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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

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ABSTRACT

The European Union-funded ECASA project (Ecosystem Approach for Sustainable Aquaculture) studied the impacts from aquaculture on ecosystems from northern Norway to Greece. The objectives of this investigation were to identify quantitative indicators of the effects of aquaculture on marine communities, and to assess their applicability over a range of ecosystems and aquaculture production systems. The study included 6 Mediterranean and 4 Atlantic sites, 7 of which produced finfish (seabream, seabass, tuna, salmon and cod), and 2 bivalve molluscs (oysters, mussels, and clams); one site produced both fish and bivalves. Cultivation methods included finfish cages, long-lines and trestles. Similar sampling methodologies were employed at the 10 study sites, obtaining sediment, hydrodynamic, and benthic faunal data. The horizontal impact from organic enrichment extended 50 m from the farms, with contradictory responses in several indicators (individual abundance, biomass) and a more consistent response of the Infaunal Trophic Index (ITI) and AZTI's Marine Biotic Index (AMBI). By means of Partial Redundancy Analysis, it was demonstrated that the environmental variables explained 53.2% of the variability in the macrofaunal variables (individual abundance, species richness, diversity, AMBI and ITI), whilst the explained variance was partialled out within three groups of variables: (i) 'hydrography' (depth, distance to farm, average current speed), which explained 11.5% of the variance; (ii) 'sediment' (Eh and percentages of silt and total organic matter), which explained 5.4%; and (iii) 'cages' (years of production and annual production), which explained 15.2%. The shared variance explained by interactions among these groups was 21.1%. These results, together with multiple regression analysis, provide an accurate assessment of the degree of impact from aquaculture. In conclusion, the use of several benthic indicators, in assessing farm impacts, together with the investigation of dynamics of the studied location, water depth, years of farm activity, and total annual production, must be included when interpreting the response of benthic communities to organic enrichment from aquaculture.

1. Introduction

Marine aquaculture continues to expand both globally and within Europe bringing benefits to society, often in fragile coastal communities where traditional employment opportunities are in decline (FAO, 2007). However, there are well-documented cases where aquaculture has a negative impact on the environment (Karakassis et al., 2000; Black, 2001; Buschmann et al., 2006; Kalantzi and Karakassis, 2006; Pergent-Martini et al., 2006; Apostolaki et al., 2007; Giles, 2008; Holmer et al., 2008). These studies have focussed on the effects of waste products (dissolved and particulate nutrients, chemicals and medicines) on benthic and planktonic communities, the transmission of genes, parasites and diseases between wild and cultured species, as well as interactions with both local and oceanic fisheries. There are also well-known conflicts between aquaculture and other coastal uses (Michler-Cieluch and Kodeih, 2008).

Although considerable work has been conducted on this topic, much of the information gathered has yet to be integrated, analysed and

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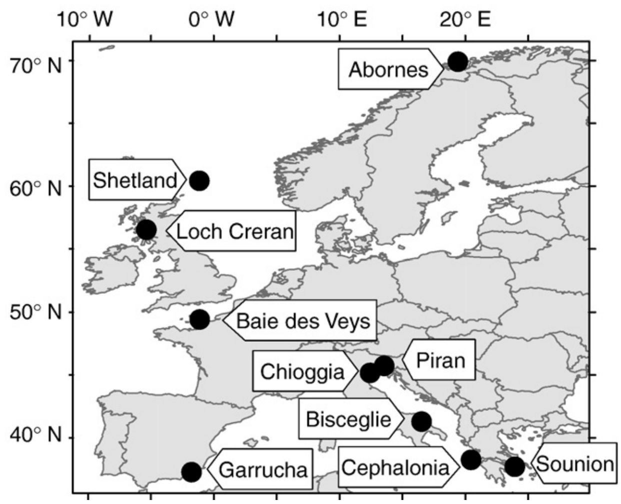

Fig. 1. Position of the sampling locations across Europe.

disseminated, and there remains a need to develop a quantitative understanding and predictive capability in the diverse European ecosystems where aquaculture is practised. In particular, the advances recently made in understanding ecosystem functioning and resilience of coastal seas through several programmes of oceanographic research (e.g. ELOISE, see Pacyna et al., 2005) have not been adequately integrated with the more applied coastal science which has, in general, focused on the local scale.

