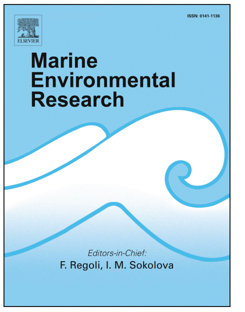
Accepted Manuscript



Application of “taxocene surrogation” and “taxonomic sufficiency” concepts to fish farming environmental monitoring. Comparison of BOPA index versus polychaete assemblage structure

F. Aguado-Giménez, J.I. Gairín, E. Martinez-Garcia, V. Fernandez-Gonzalez, M. Ballester Moltó, J. Cerezo-Valverde, P. Sanchez-Jerez

PII: S0141-1136(14)00184-6

DOI: 10.1016/j.marenvres.2014.10.006

Reference: MERE 3946

To appear in: *Marine Environmental Research*

Received Date: 3 July 2014

Revised Date: 27 October 2014

Accepted Date: 31 October 2014

Application of "taxocene surrogation" and "taxonomic sufficiency" concepts to fish farming environmental monitoring. Comparison of BOPA index versus polychaete assemblage structure

Aguado-Giménez, F.[1]*, Gairín, J.I.[2], Martinez-Garcia, E.[3], Fernandez-Gonzalez, V.[3], Ballester Moltó, M.[1], Cerezo-Valverde, J.[1], Sanchez-Jerez, P.[3]

[1]. Instituto Murciano de Investigación y Desarrollo Agrario y Alimentario (IMIDA). Estación de Acuicultura Marina. Puerto de San Pedro del Pinatar 30740. Murcia, Spain.

[2]. IRTA Investigación y Tecnología Agroalimentaria. Ctra. De Poblenou Km 5.5, 43450 Sant Carlos de la Rápita. Tarragona, Spain.

[3]. Departamento de Ciencias del Mar y Biología Aplicada. Universidad de Alicante. Ap. 99, 03080 Alicante, Spain.

* Corresponding author. Tel.: +34968184518; Fax: +34968181116; E-mail: felipe.aguado@carm.es

Keywords: benthic index; fish farming; monitoring; polychaete assemblage; taxocene surrogation; taxonomic sufficiency

Abstract

"Taxocene surrogation" and "taxonomic sufficiency" concepts were applied to the monitoring of soft bottoms macrobenthic assemblages influenced by fish farming following two approaches. Polychaete assemblage evaluation through multivariate analysis and the benthic index BOPA were compared. Six fish farms along the Spanish Mediterranean coast were monitored. Polychaete assemblage provided a suitable picture of the impact gradient, being correlated with total free sulphides. BOPA did not support the impact gradient described by the polychaete assemblage, providing erroneous categorizations. The inclusion of several polychaete families, which were locally identified as indicative of affection to recalculate BOPA, resulted in an improved diagnosis and correlation with the impact gradient. Nevertheless, frequent misclassifications occurred. These results suggest that the structure of polychaete families, sulphides and granulometry conform an appropriate strategy for fish farming monitoring. Biotic indices need to be specifically designed for concrete activities, and regionally validated, because of the environmental plasticity of benthic invertebrates.

Introduction

Marine soft-bottom macrozoobenthic communities have the inherent ability of integrating the environmental quality status reflecting the system condition adequately. They are relatively immobile residents and exhibit a wide range of tolerance or sensitivity to different stressors (Tataranni and Lardicci, 2010). Also, soft-bottom macrobenthic communities play a key role in the provision of ecosystem services, mainly the cycling of nutrients and material in the sediments, and the maintenance of the benthic food web (Gray and Elliott, 2009). Owing to both prerogatives, macrobenthic invertebrate communities have been widely used as an indicator for environmental assessments, particularly for fish farming monitoring (Karakassis et al., 2000; Carroll et al., 2003; Lee et al., 2006; Aguado-Giménez et al., 2007).

Information from scientific studies on the marine benthic communities applicable to the management of coastal resources is not always easily understandable for a non specialist audience. To facilitate the management and decision-making processes, marine benthic scientists have developed ecological indicators as benthic biotic indices (BBIs hereafter), which supply synoptic information of the ecosystems (Salas et al., 2006). BBIs attempt to simplify the complex multivariate structure extracted from benthic assemblages up to a single (univariate) value that summarizes the ecological status as a function of some ecological characteristic (e.g. sensitivity or tolerance to pollution, trophic strategy, combined with species' richness, abundance, presence – absence, diversity, etc.) (Pinto et al., 2009; Dauvin et al., 2010).

Over the last decades, coinciding with the publication of the European Water Framework Directive (WFD, 2000/60/EC) and the Marine Strategy Framework Directive (MSFD, 2008/56/EC), there was an intensive work for the development of monitoring tools (Dauvin, 2007). In the case of the marine benthic environment, it has led to a revival and emergence of old and new BBIs (Díaz et al., 2004; Devlin et al., 2007; Pinto et al., 2009), with the aim of standardising methodologies for typifying and monitoring the environment quality of European water bodies. Many of the recently emerged or created BBIs (e.g. AZTI´s Marine Biotic Index, AMBI: Borja et al., 2000; BENTIX: Simboura and Zenetos, 2002; Benthic Quality Index, BQI: Rosenberg et al., 2004; Benthic Opportunistic Polychaetes Amphipods Index, BOPA: Dauvin and Ruellet, 2007; MEDiterranean OCCidental Index, MEDOCC: Pinedo and Jordana, 2007; and others) have already been applied to assess the ecological quality status of water bodies at different locations worldwide (e.g. Pranovi et al., 2007: lagoon of Venice, Italy; Afli et al., 2008: Tunisian coasts and lagoons; Labrune et al., 2012: Rhône river, France; Pinedo et al., 2012: Spanish Mediterranean coast; Quiroga et al., 2013: Patagonian fjords, Chile). Most of these indices are based on the concept of macrobenthic sensitivity or tolerance *sensu*, the Pearson and Rosenberg (1978) organic enrichment paradigm. As there are different sources of organic enrichment and, furthermore, this is not the only source of pollution affecting benthic communities, most of BBIs have also been tested under particular sources of stress, such as domestic sewage (de la Ossa Carretero et al., 2009; Sampaio et al., 2011), mining (Marín-Guirao et al., 2005; Gray and Delaney, 2008), dredging, industrial and agricultural wastes (Borja et al., 2003), metalliferous wastes (Simboura et al., 2007), and aquaculture (Aguado-Giménez et al., 2007; Bouchet and Sauriau, 2008; Borja et al., 2009; Nickell et al., 2009; Forchino et al., 2011; Keeley et al., 2012; Karakassis et al., 2013).

However, after several decades of widespread use of these BBIs, their application is still questioned, mainly as regards the sources of stress and geographical plasticity (Green and Chapman, 2011; Keeley et al., 2012). Beside these overall discrepancies, there are many other controversial aspects, such as the assignation of taxa to sensitivity/tolerance levels (Carvalho et al., 2006; Labrune et al., 2012), misclassification of the ecological quality status (Quintino et al., 2006; Bouchet and Sauriau, 2008; Callier et al., 2008), differences in discriminating power among indices (Pranovi et al., 2007), loss of essential information causing loss of diagnostic capability (Sampaio et al., 2011), difficulty in distinguishing natural from human-induced stress in transitional waters (Dauvin, 2007; Elliott and Quintino, 2007), the need of assessing the spatial and temporal variability of the BBI performance (Kröncke and Reiss, 2010; Tataranni and Lardizzi, 2010; Quintino et al., 2012), the availability of taxonomic expertise (the so-called "taxonomic impediment; "Wheeler, 2004; Bevilacqua et al., 2013), and several more as Dauvin et al. (2012) summarized. Finally, the main criticisms of the indices are the huge loss of information by reducing the complexity of a community to a single value, and the misleading biological interpretation of the data they are intending to summarize (Green and Chapman, 2011). Despite this, some of the above-mentioned or other BBIs have been postulated as reliable tools not only in the context of the WFD, but also for the mandatory monitoring of specific operational aquaculture activities like mussel and fish farming as well (Borja et al., 2009; Forchino et al., 2011; Keeley et al., 2012; Karakassis et al., 2013).