Similarly, while much effort has been expended on the environmental requirements of farmed species, there is a need for good examples where the ecosystem services required by aquaculture are considered at the broader scale. Such considerations allow assessment of resource use and minimisation of risks to the aquaculture sector from ecosystem degradation (FAO, 2007). In the absence of a holistic ecosystem approach, selection of sites for aquaculture is therefore often inadequate both with respect to the receiving environment and to ecosystem services to cultured species. The concepts of carrying capacity (i.e. available resources, particularly food), assimilative capacity (i.e. recycling capacity of organic matter and nutrients) and holding capacity (i.e. sustainable production) as they relate to aquaculture, are important, but elusive aspects, though they have received considerable attention from regulatory bodies and investigators (e.g. Fernandes et al., 2001, Stigebrandt et al., 2004; McKindsey et al., 2006; Ferreira et al., 2008; Sequeira et al., 2008).

Recent legislation worldwide has emphasised the importance of an ecosystem-based approach when managing marine activities (Garcia et al., 2003; Borja et al., 2008). In this context, the European project ECASA (Ecosystem Approach for Sustainable Aquaculture) (http://www.ecasa.org.uk/) focused on functional relationships and processes within ecosystems in response to different aquaculture impacts, from Greece to northern Norway (Fig. 1). To achieve these objectives, knowledge of coastal hydrodynamics, biogeochemistry, sediment dynamics, and understanding of the community ecology of benthic and pelagic components that are impacted by anthropogenic activities, are needed.

In this contribution, only the benthic component has been investigated. The process of testing benthic indices for the assessment of environmental impacts has been reviewed by Borja and Dauer (2008). Several indicators of the effects of aquaculture on benthic communities have been proposed but only a few are suitable for use across a wide range of different ecosystem types. The *Infaunal Trophic Index* (ITI) (Word, 1979) has been used in aquaculture impact assessment in a variety of environments (Maurer et al., 1999; Cromey et al., 2002a; Aguado-Giménez et al., 2007). Another index, *AZTI's Marine Biotic Index* (AMBI) (Borja et al., 2000), has been used to detect different sources of impact along European coasts (Borja et al., 2003) including aquaculture (Muxika et al., 2005; Carvalho et al., 2006; Sanz-Lázaro and Marín, 2006; Aguado-Giménez et al., 2007; Bouchet and Sauriau, 2008; Callier et al., 2008). Finally, many other indicators and indices, such as diversity, richness, abundance, biomass, dominance, evenness, etc., have been investigated elsewhere in assessing aquaculture impacts (Jones, 2002; Hargrave, 2004; Edgar et al., 2005; Aguado-Giménez et al., 2007; Gibbs, 2007).

The objectives of this investigation were to identify the suitability of selected quantitative indicators in the assessment of the effects of aquaculture on benthic communities in soft sediments, to assess their applicability over a range of ecosystems and aquaculture production systems, and to study the factors (e.g. dynamic, production, location, etc.) to which these indicators respond, on a pan-European scale.

The benthic indicators to be tested were selected on the basis of the following criteria: (i) direct relevance to ECASA project objectives; (ii) easily understood by stakeholders and fit for purpose (indicators were defined clearly in order to avoid confusion in their development or interpretation); (iii) realistic collection or development costs (indicators need to be practical and cost-effective); (iv) high quality and reliability (the information they provide is only as good as the data from which they are derived) and (v) appropriate spatial and temporal scale.

2. Methods

2.1. Sampling design

The ECASA study sites ranged across Europe and covered a wide range of latitudinal locations, water depths, bottom sediment types, and different cultured species (Fig. 1, Table 1). This investigation encompassed 10 study sites, consisting of 6 Mediterranean and 4 Atlantic locations (Table 1). Most of the locations (7) were finfish farms (seabream, seabass, tuna, salmon and cod), 2 were bivalve mollusc farms (oyster, mussel, and clam), and one produced both fish

Table 1
Locations sampled in each country, including the number of sampling stations, the species farmed, the culture method, annual production (during the year previous to the sampling), years of production, average current speed at 15 m water depth (except in Piran, which was at 7 m).

Country	Location	Stations (nr)	Depth range (m)	Silt content range (%)	Species	Culture method	Annual production (tons)	Years of production (nr)	Averaged current speed (cm s^{-1})
Greece	Sounion	6	12.5–17.1	0.1–2.6	Seabream-Seabass	Cages	320	8	6.5
Greece	Cephalonia	6	19.7–20.6	53.5–94.2	Seabream-Seabass	Cages	1000	22	2.4
Slovenia	Piran	8	13.0–13.0	100	Seabream-Mussel	Cages-Long-lines	71	14	2.5
Italy	Bisceglie	7	19.4–22.3	40.0–67.2	Seabream-Seabass	Cages	800	15	13.8
Italy	Chioggia	4	22.5–25.1	21.8–58.0	Mussel	Long-lines	600	13	12
Spain	Garrucha	10	53.0–62.0	11.7–37.0	Tuna	Cages	218	3	14
France	Baie des Veys	6	Intertidal	–	Oyster-Mussel-Clams	Trestle and pole	No data	No data	No data
United Kingdom	Loch Creran	7	22.0–31.0	33.9–83.1	Salmon	Cages	1500	23	5
United Kingdom	Shetland	7	16.0–32.0	12.3–34.1	Cod	Cages	1390	21	5.2
Norway	Abornes	4	54.0–58.0	9.6–12.0	Salmon	Cages	1516	0.5	7.2
TOTAL AND TOTAL RANGE		65	0.0–62.0	0.1–100			71–1516	0.5–23	2.4–14

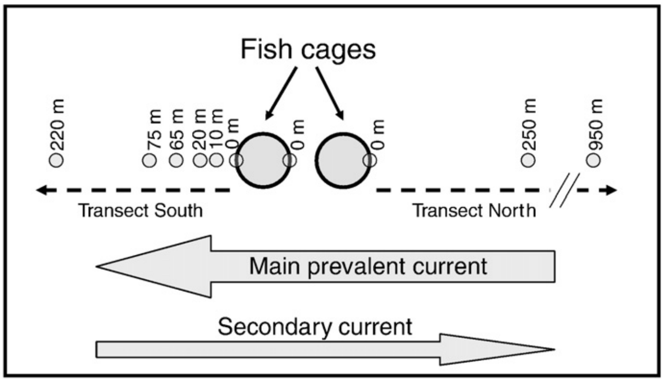

Fig. 2. Example of the sampling design in Garrucha (Spain), oriented along the main current direction in the area.

and bivalves. Farming methods included cages, long-lines and trestles or 'trays' (Table 1), being the later an oyster aquaculture method consisting on timber or metal frames (trestles), supporting the mesh oyster bags, and sited on the foreshore in the intertidal area.

At each of the study sites sampling station positions were selected to reflect gradients of impact, where maximum impact was assumed to occur at, or adjacent to, the farm and decrease with distance, dropping to "nil" at the reference station. Where possible station selection was performed by preliminary modelling (using current records, e.g. Garrucha, Loch Creran, Shetland), whilst in other locations (e.g. Bisceglie, Piran) pre-survey testing was done to detect such gradients. To maximise chances of detecting a gradient, and to include distance from the farm as a variable, 4 to 10 stations per site were sampled (at 0 m, 5 m, 10 m, 25 m, 50 m, 100 m, and 200 m from the farm). At least one reference station, at similar depth and sediment type but at a location considered not to be influenced by the farm, in terms of organic matter deposition, was sampled at each location (see Fig. 2, for an example of a sampling scheme). At Creran, Shetland, Garrucha and Bisceglie two opposed transects (starting beneath the cages) within the main current direction were sampled, whereas at Piran three transects were sampled. Samples were taken when impacts were likely to be maximal, i.e. at the end of the production cycle within the farm (normally in summer, except for tuna, which is in March-April). All sampling was carried out in 2006.

At each subtidal station, at least 4 grab (van Veen, 0.1 m^2) replicates were taken for macrofauna, and triplicate cores (generally