Nevertheless, all of the very few comparative studies contrasting the univariate information provided by some BBIs *versus* the multivariate information from the whole assemblage data set, in the context of aquaculture, agree that the multivariate approach is more appropriate to detect the influence of aquaculture on the benthic environment (Aguado-Giménez et al., 2007; Callier et al., 2008; Quintino et al., 2012). Considering that mandatory survey is an additional economic charge for fish farmers, it would be desirable that these studies were very well balanced from a cost/benefit point of view, without forgetting representativeness and robustness. Therefore, a selection of informative but cheap impact indicators (Riera et al., 2012) and the establishment of an adequate sampling design (Fernandes et al., 2001; Aguado-Giménez et al., 2012a; Fernandez-Gonzalez et al, 2013) are needed. The application of some BBIs such as AMBI, BENTIX, BQI, MEDOCC or others, is an expensive and very time-consuming task requiring practised taxonomists for identifying all the fauna to species level (Dauvin et al., 2003; De Biasi et al., 2003; Riera et al., 2012). Nevertheless, others BBIs such as BOPA only works with two faunal groups (polychaetes and amphipods), so its taxonomic effort is much lower. On the other hand, some authors have even proposed that surrogating the whole benthic assemblage to a particular taxonomic group which was able to reflect the natural or human-induced development of the entire assemblage, would represent a significant cost reduction with a minimal loss of relevant information (Olsgard and Sommerfield, 2000; Bertasi et al., 2009; Soares-Gomes et al., 2012). This is the case of polychaetes, whose frequency and abundance in soft bottom and its proven sensitivity to environmental changes makes them an appropriate surrogate taxocene for monitoring programmes (Olsgard et al. 2003; Giangrande et al., 2005; Del-Pilar-Ruso et al., 2009; Musco et al., 2009; Soares-Gomes et al., 2012). Likewise, following the concept of taxonomic sufficiency (Ellis, 1985), it has been evidenced that identifying polychaetes to family level - as unique biological indicator - provides sufficiently accurate assessments in monitoring surveys of aquaculture activities (Tomassetti and Porrello, 2005; Lee et al., 2006; Aguado-Giménez et al., 2012b; Martínez-García et al., 2013).

The aim of this work is to evaluate some indicators that meet some criteria of simplicity (as indicated by the concepts of “taxonomic sufficiency” and “taxocene surrogation” proposed by Ellis (1985) and *sensu* Olsgard and Sommerfield (2000), respectively) as potential tools for the monitoring of fish farming effects on soft bottoms. For that, we assess two alternative approaches under an *a priori* well-defined impact gradient: univariate BOPA index, which uses only polychaetes and amphipods identified to family level, and the multivariate structure of polychaete assemblage also identified to family level, both together with a good sediment descriptor (granulometry) and a very sensitive chemical variable (total free sulphides), as Aguado-Giménez et al (2012a) suggested. We considered that both approaches provide a good balance between simplicity and robustness. Both methods were compared in terms of discriminating capacity and susceptibility of application in compulsory surveys. In order to ascertain whether specific polychaete families which act as indicator of the impact derived from fish farming would improve BBI results, we also recalculate BOPA using those tolerant polychaete families derived from the results of this study and the definitions from Martínez-García et al. (2013).

Materials and Methods

Study area and proceedings for sampling and analyses

The study was carried out at six gilthead seabream (*Sparus aurata*) and European seabass (*Dicentrarchus labrax*) cage fish farms randomly chosen along the Spanish Mediterranean coast (Figure 1). At each farm, three zones at increasing distances from the farm were established following a theoretical enrichment gradient downstream: beneath the cages, just outside the farm lease boundaries (60 – 100m away from the cages), and a reference zone (0.5 – 1km away from the lease boundary). This zoning was set in agreement with the proposal of “allowable zone of effects” suggested by Aguado-Giménez et al. (2012a). At each distance, three sites were randomly sampled, where three sediment replicates were collected for polychaete and amphipod assemblage analyses. Three additional samples were also taken for sediment physico-chemical analyses. All sediment samples were collected using a 0.04 m^2 Van-Veen grab. Two sampling campaigns were conducted, at late-summer 2009 and mid-winter 2010.

Samples for the study of polychaete and amphipod assemblages were sieved (1mm) on board and fixed in formalin 4%. Polychaete and amphipod specimens were sorted under a magnifying glass using forceps, and preserved in ethanol 70% until identification. All the specimens were identified to family level. Using these data, we calculated the BOPA index (Dauvin and Ruellet, 2007) for each replicate as follows:

$$BOPA = \log\left(\frac{fp}{fa+1}+1\right),$$

where fp is opportunistic polychaete frequency, and fa is amphipod frequency (excluding Genus *Jassa*). The BOPA index was calculated using the list of opportunistic polychaetes families given in Gómez Gesteira and Dauvin (2000). We also recalculated BOPA (as “BOPA-fish farming”: hereafter BOPA-FF) including not only some of the typical polychaete families which are worldwide considered as indicator of organic enrichment (*Capitellidae* and *Spionidae*, also included in BOPA), but also those families which showed a higher contribution to dissimilarities along the gradient in the present work (see further on), and also those families tolerant to the fish farming influence proposed by Martínez-García et al (2013). We used the same ecological classification for both BOPA and BOPA-FF (Table 1).

Subsamples were extracted from each sediment sample for physical and chemical analyses. We selected the finest fraction of the sediment (FFS: < 63µ) as benthic environment descriptor (Buchanan, 1984), and total free sulphides concentration (TFS; Wildish et al., 1999) as chemical indicator of environmental impact which would serve us to confirm the impact gradient , in agreement with the proposal of Aguado-Giménez et al. (2012a).

Statistical procedures

All of the multivariate data were analysed using PRIMER-E software (PRIMER, 2006) with the add-on package PERMANOVA+ (Anderson et al., 2008).

Polychaete abundance, environmental variables, BOPA and BOPA-FF data were analysed separately by means of a 4-factor model using permutational multivariate analysis of variance (PERMANOVA) based on the Bray-Curtis dissimilarities of untransformed data (Anderson, 2001). The analyses were tested using 4999 permutations of residuals under a reduced model. We considered different spatial scales and a short term temporal scale (factor “time”; T: random and orthogonal with two levels: summer and winter) around the main factor “distance” (D: fixed and orthogonal with three levels: BC: below cages, LB: just outside the lease boundaries, and RF: reference). Spatial scales were represented by the factors “farm” (F: random and orthogonal) with six levels (six randomly selected farms along the Spanish Mediterranean coast as a representative sample of all the possible fish farms), and “site” (S: random and nested in F, T and D, with three levels: three sites within each distance and sampling time as a random replication of the main factor D). The number of replicates for each combination of factors was n = 3. Our interesting null-hypothesis was that there were not significant differences between distances for the whole set of farms sampled at different times for the selected biotic and abiotic variables, i.e. the contrasting term is D. In the case that any random factor or their interactions resulted significant, the fixed main effect D was, regardless, interpreted accordingly with Quinn and Keought (2002). Random effects were not relevant for the objectives of the work, in accordance with Underwood (1997), neither their interactions since they only represent the expected spatio-temporal variability around the null-hypothesis, and therefore they were not interpreted.

We used Spearman rank correlation test to investigate the relationship between the multivariate polychaete assemblage structure and environmental variables (FFS and TFS), following the BIOENV routine (Clarke and Gorley, 2006).

SIMPER test (Clarke, 1993) was performed to enquire on those polychaete families with a greater contribution to the differences among the levels of the fixed factors PI and D. The families with a greater contribution were used to calculate BOPA-FF. Comparisons between polychaete multivariate structure versus BOPA and BOPA-FF were conducted following the data processing proposed by Warwick et al. (2010): the values of the PC1 score from a Principal Component Analysis of the polychaete abundance data set can be interpreted as a disturbance index to be compared with BOPA and BOPA-FF values. PC1 values are plotted against BOPA and BOPA-FF values and the Spearman rank correlation test was run.