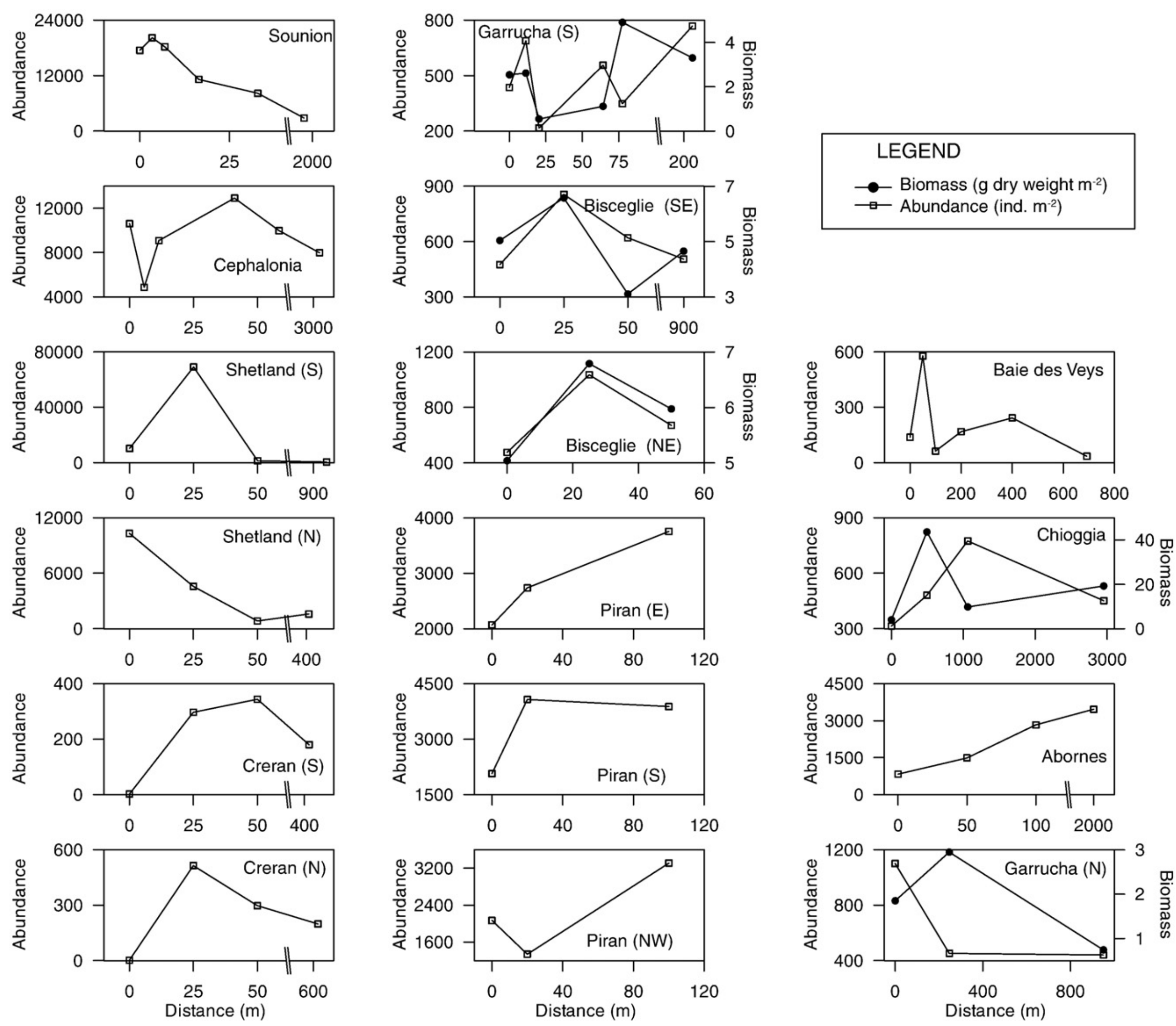

Fig. 3. Spatial variability of macrofaunal abundance across the studied transects. The direction is shown between brackets at locations with several transects sampled (S: South, N: North, E: East, W: West). At Bisceglie, Chioggia and Garrucha the macrofaunal biomass is also shown. Distance '0' m is beneath the cages (see Fig. 1).

Table 2
Pearson's correlation coefficients and *p*-values between the studied variables, taking into account data from sampled sites beneath the cages.

	Latitude	Production	Depth (sr)	Current (log)	Years	Silt	TOM	Eh	Abundance (log)	Richness	Diversity	AMBI
Production	0.77*											
Depth (sr)	0.46	0.39										
Current (log)	−0.09	−0.10	0.46									
Years	−0.03	0.32	−0.53	−0.49								
Silt	−0.34	−0.34	−0.56	−0.44	0.40							
TOM	0.06	0.10	−0.45	−0.79*	0.49	0.67*						
Eh	0.12	0.12	0.24	0.25	−0.24	−0.67*	−0.72*					
Abundance (log)	−0.34	−0.40	−0.20	−0.15	−0.25	−0.07	−0.33	0.54				
Richness	−0.05	−0.13	0.70*	0.49	−0.77*	−0.41	−0.59	0.27	0.37			
Diversity	−0.11	−0.33	0.30	0.51	−0.84**	−0.09	−0.36	0.02	0.28	0.71*		
AMBI	0.15	0.37	−0.28	−0.71*	0.78*	0.08	0.51	−0.13	−0.15	−0.58	−0.91***	
ITI	−0.15	−0.41	0.44	0.75*	−0.86**	−0.19	−0.58	0.16	0.10	0.67*	0.86**	−0.96***

Key: log — log-transformed; sr — square root transformed. Correlations significant at $p<0.001$ (***), $p<0.01$ (**) and $p<0.05$ (*). Note: only subtidal locations have been used in the calculation.

5 cm of diameter, 30 cm length) for sediment analyses. In Baie des Veys all the stations were located in intertidal areas and sampled directly by hand cores. Hand coring was also performed at the two Greek sites, Sounion and Cephalonia, using SCUBA-divers. Sieves used for extraction of the macrofauna from sediments had a mesh size of 1.0 mm. After sorting out the samples, taxa were identified to species level.

The variables used to characterize sediments were organic matter content (here expressed as percentage of loss on ignition; Total Organic Matter (TOM)), percentage of silt (i.e., sediment fraction

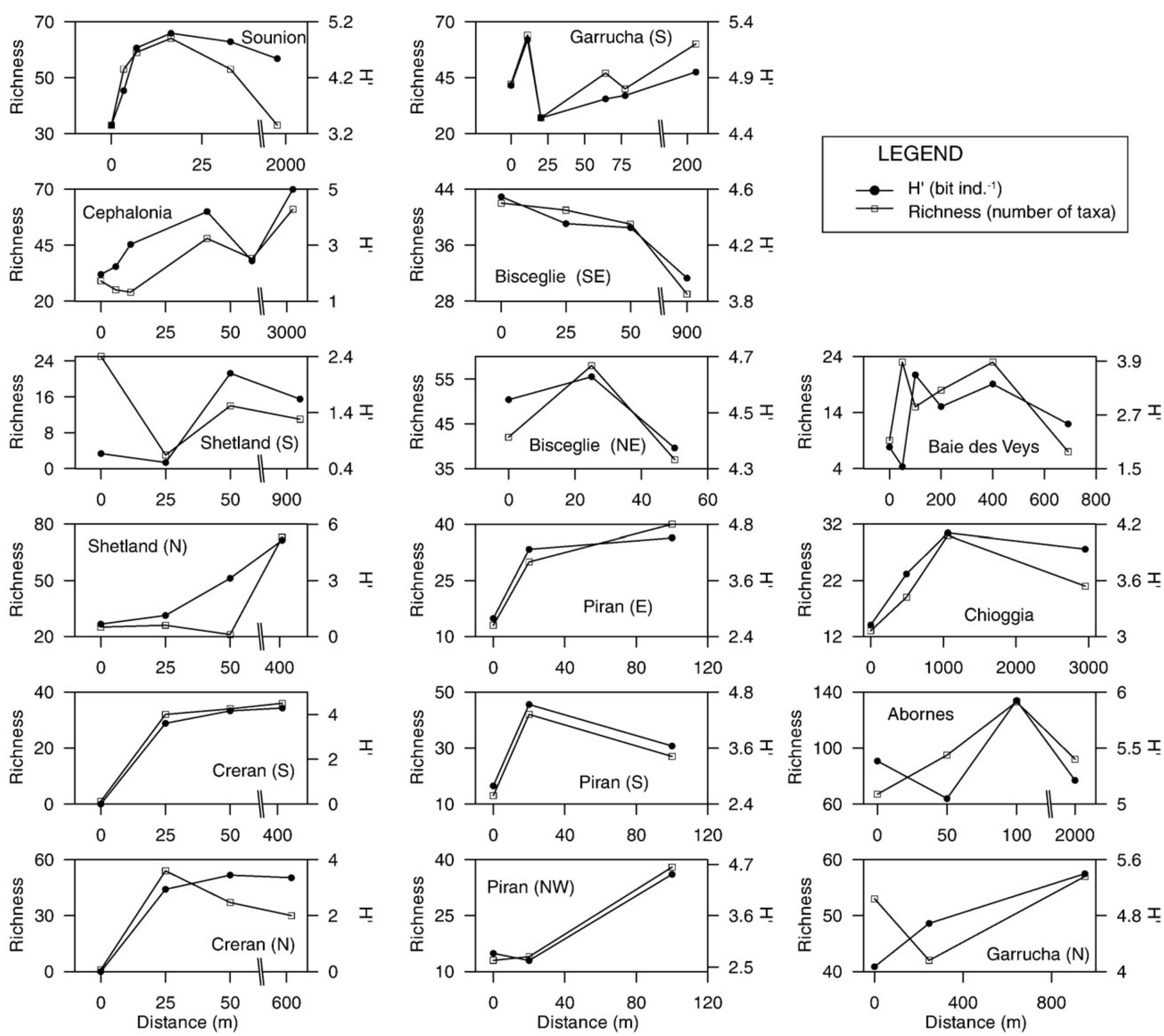

Fig. 4. Spatial variability of diversity (H') and species richness of macrofauna assemblages across the studied transects. The direction is indicated as in Fig. 2. Distance '0' m is beneath the cages (see Fig. 1).