Results

Polychaete assemblage

For the whole farms and sampling times, a total of 48 polychaete families were identified, and a total 37233 specimens were recorded. Total abundance decreased from farm to reference locations, but families' richness was very similar through the gradient (Table 2). PERMANOVA (Table 3) showed significant differences for the fixed factor "distance" (D: $P < 0.05$). Pairwise PERMANOVA for the levels of the factor D showed significant differences between BC and RF ($t = 1.8234$; $P(\text{perm}) = 0.0260$) and between BC and LB ($t = 1.5415$; $P(\text{perm}) = 0.0380$), but not between LB and RF ($t = 0.9167$; $P(\text{perm}) = 0.6660$).

Average abundances of the polychaete families with highest contribution to dissimilarities between samples after SIMPER test are shown in Table 2. The families that most contributed to the dissimilarities between the samples from BC and LB were *Dorvilleidae*, *Nereididae*, *Oweniidae* and *Capitellidae* (cumulative contribution: 46.45%), the three former being more abundant in BC and the latter in LB. Dissimilarities among the samples obtained in BC and RF locations were mainly caused by *Dorvilleidae*, *Nereididae*, *Oweniidae Capitellidae*, and *Paraonidae* (cumulative contribution: 55.18%). The first three families were more abundant in BC, *Capitellidae* was only slightly plentiful in BC and the latter was more abundant in RF. *Capitellidae* and *Paraonidae* were the families which showed a relevant contribution to the dissimilarities between RF and LB samples (cumulative contribution: 27.49%). In addition to these, some other less abundant families all contributed even further to the dissimilarities among distances. *Capitellidae* was more abundant on average at LB distance than at BC. In RF, despite its average abundance was lower than in BC and LB, *Capitellidae* remains significantly high. A similar situation was observed for *Cirratulidae*, but with lower average abundances. *Dorvilleidae* was much more abundant on average in BC samples than in LB and RF. The same applies to *Nereididae*, but with lower average abundances too. *Paraonidae* average abundance was very similar in BC and RF samples, while it was about half at LB. Such a situation also occurred for *Spionidae* and *Lumbrineridae*, but with lower average abundances. *Oweniidae* was more abundant in BC samples than at LB and RF.

Environmental variables

On average, FFS was very similar through the impact gradient (Figure 2A) for the set of farms, and PERMANOVA did not reveal significant differences between D levels (Table 3). Despite this, an increase of the FFS occurred in BC and LB from summer to winter (Figure 2A). A gradient of affection was accurately described by TFS (Figure 2B). PERMANOVA revealed significant differences for the levels of the factor D for TFS (Table 3), between BC and RF (pairwise PERMANOVA test,t = 3.0529; *P*(perm) = 0.0120), between BC and LB (t = 2.1657; *P*(perm) = 0.0460), and between LB and RF (t = 2.2186; *P*(perm) = 0.0270),

BOPA and BOPA-FF

PERMANOVA for BOPA data (Table 3) did not indicate significant differences among distances ($P > 0.1$) However, BOPA-FF showed significant differences among distances ($P < 0.05$) (Table 3), which only occurred between BC and RF (pairwise PERMANOVA test, t = 3.2924; $P = 0.0070$). Using the ecological quality classification from Table 1, BOPA gives an EcoQ of "good" to all the distances, whereas BOPA-FF gives a status of "moderate" to BC and "good" to LB and RF (Figures 2C and D), unlike sampling time.

Correlation among variables

Polychaete assemblage structure correlated significantly with TFS (ρ = 0.262; $P < 0.05$), but not with FFS (ρ= 0.105; $P > 0.05$). Correlation between the impact gradient described by polychaete assemblage structure (PC1 axis: Figure 3) and BBI´s was negative for BOPA (ρ = -0.62; $P > 0.05$) which means that the trend was to decrease BOPA values as the impact gradient progresses. On the other hand, correlation between BOPA-FF and the impact axis was positive (ρ = 0.38; $P > 0.05$), meaning that BOPA-FF describes the impact gradient in a similar way as the polychaete assemblage structure did. Nevertheless, both BBI´s provided very high values (poor to bad status) for many samples from the lower part of the impact gradient (Figure 3).

Discussion

Surrogating the whole soft-bottom benthic community to the abundance of polychaete families has provided a very accurate picture of the impact gradient around offshore western Mediterranean fish farms. TFS also showed a clear gradient of affection as moving away from the farms. In contrast, BOPA index did not support the impact gradient described by the polychaeta assemblage, and provide a generalized misclassification. However, when polychaeta families tolerant to fish farming influence (*Dorvilleidae*, *Nereididae*, *Oweniidae* and *Glyceridae* (Table 2), the latter as Martínez-García et al. (2013) suggested) were considered to recalculate BOPA (as BOPA-FF), the predictive capability and correlation with the impact gradient improved. Nevertheless, many erroneous categorisations were obtained with both BBIs particularly in the lower part of the impact gradient. This may be a consequence of the low abundance of amphipods over the whole study area, and also to the abundance of classical tolerant polychaete families – mainly *Capitellidae*, *Cirratulidae* and *Spionidae* - in reference locations.

Average FFS for the whole farms was very similar at all the distances from the farms, despite slight variations between sampling times. Therefore, the affection gradient described by TFS -accordingly with the thresholds proposed by Carballeira et al. (2011)- and polychaete assemblage is not attributable to the physical characteristics of the sediment, but to the farms´ influence. The physical characteristics of the sediment largely determine the faunal composition (Labrune et al., 2007; Mutlu et al., 2010; Martínez-García et al., 2013) and, accordingly, the expected response can be different (Fernandez-Gonzalez et al., 2013). Therefore, sediment grain size - specially the finest fraction - can act as a good environmental descriptor, being also very useful for the interpretation of other variables.

The response of soft-bottom macrobenthic communities to the influence of aquaculture activities can be very variable and, therefore, monitoring requires robust indicators. Despite this, some authors consider that it is predictable to anticipate how will be the macrobenthic behaviour (Borja et al., 2009). Oceanographic conditions and husbandry practices are strongly correlated with this variability (Borja et al., 2009). Both culture and oceanographic conditions will ultimately determine the gross waste output, the degree of organic enrichment, the waste’s dispersion and the extent of the affected area (Tomassetti et al., 2009), which obviously have an influence on the environment response. Nevertheless, the response itself depends not only on these external factors, but also and mainly on the physic, chemical and biologic characteristics of the receiving benthic environment: type of sediment – which, in turn, determines the community composition (Martínez-García et al., 2013) - and its functional status at a regional scale. Miron et al. (2005) suggested that this wide range of response also largely depends on what individual indicator was used. The response of the benthic environment to the organic contribution derived from fish farming has been usually explained by the greatly accepted reference model of Pearson and Rosenberg (1978). This model postulates a progressive loss of diversity, a decline in species’ richness and an increase in abundance of opportunistic species which dominate the community, over a gradient of affection. The quintessential polychaete family considered as indicator of these changes is *Capitellidae*, which usually predominates in the sediments closer to the farms (Karakassis and Hatziyanni, 2000; Karakassis et al., 2000; Lee et al., 2006). In our work, polychaete family richness below cages was high, and very similar to that in intermediate and reference locations. We also observed that the abundance of *Capitellidae* was even larger in reference locations than below cages. Similarly, other families such as *Spionidae* and *Cirratulidae*, which are also outlined as indicative of organically enriched sediments (Méndez, 2002; Tomassetti and Porrello, 2005) also showed larger frequencies of presence at intermediate and reference locations. These results confirm the assertion that the ubiquitous usage of indicator taxa does not always work (Bustos-Baez and Frid, 2003). Martínez-García et al. (2013), as a result of a meta-analysis study, evidenced that polychaete families related with fish farming impacted areas could proliferate in non-polluted areas too when these areas are naturally enriched with organic matter. Despite these apparently controversial results, polychaete composition analyses revealed significant differences among impacted, intermediate and reference locations. This corroborates the above-mentioned variability of the benthic response, indicating that this response does not need to be generally associated to the dynamic of classical indicator taxa. On the other hand, polychaete families such as *Dorvilleidae*, *Nereididae* and *Oweniidae*, which are not considered as classical indicators but have also been consistently found in organically polluted areas (Méndez et al., 1998; Lee et al., 2006), played the role of the typical indicator families in relation with the expected assemblage distribution pattern for the six farms studied. Probably only under severe impact conditions, the typical indicator families assert their prominence. Hence, the previous definition of sensitive taxa and their weighing will be crucial for the development and application of BBIs.

Long-term temporal changes expected in soft-bottom polychaete assemblages influenced by fish farming include a reduction of diversity as a consequence of sediment chemical deterioration (Lee et al., 2006). However, this response was not so evident at shorter temporal scales, such as in our study, despite indicative species held a higher relative abundance. Consequently, longer-term monitoring of robust indicators at an appropriate spatial scale is needed to characterize the impacts (press or pulses) with certainty, in the sense of Underwood (1991). What really reveals whether a polychaete assemblage is or is not affected is not only the development of “universal” indicator species, but the spatio-temporal dynamic of the whole assemblage, always contrasting with natural variability.

BOPA index is based on the Pearson and Rosenberg (1978) paradigm and also on those opportunistic polychaete families considered as “universal” indicators of organic pollution (Gómez-Gesteira and Dauvin, 2000; Dauvin and Ruellet, 2007). When we applied BOPA to our six fish farms data set, all distances obtained the same average ecoQ: “good”. Neither BOPA quality classification nor data set (as a quantitative variable) were able to distinguish between impacted, intermediate and reference locations as polychaete multivariate analysis did. Also, its relationship with the impact gradient described by polychaete families’ structure was upside down. This disagreement was observed both in the lower and the upper levels of the impact gradient. BOPA values close to zero are indicative of undisturbed environments. These values can be obtained when the frequency of amphipods is quite larger than the frequency of opportunistic polychaetes, but also when opportunistic polychaetes are absent, as would occur in an azoic situation or whether other families different from those considered by BOPA dominate. In this case, as occurred in some situations in our study, the assigned health condition was absolutely erroneous. On the other hand, high BOPA values – indicative of disturbed environments - are obtained when the frequency of amphipods is low or whether frequency of tolerant polychaetes is high. In our case, intermediate and reference locations were poor in amphipods with some exception and abundant in some opportunistic polychaete families included in BOPA calculation, such as *Capitellidae* and *Cirratulidae*. Also, the total number of polychaetes plus amphipods is very high, which decreases the frequency of amphipods. Then, BOPA values were higher than expected. Keeley et al. (2012) obtained similar misclassifications using BOPA, and suggested that indices which are based on a limited number of taxa – as occurs with BOPA - are unlikely to be suitable for broad geographical comparisons.

When we changed the polychaete families suggested by Gómez-Gesteira and Dauvin (2000) to our more indicative families and some of those proposed by Martínez-García et al. (2013) to calculate BOPA-FF (Table 2), the obtained average ecoQs were more in accordance with the impact gradient described by TFS and polychaete assemblage structure: BC as “moderate”, LB and RF as “good”. Furthermore, BOPA-FF data set was able to discriminate between distances significantly, being the data correlated with the impact gradient defined by the polychaete assemblage structure. The consideration of polychaete families which had been previously confirmed as local (or regional), indicative of affection of a particular stressor, has meant a significant improvement in the diagnosis. However, we observed the same type of erroneous classification as with BOPA. We suggest, as well as Keeley et al. (2012), that benthic indices need to be regionally validated in order to avoid the risk of under- or overestimating the ecological status. These results reveal that any benthic index that aims to be used to evaluate a particular activity needs to be specifically designed. Then, local or regional indicative taxa should be defined previously by pilot studies and/or bibliography revisions for a more accurate application of BOPA-FF or similar indices. Also, an appropriate sampling design, including several reference locations (Tataranni and Lardicci, 2010) and a proper statistical data management, should be used for diagnosis rather than the establishment of thresholds. Despite the improvements achieved with BOPA-FF, we really believe that using this index can be unwise without accompanying it with any other statistic approach that ensure no loss of information (Green and Chapman, 2011). A misclassification might imply wrong management measures too, and injuries to the environment or the producers.

Nevertheless, several authors concur that the “picture” of areas influenced by fish farming provided by the multivariate analysis of assemblages composition was more suitable and statistically validated than that provided by univariate benthic indices (Aguado-Giménez et al., 2007; Callier et al., 2008; Quintino et al., 2012), regardless of taxonomic resolution. Quintino et al. (2012) also showed that multivariate abundance data, together with primary biological variables (richness and total abundance) were more effective than synthetic biotic indices to diagnose benthic alterations related with oyster faming. Likewise, assemblage composition analyses let us to enquire in several aspects of the community which benthic indices obviate, such as contribution of species, differences in the composition depending on environmental characteristics, identifying local or regional indicative taxa and seasonal changes. Callier et al. (2008) proposed that in any case benthic indices could constitute a complementary analysis to facilitate interpretation of the results, but multivariate statistics is the more powerful method to detect changes in benthic assemblages. In the case of BBI utilisation, it would be desirable that they could be regionally adapted for specific applications to avoid erroneous verdicts that could result in wrong management measures.

On the basis of all above-mentioned, we propose the utilisation of TFS as a reliable indicator of impact derived of fish farming (cause-effect relationship), and FFS as benthic descriptors. Both variables should complement the multivariate analysis of polychaete assemblages as a biological indicator in the compulsory monitoring programmes of cage fish farming. At least, this strategy worked well in western Mediterranean farms. These three variables allow us to perform an appropriate interpretation and diagnosis for any type of sediment. A monitoring programme should be simple and robust. Therefore, surrogating the whole benthic assemblage to a single but sensitive taxocene, reducing taxonomic resolution up where sensibility allows, and choosing the strictly necessary but indicative physic-chemical descriptors or indicators as complementary variables should be compensated improving sampling designs, e.g. spatio-temporal nested design and the use of several controls. We consider that this strategy can be applied in any geographical area.

One of the great differences between using biotic indices and multivariate assemblage data is that the information finally provided by the indices (ecological quality status) is easier to understand and manage. The problem is that this categorisation may not be a real reflection whether the used index is not performed for a particular geographical area, habitat or type of impact. Since reaching an accurate diagnosis is as important as providing easily manageable information, we believe that it is necessary to improve monitoring programmes to ensure that complex systems are evaluated in the most reliable way, providing correct information for managers. Simultaneous utilisation of multivariate analysis of polychaete assemblage and locally adapted BBIs, such as BOPA-FF, would contribute to communicate properly the monitoring results to decision-makers. However, as Green and Chapman (2011) suggested, it is also necessary a better education for managers to deal with complex scenarios.

Acknowledgements

This research was funded by the Spanish National Plans of Aquaculture (JACUMAR). Authors wish to thank to the staff of the fish farms that gave us access and help during the study. We also thank to all the colleagues who helped us during the sampling campaigns and in the processing of samples at the lab.

Bibliography

Afli, A., Ayari, R., Zaabi, S. (2008). Ecological quality of some Tunisian coast and lagoon locations, by using benthic community parameters and biotic indices. Est. Coast Shelf Sci. 80: 269-280.

Aguado-Giménez, F., Marín, A., Montoya, S., Marín-Guirao, L., Piedecausa, M.A., García-García, B. (2007). Comparison between some procedures for monitoring offshore cage culture in western Mediterranean Sea: Sampling methods and impact indicators in soft substrata. Aquaculture 271: 357-370.