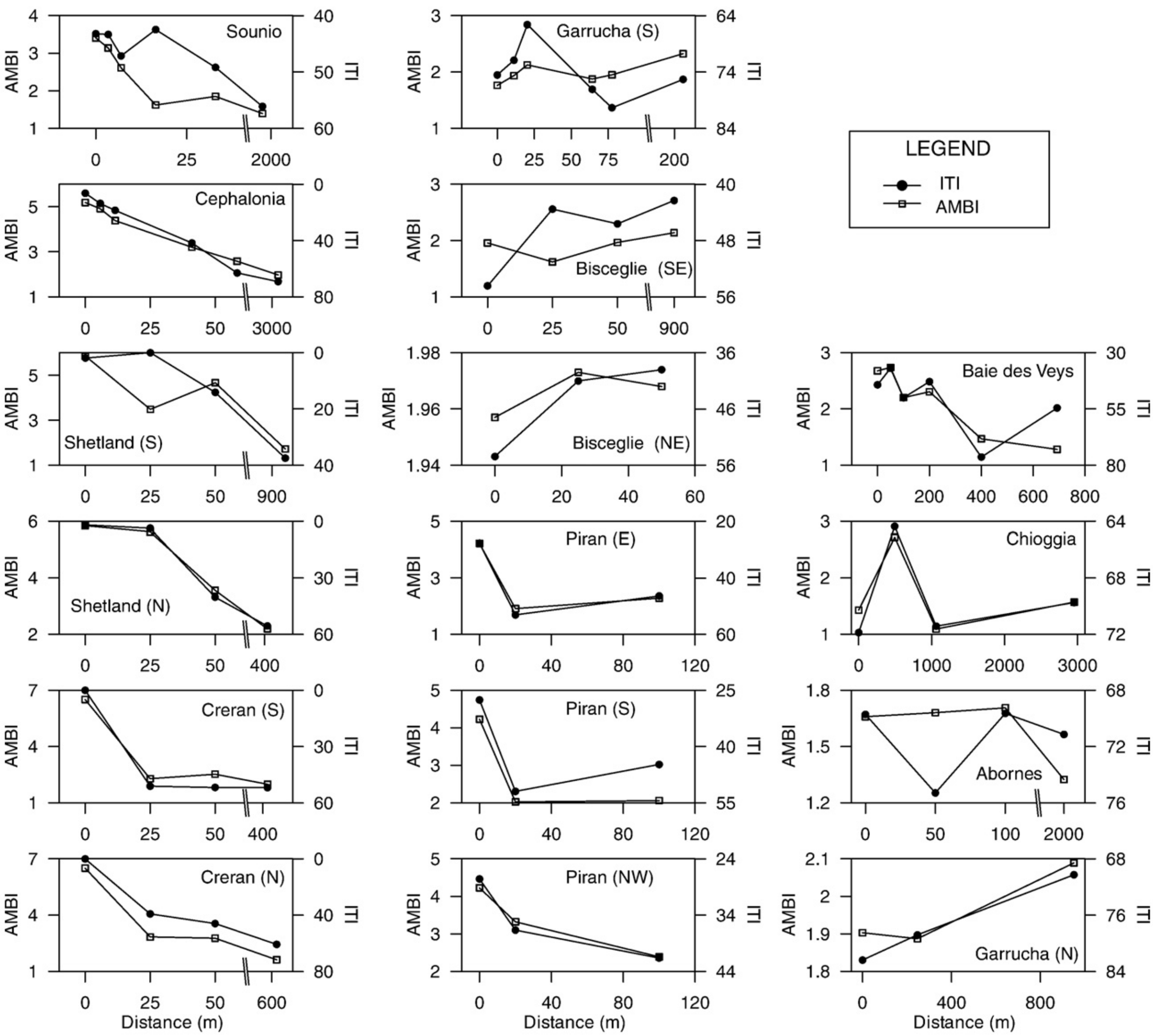

Fig. 5. Spatial variability of ITI (Infaunal Trophic Index) and AMBI (AZTI's Marine Biotic Index) across the studied transects. The direction is indicated as in Fig. 2. Distance '0' m is beneath the cages.

<63 µm: Eleftheriou and MacIntyre, 2005) and redox potential (i.e., Eh, following Langmuir, 1971).

The hydrographic environment of each site is considered an important variable in structuring benthic impacts (Giles, 2008). Hence, current velocity and direction were measured at each site by means of Acoustic Doppler Current Profilers (ADCP) or by conventional current meters (e.g. InterOcean S4). Instruments were deployed for a period of one month or more, and data were recorded using a sampling interval of 10 min. In addition, data on years of farm activity and production volumes were obtained for each study site.

2.2. Biological indicators

The biological data sets, containing individual abundance and species composition, per replicate and station, were checked and standardised (correcting taxonomic and typographical errors), and the key structural parameters (e.g. individual abundance) were standardised per square meter. At several of the locations macrobenthic biomass was derived as dry weight (by drying samples at 80 °C until constant weight). Univariate variables were calculated from this dataset, including Shannon-Wiener's diversity, ITI, and AMBI. AMBI was calculated using AMBI software (version 4.1, downloadable from www.azti.es) and a species-list established in December 2007. Guidelines specified in Borja and Muxika (2005) were referred to in deriving this index.