Aguado-Giménez, F., Carballeira, A., Collado, C., González, N., Sánchez-Jerez, P. (2012a). Methodological proposal for the development of environmental monitoring plans for marine farming in floating cages (In Spanish). Ministry of Agriculture Food and Environment of Spain. Available in: http://www.magrama.gob.es/app/jacumar/recursos_informacion/Documentos/Noticias/57_Planes_Vigilancia_Ambiental_JACUMAR_v.0.pdf (last access: March 2013).

Aguado-Giménez, F., Piedecausa, M.A., Gutiérrez, J.M., García-Charton, J.A., Belmonte, A., García-García, B. (2012b). Benthic recovery after fish farming cessation: a ‘‘beyond-BACI’’ approach. Mar. Poll. Bull. 64: 729-738.

Anderson, M.J. (2001). A new method for non-parametric multivariate analysis of variance. Austral. Ecol. 26: 32-46.

Anderson, M.J., Gorley, R.N., Clarke, K.R. (2008). PERMANOVA+ for PRIMER: guide to software and statistical methods. PRIMER-E, Plymouth, UK.

Bertasi, F., Colangelo, M.A., Colosio, F., Gregorio, G., Abbiati, M., Ceccherelli, V.U. (2009). Comparing efficacy of different taxonomic resolutions and surrogates in detecting changes in soft-bottom assemblages due to coastal defence structures. Mar. Poll. Bull. 58: 686-694.

Bevilacqua, S., Claudet, J., Terlizzi, A. (2013). Best practicable aggregation of species: a step forward for species surrogacy in environmental assessment and monitoring. Ecol. Evol. DOI: 10.1002/ece3.715.

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. Poll. 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. Poll. Bull. 46: 835–845.

Borja, A., Rodríguez, J.G., Black, K., Bodoy, A., Emblow, C., Fernandes, T.F., Forte, J., Karakassis, I., Muxika, I., Nickell, T.D., Papageorgiou, N., Pranovi, F., Sevastou, A., Tomassetti, P., Angel, D. (2009). Assessing the suitability of a range of benthic indices in the evaluation of environmental impact of fin and shellfish aquaculture located in sites across Europe. Aquaculture 293: 231-240.

Bouchet, V.M.P., Sauriau, P-G. (2008). Influence of oyster culture practices and environmental conditions on the ecological status of intertidal mudflats in the Petrus Charentais (SW France): a multi-index approach. Mar. Poll. Bull. 56: 1898-1912.

Buchanan, J.B. (1984). Sediment analysis. In: Holme, N.A., and McIntyre, A.D. (Eds.). Methods for the study of marine benthos. Blackwell Scientific Publications. Oxford, p 41-64.

Bustos-Baez, S., Frid, C. (2003). Using indicator species to assess the state of macrobenthic communities. Hydrobiologia 496: 299-309.

Callier, M.D., McKindsey, C.W., Desrosiers, G. (2008). Evaluation of indicators used to detect mussel farm influence on the benthos: two case studies in the Magdalen Islands, Eastern Canada. Aquaculture 278: 77-88.

Carballeira, A., Aguado-Giménez, F., González, N., Sánchez-Jerez, P., Texeira, J.M., Gairin, J.I., García-García, B., Fernández-González, V., Carreras, J., Macías, J.C., Acosta, D., Collado, C. (2011). Utilization of ecological profiles for the selection of sediment geochemical variables as indicators of the environmental impact generated by offshore mariculture. In: FOESA, M. Villarroel, L. Reig and C. Almansa (Eds.). Proceedings of the XIII Spanish National Symposium of Aquaculture: "In Balance with the Environment" (ISBN: 978-84-937611-9-6). pp. 41-42. Available at: http://www.congresodeacuicultura.es/uploads/a_9467_actas_xiii_cna.pdf (In Spanish).

Carroll, M.L., Cochrane, S., Fieler, R., Velvin, R., White, P. (2003). Organic enrichment of sediments from salmon farming in Norway: environmental factors, management practices and monitoring techniques. Aquaculture 226: 165-180.

Carvalho, S., Barata, M., Pereira, F., Gaspar, M.B., Cancela da Fonseca, L., Pousao-Ferreira, P. (2006). Distribution patterns of macrobenthic species in relation to organic enrichment within aquaculture earthen ponds. Mar. Poll. Bull. 52: 1573-1584.

Clarke, K.R. (1993). Non-parametric multivariate analyses of changes in community structure. Aust. J. Ecol. 18, 117–143.

Clarke, K.R., Gorley, R.N. (2006). PRIMER v6: User Manual/Tutorial. PRIMER-E, Plymouth, UK.

Dauvin, J.C. (2007). Paradox of estuarine quality: benthic indicators and indices, consensus or debate for the future. Mar. Poll. Bull. 55: 271-281.

Dauvin, J.C., Ruellet, T. (2007). Polychaete / amphipod ratio revisited. Mar. Poll. Bull. 55: 215-224.

Dauvin, J.C., Gómez-Gesteira, J.L., Salvande Fraga, M. (2003). Taxonomic sufficiency: an overview of its use in the monitoring of sublittoral benthic communities after oil spills. Mar. Poll. Bull. 46: 552-555.

Dauvin, J.C., Bellan, G., Bellan-Santini, D. (2010). Benthic indicators: from subjectivity to objectivity – where is the line? Mar. Poll. Bull. 50: 947-953.

Dauvin, J.C., Alizier, S., Rolet, C., Bakalem, A., Bellan, G., Gomez Gesterira, J.L., Grimes, S., de-la-Ossa-Carretero, J.A., Del-Pilar-Ruso, Y. (2012). Response of different benthic indices to diverse human pressures. Ecol. Indicat. 12: 143-153.

De Biasi, A.M., Bianchi, C.N., Morri, C. (2003). Analysis of macrobenthic communities at different taxonomic levels: an example from an estuarine environment in the Ligurian Sea NW Mediterranean. Est. Coast Shelf Sci. 58: 99-106.

de-la-Ossa-Carretero, J.A., Del-Pilar-Ruso, Y., Giménez-Casalduero, F., Sánchez-Lizaso, J.L. (2009). Testing BOPA index in sewage affected soft-bottoms communities in the north-western Mediterranean. Mar. Poll. Bull. 58: 332-340.

Del-Pilar-Ruso, Y., de-la Ossa-Carretero, J.A., Angel, L.F., Ferrero-Vicent, L.M., Giménez-Casalduero, F., Sánchez-Lizaso, J.L. (2009). Assessment of soft-bottom polychaete assemblage affected by a spatial confluence of impacts: sewage and brine discharges. Mar. Poll. Bull. 58: 795-786.

Devlin, M., Best, M., Haynes, D. (2007). Implementation of the Water Framework Directive in European marine waters. Mar. Poll. Bull. 55: 1-2.

Díaz, R.J., Solan, M., Valente, R.M. (2004). A review of approaches for classifying benthic habitats and evaluating habitat quality. J. Environ. Manage. 73: 165-181.

Elliott, M., Quintino, V. (2007). The estuarine quality paradox environmental homeostasis and the difficulty of detecting anthropogenic stress in naturally stressed areas. Mar. Poll. Bull. 54: 640-645.

Ellis, D. (1985). Taxonomic sufficiency in pollution assessment. Mar. Poll. Bull. 16: 549.

Fernandes, T.F., Eleftheriou, A., Ackefors, H., Eleftheriou, M., Ervik, A., Sánchez-Mata, A., Scanlon, T., White, P., Cochrane, S., Pearson, T.H., Read, P.A. (2001). The scientific principles underlying the monitoring of the environmental impacts of aquaculture. J. App. Ichthyol. 17: 181-193.

Fernández-González, V., Aguado-Giménez, F., Gairin, I., Sánchez-Jerez, P. (2013). Exploring patterns of variation in amphipod assemblages at multiple spatial scales: natural variability versus coastal aquaculture effects. Aquacult. Environ. Interact. 3: 93-105.

Forchino, A., Borja, A., Brambilla, F., Rodríguez, J.G., Muxika, I., Terova, G., Saroglia, M. (2011). Evaluating the influence of off-shore cage aquaculture on the benthic ecosystem in Alghero Bay (Sardinia, Italy) using AMBI and M-AMBI. Ecol. Indicat. 11: 1112-1122.