2.3. Statistical analyses of data from subtidal areas

Partial ReDundancy Analysis (pRDA) was carried out to study the variation in the macrobenthic univariate summary statistics (individual abundance, diversity, richness, AMBI, ITI) among (i) hydrographical features (average current speed, depth, distance from farms), (ii) farm characteristics or husbandry practices (years of production, annual production in tons), and (iii) sediment characteristics (Eh, percentage of silt, TOM). The pRDA was carried out following Bocard et al. (1992) and Legendre et al. (2005).

Multiple regression models assessing the suitability of predictor responses of the benthic characteristics (individual abundance, diversity, richness, AMBI, ITI) were carried out taking as independent variables the parameters from aquaculture farm and from hydrography (see above).

The statistical analyses were performed by S-Plus 2000, R Commander (Fox, 2005) and CANOCO for Windows (ter Braak and Smilauer, 2002).

3. Results

3.1. Spatial variability of the benthic characteristics in relation to distance to the farm

Macrofaunal abundance varied along the transects and differed between sites (Fig. 3). At 11 out of the 16 studied transects, the abundance increased and peaked within a radius of 5 to 50 m from the edge of the farm (Fig. 3). In Abornes and Piran (transect E) macrofaunal abundance increased with distance from the cages, whilst at Sounion, Shetland (transect N) and Garrucha (transect N) the opposite pattern was found (Fig. 3). In Garrucha (transect S) the maximum abundance was found at 75 m from the cages (Fig. 3). Macrofaunal biomass was also evaluated along 5 transects. With the exception of Bisceglie, the maximum biomass did not coincide within the maximum abundance (Fig. 3). Taking subtidal data into account from sampling points located beneath the farms, abundance was not significantly correlated with farm husbandry, hydrography or sediment characteristics (Table 2).

Macrofaunal diversity (Shannon-Wiener H') and species richness showed relatively similar patterns within each of the studied transects, but dissimilar patterns among the different studied sites (Fig. 4). Thus, at 10 out of the 16 studied transects the diversity and richness were minimal beneath the farm, whereas at others (e.g. Bisceglie and Garrucha (transect S)), this was not observed (Fig. 4). Considering subtidal data from stations located beneath the farm, species richness was positively and significantly correlated with depth, and both species richness and diversity were negatively correlated with the number of years of production (Table 2).

ITI and AMBI showed relatively similar patterns within each studied transect, suggesting an increase in benthic quality with distance from the farms in Sounion, Cephalonia, Shetland, Creran, Piran and Baie des Veys (Fig. 5). Some differences were apparent along other transects. In Bisceglie for example, AMBI was almost constant with distance, whereas ITI indicated a decrease in benthic quality with distance from the farm (Fig. 5). In Abornes, AMBI was almost constant within the first 100 m, whilst ITI identified better benthic quality at a distance of 50 m from the farm (Fig. 5). It is noteworthy that both ITI and AMBI were significantly (though inversely) correlated with the average current speed and the number of years of production (Table 2). These relationships indicated an increase in the benthic quality with higher current speed and fewer years of production.

3.2. Relationships between benthic characteristics and cages and hydrography in subtidal areas

These relationships are summarized in Table 3. The redundancy analysis shows that environmental variables explain 53.2% of the variability in the macrofauna parameters (abundance, richness, diversity, AMBI and ITI) (F-ratio = 6.68; p-value < 0.001; Monte Carlo permutation test). Species richness, diversity and ITI were positively correlated with Eh, distance from farm, current speed and depth, and negatively correlated with TOM, years of operation and the annual production (Fig. 6a). AMBI was inversely correlated with Eh, distance from farm, current speed and depth, and positively correlated with the TOM content, the years and the annual rate of production (Fig. 6a). The explained variance was partialled out within three groups of variables: (i) 'hydrography' (depth, distance to the farm, average current speed), (ii) 'sediment' (Eh and percentages of silt and TOM), and (iii) 'cages' (years of operation and annual production). Thus, the proportion of the total variation in macrofaunal structure, explained by interactions among these groups (i.e., shared variance), was 21.1%, and the variance explained by 'cages', 'hydrography', and 'sediment' was 15.2%, 11.5% and 5.4%, respectively (Fig. 6b).