Giangrande, A., Licciano, M., Musco, L. (2005). Polychaetes as environmental indictors revisited. Mar. Poll. Bull. 50: 1153-1162.

Gómez Gesteira, J.L., Dauvin, J.C. (2000). Amphipods are good bioindicators of the impact of oil spill on soft-bottom macrobenthic communities. Mar. Poll. Bull. 40(11): 1017-1027.

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.

Gray, J.S., Elliott, M. (2009). Ecology of marine sediments. From science to management. Oxford University Press (2nd Ed.). New York. 225 pp.

Gray, N.F., Delaney, E. (2008). Comparison of benthic macroinvertebrate indices for the assessment of the impact of acid mine drainage on an Irish river below an abandoned Cu-S mine. Environ. Pollut. 155: 31-40.

Green, R., Chapman, P.M. (2011). The problem with indices. Mar. Poll. Bull. 62: 1377-1380.

Karakassis, I., Hatziyanni, E. (2000). Benthic disturbance due to fish farming analyzed under different levels of taxonomic resolution. Mar. Ecol. Prog. Ser. 203: 247-253.

Karakassis, I., Tsapakis, M., Hatziyanni, E., Papadopoulou, K.N., Plaiti, W. (2000). Impact of cage fish farming on the seabed in three Mediterranean coastal areas. ICES J. Mar. Sci. 57: 1462-1471.

Karakassis, I., Dimitriou, P.D., Papageorgiou, N., Apostolaki, E.T., Lampadariou, N., Black, K.D. (2013). Methodological considerations on the coastal and transitional benthic indicators for the Water Framework Directive. Ecol. Indicat. 29: 26-33.

Keeley, N.B., Forrest, B.M., Crawford, C., Macleod, C.K. (2012). Exploiting salmon farm benthic enrichment gradients to evaluate the regional performance of biotic indices and environmental indicators. Ecol. Indicat. 23: 453-466.

Kröncke, I., Reiss, H. (2010). Influence of macrofauna long-term natural variability on benthic indices used in ecological quality assessment. Mar. Poll. Bull. 60: 58-68.

Labrune, C., Grèmare, A., Amouroux, J.M., Sardá, R., Gil, J., Taboada, S. (2007). Assessment of soft-bottom polychaetes assemblages in the Gulf of Lion (NW Mediterranean) based on a mesoscale survey. Est. Coast. Shelf Sci. 71: 133-147.

Labrune, C., Romero-Ramírez, A., Amouroux, J.M., Duchêne, J.C., Desmalades, M., Escoubeyrou, K., Buscail, R., Grémare, A. (2012). Comparison of ecological quality indices based on benthic macrofauna and sediment profile images: a case study along an organic enrichment gradient off the Rhône River. Ecol. Indicat. 12: 133-142.

Lee, H.W., Bailey-Brock, J.H., McGurr, M.M. (2006). Temporal changes in the polychaete assemblage surrounding a Hawaiian mariculture operation. Mar. Ecol. Prog. Ser. 307: 175-185.

Marín-Guirao, L., Cesar, A., Marín, A., Lloret, J., Vita, R. (2005). Establishing the ecological quality of soft-bottom mining-impacted coastal water bodies in the scope of the Water Framework Directive. Mar. Poll. Bull. 50: 374-387.

Martínez-García, E., Sánchez-Jerez, P., Aguado-Giménez, F., Ávila, P., Guerrero, A., Sánchez-Lizaso, Fernández-González, V., González, N., Gairin, J.I., Carballeira, C., García-García, B., Carreras, J., Macías, J.C., Carballeira, A., Collado, C. (2013). A meta-analysis approach to the effects of fish farming on soft-bottom polychaeta assemblages in temperate regions. Mar. Poll. Bull. 69(1-2): 165-171.

Méndez, N. (2002). Annelid assemblages in soft bottoms subjected to human impact in the Urías estuary (Sinaloa, México). Oceanol. Acta 25: 139-147.

Méndez, N., Flos, J., Romero, J. (1998). Littoral soft-bottom polychaete communities in a pollution gradient in front of Barcelona (Western Mediterranean, Spain). Bull. Mar. Sci. 63: 167-178.

Miron, G., Landry, T., Archambault, P., Frenette, B. (2005). Effects of mussel culture husbandry practices on various benthic characteristics. Aquaculture 250: 138-154.

Musco, L., Terlizzi, A., Licciano, M., Giangrande, A. (2009). Taxonomic structure and the effectiveness of surrogates in environmental monitoring: a lesson from polychaetes. Mar. Ecol. Prog. Ser. 383: 199-210.

Mutlu, E., Cinar, M.E., Ergev, M.B. (2010). Distribution of soft-bottom polychaetes of the Levantine coast of Turkey, Eastern Mediterranean Sea. J. Mar. Syst. 79: 23-35.

Nickell, T.D., Cromey, C.J., Borja, A., Black, K.D. (2009). The benthic impacts of a large cod farm. Are there indicators for environmental sustainability? Aquaculture 295: 226-237.

Olsgard, F., Sommerfield, P.J. (2000). Surrogates in marine benthic investigations: which taxonomic unit to target? J. Aquat. Ecosys. Stress Rec. 7: 25-42.

Olsgard, F., Brattegard, T., Holthe, T. (2003). Polychaetes as surrogates for marine biodiversity: lower taxonomic resolution and indicators groups. Biodivers. Conserv. 12: 1033-1049.

Pearson, T.H., Rosenberg, R. (1978). Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. Oceanogr. Mar. Biol Ann. Rev. 16: 229-311.

Pinedo, S., Joedana, E. (2007). Spain (Catalonia and Balearic Islands). In: Carletti, A., Heiskanen, A-S. (Eds.). Water Framework Directive Intercalibration Technical Report

Part 3: Coastal and Transitional Waters. JRC Scientific and Technical Reports. JRC, ies. pp. 62-70.

Pinedo, S., Jordana, E., Salas, F., Subida, M.D., García Adiego, E., Torres, J. (2012). Testing MEDOCC and BOPA indices in shallow soft-bottom communities in the Spanish Mediterranean coastal waters. Ecol. Indicat. 19: 98-105.

Pinto, R., Patricio, 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. Indicat. 9: 1-25.

Pranovi, F., Da Ponte, F., Torricelli, P. (2007). Application of biotic indices and relationship with structural and functional features of macrobenthic community in the lagoon of Venice: an example over a long time series of data. Mar. Poll. Bull. 54: 1607-1618.

PRIMER (2006). PRIMER 6: for Windows, Version 6.1.8. PRIMER-E, Plymouth, UK.

Quinn, G.P., Keough, M.J. (2002) Experimental design and data analysis for biologist. Cambridge University press. UK. 557 pp.

Quintino, V., Elliott, M., Rodrigues, A.M. (2006). The derivation, performance and role of univariate and multivariate indicators of benthic change: case studies at different spatial scales. J. Exp. Mar. Biol. Ecol. 330: 368-382.

Quintino, V., Azevedo, A., Magalhaes, L., Sampaio, L., Freitas, R., Rodrigues, A.M., Elliott, M. (2012). Indices, multispecies and synthesis descriptors in benthic assessments: intertidal organic enrichment from oyster farming. Est. Coast Shelf Sci. 110: 190-201.

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. Poll. Bull. 68(1-2): 117-126.

Riera, R., Sánchez-Jerez, P., Rodríguez, M., Monterroso, O., Ramos, E. (2012). Long-term monitoring of fish farms: application of Nematode/Copepod index to oligotrophic conditions. Mar. Poll. Bull. 64: 844-850.

Rosenberg, R., Blomqvist, M., Nilsson, H.C., Cederwall, H., Dimming, A. (2004). Marine quality assessment by use of benthic species-abundance distributions: a proposed new protocol within the European Union Water Framework Directive. Mar. Poll. Bull. 49: 728-739.

Salas, F., Marcos, C., Neto, J.M., Patricio, J., Pérez-Ruzafa, A., Marques, J.C. (2006). User-friendly guide for using benthic ecological indicators in coastal and marine quality assessment. Ocean Coast Manage. 49: 308-311.

Sampaio, L., Rodrigues, A.M., Quintino, V. (2011). Can biotic indices detect mild organic enrichment of the seafloor? Ecol. Indicat. 11: 1235-1244.

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

Soares-Gomes, A., Mendes, C.L.T., Tavares, M., Santi, L. (2012). Taxonomic sufficiency of polychaete taxocenes for estuary monitoring. Ecol. Indic. 15: 149-156.

Tataranni, M., Lardizzi, C. (2010). Performance of some biotic indices in then real variable world: a case study at different spatial scales in North-Western Mediterranean Sea. Environ. Pollut. 158: 26-34.

Tomassetti P, Porrello S (2005). Polychaetes as indicators of marine fish farm organic enrichment. Aquac. Int. 13:109-128.

Tomassetti, P., Persia, E., Mercatali, I., Vani, D., Marusso, V., Porrello, S. (2009). Effects of mariculture on macrobenthic assemblages in a western Mediterranean site. Mar. Poll. Bull. 58: 533-541.

Underwood, A. J. (1991). Beyond BACI: experimental designs for detecting human environmental impacts on temporal variations in natural populations. Aust. J. Mar. Fresh. Res. 42: 569–587.

Underwood, A.J. (1997). Experiments in Ecology: Their Logical Design and Interpretation Using Analysis of Variance. Cambridge University Press, Cambridge, pp. 504.

Warwick, R.M., Clarke, K.R., Somerfield, P.J. (2010). Exploring the marine biotic index (AMBI): variations on a theme by Ángel Borja. Mar. Poll. Bull. 60: 554-559.

Wheeler, Q. D. (2004). Taxonomic triage and the poverty of phylogeny. Philos. Trans. R. Soc. Lond. B Biol. Sci. 359:571–583.

Wildish, D.J., Akagiu, H.M., Hamilton, N., Hargrave, B.T. (1999). A recommended method for monitoring sediments to detect organic enrichment from mariculture in the Bay of Fundy. Can. Tech. Rep. Fish. Aquat. Sci. 2286, iii + 31p.

Table 1: Ecological Quality Classification of BOPA index (Dauvin and Ruellet, 2007).

BOPA range of values	EcoQ.
0.00000 – 0.04576	High
0.04576 – 0.13966	Good
0.13966 – 0.19382	Moderate
0.19382 – 0.26761	Poor
0.26761 – 0.30103	Bad

Table 2: Polychaete families´richness and abundance (mean ± se) of the whole assemblage and of those polychaete families involved in BBI´s calculations with a higher contribution to dissimilarities among distances over the impact gradients.

		BC (42 families; 19850 indiv.; 184 indiv. per sample)		LB (46 families; 9768 indiv.; 90 indiv. per sample)		RF (44 families; 7705 indiv.; 71 indiv. Per sample)		TOTAL (48 families; 37323 indiv.)	
BBI presence	Family	Abund.	mean. ± sem	Abund.	mean. ± sem	Abund.	mean. ± sem	Abund.	mean. ± sem
Both	*Capitellidae*	1456	13.4 ± 1.5	2188	20.5 ± 2.7	1146	10.6 ± 0.8	4790	14.8 ± 1.1
BOPA	*Cirratulidae*	455	4.2 ± 0.8	830	7.6 ± 1.0	616	5.7 ± 0.7	1901	5.8 ± 0.5
BOPA-FF	*Dorvilleidae*	6672	61.7 ± 7.2	174	1.6 ± 0.3	250	2.3 ± 0.9	7096	21.9 ± 2.8
BOPA	*Eunicidae*	740	6.8 ± 1.1	234	2.1 ± 0.3	86	0.8 ± 0.1	1060	3.2 ± 0.4
BOPA-FF	*Glyceridae*	256	2.3 ± 0.3	83	0.7 ± 0.1	86	0.7 ± 0.1	425	1.3 ± 0.1
BOPA-FF	*Nereididae*	2518	23.3 ± 5.6	387	3.5 ± 1.1	43	0.4 ± 0.1	2948	9.1 ± 2.0
BOPA-FF	*Oweniidae*	639	5.9 ± 1.1	230	2.1 ± 0.4	210	2.0 ± 0.3	1079	3.3 ± 0.4
None	*Paraonidae*	1475	13.6 ± 2.8	788	7.2 ± 0.8	1471	13.6 ± 1.5	3734	11.5 ± 1.1
BOPA	*Spiochaetopteridae*	0	-	0	-	0	-	0	-
Both	*Spionidae*	719	6.6 ± 0.9	536	4.9 ± 0.5	695	6.4 ± 0.6	1950	6.0 ± 0.4

Table 3: Permutational multivariate analyses of variance (PERMANOVA) of polychaete assemblage, environmental variables, BOPA and BOPA-FF data sets. F: fish farms; T: time; D: distances; S: sites within F, T and D.

			Polychaete assemblage		*FFS*		*TFS*		*BOPA*		*BOPA-FF*	
Source of variation	d.f.	Denom. d.f.	MS	*P*(perm)	MS	*P*(perm)	MS	*P*(perm)	MS	*P*(perm)	MS	*P*(perm)
F	5	5	29130	**0.0018**	25931	0.0556	29900	**0.0064**	6071	0.1162	1633	0.5800
T	1	5	22357	**0.0352**	17190	0.1670	4287	0.3304	1218	0.5986	719	0.6162
D	2	10 + 2	53862	**0.0388**	2564	0.8612	64397	**0.0074**	13144	0.1284	24332	**0.0416**
F x T	5	72	5534	**0.0002**	6416	**0.0002**	3710	**0.0002**	2139	**0.0014**	1789	**0.0034**
F x D	10	10	22030	**0.0002**	5283	**0.0226**	9515	**0.0002**	6417	**0.0046**	7784	**0.0002**
T x D	2	10	4228	0.5692	3504	0.1678	3933	0.1662	742	0.7514	231	0.9398
F x T x D	10	72	4910	**0.0002**	1750	**0.0002**	2287	**0.0110**	1560	**0.0020**	983	0.0600
S(F x T x D)	72	216	1927	**0.0002**	262	**0.0002**	1297	**0.0002**	638	**0.0002**	605	**0.0002**
Residual	216		807		24		223		307		255	
Total	323											

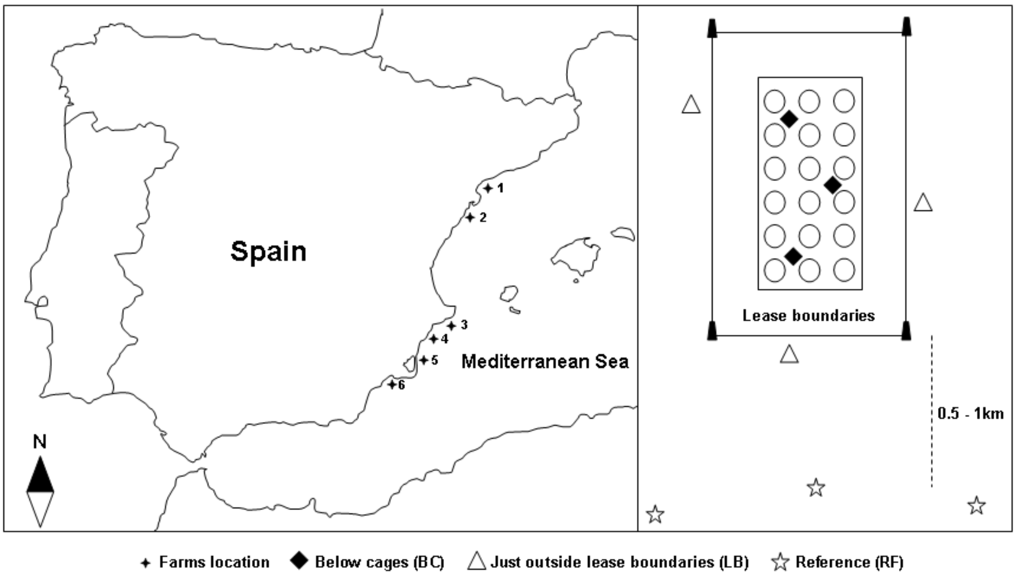


Figure 1: Localisation of the study area and diagram of the sampling design.

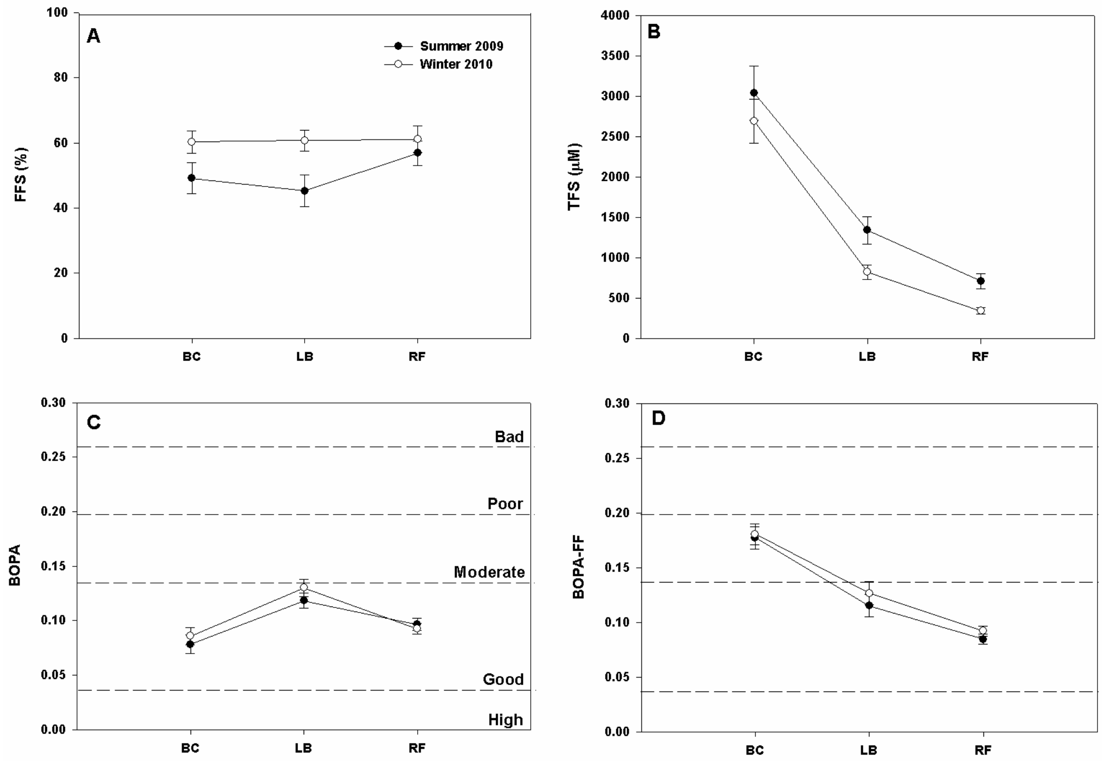


Figure 2: Environmental variables and Benthic Biotic Indices results (mean ± s.e.) for the different distances and sampling times (FFS: finest fraction of the sediment; TFS: total free sulphides; BC: below cages; LB: lease boundaries; RF: reference locations; PI: production intensity).

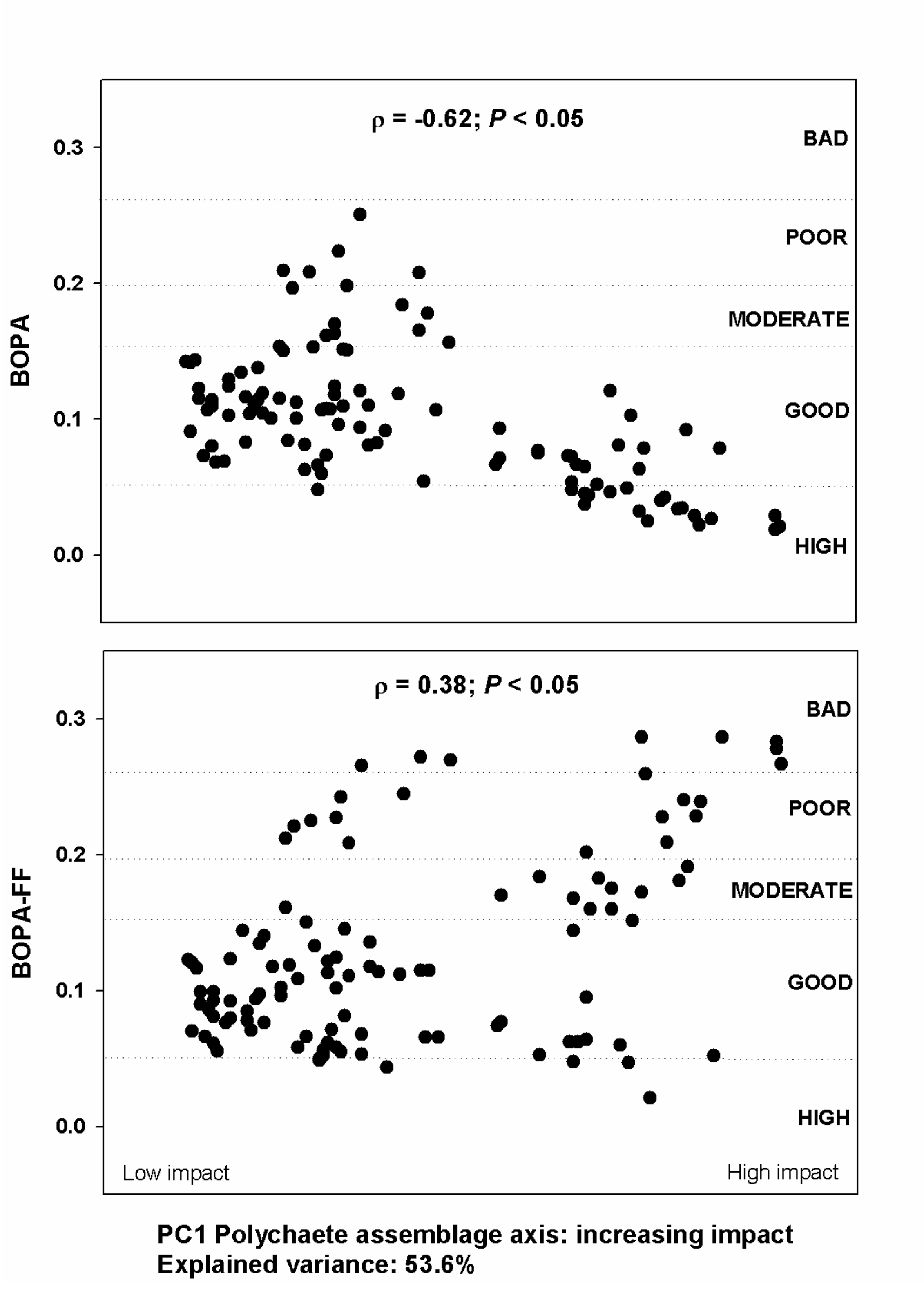


Figure 3: Plots of BOPA and BOPA-FF averaged scores by sites in relation with the impact gradient (PC1) described by the polychaete assemblage structure (ρ: Spearman rank correlation coefficient).

- We applied taxocene surrogation and taxonomic sufficiency to fish farming monitoring
- Multivariate assessment of polychaete assemblage and BOPA index were compared
- Polychaete assemblage provided a suitable picture of the impact gradient
- We recalculate BOPA-FF including polychaete locally identified as indicative
- BOPA-FF correlated with the impact gradient and improved the diagnosis