Macrofaunal responses to the 'hydrography' and 'cages' were adjusted by multiple linear regressions (Table 4). The percentage of

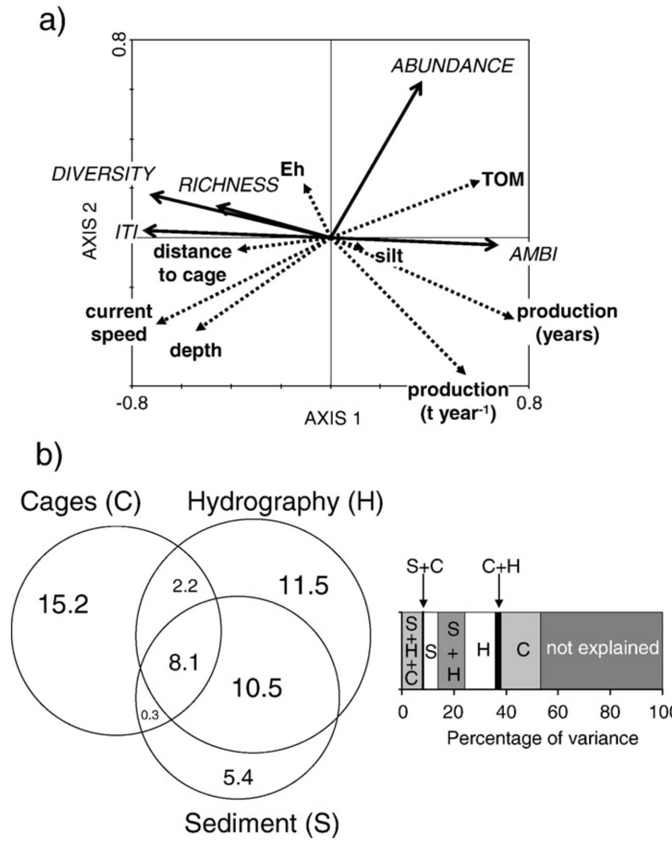

Fig. 6. a) ReDundancy Analysis (RDA) biplot of the relationships between the macrofaunal characteristics (full-line arrows) and the environmental parameters (dashed arrows). The arrows indicate the direction of increase for the variables studied. The angles between variables reflect their correlations (angles near 90° indicate no correlation, angles near 0° indicate high positive correlation and angles near 180° indicate high negative correlation). b) Partition of the variation of the macrobenthic characteristics (obtained by partial RDA) among three groups of explanatory variables: (H) hydrographical characteristics: average current speed, depth, and distance to cages; (C) characteristics of farms: years of production and annual production in tons; and (S) sediment characteristics: Eh, percentage of silt, and total organic matter (TOM).

Table 3
Pearson's correlation coefficients and p-values between the studied variables, taking into account data from all the studied sites.

	Latitude	Production	Depth (sr)	Current (log)	Years	Distance (log1)	Silt	TOM	Eh	Abundance (log)	Richness	Diversity	AMBI
Silt	−0.14	−0.20	-0.50^{***}	-0.53^{***}	0.41^{***}	0.10							
TOM	0.07	−0.05	-0.62^{***}	-0.85^{***}	0.46^{***}	−0.02	0.78^{***}						
Eh	0.20	0.27^{*}	0.28^{*}	0.20	−0.09	0.24	-0.70^{***}	-0.57^{***}					
Abundance (log)	−0.18	−0.19	-0.38^{**}	-0.40^{**}	−0.00	−0.06	0.03	0.24	0.15				
Richness	0.04	0.01	0.48^{***}	0.25	-0.53^{***}	0.05	-0.29^{*}	-0.26^{*}	0.16	0.16			
Diversity	-0.29^{*}	-0.37^{**}	0.35^{**}	0.37^{**}	-0.62^{***}	0.25^{*}	−0.09	−0.25	0.04	−0.01	0.69^{***}		
AMBI	0.18	0.29^{*}	−0.20	-0.44^{***}	0.47^{***}	-0.38^{**}	0.06	0.30^{*}	−0.06	0.11	-0.42^{***}	-0.77^{***}	
ITI	−0.23	-0.36^{**}	0.56^{***}	0.46^{***}	-0.66^{***}	0.19	−0.17	-0.40^{**}	0.13	−0.23	0.50^{***}	0.76^{***}	-0.79^{***}

Key: log — log-transformed, sr — square root transformed, log1 — transformed by $\log(X+1)$. Correlations significant at $^{***}p<0.001$, $^{(**)}p<0.01$ and $^{*}p<0.05$. Note: only subtidal locations have been used in the calculation.

Table 4
Multiple regression models taking into account data from all the subtidal sampled sites.

Dependent variable	Abundance log(ind. m^{-2})	Richness (nr. of taxa)	Diversity (ind. bit^{-1})	AMBI	ITI
intercept	4.2703645	−7.524	1.774	4.496	−3.477
De = Depth (square root (meters))		8.402	0.397	−0.0486	9.371
C = Current speed (log(cm s^{-1}))	−1.072			−1.615	6.226
P = Production (tons yr^{-1})	−0.000335	−0.000599	−0.00104	0.000665	−0.0169
Di = Distance to cages (log(1+ m))		3.514	0.534	−0.593	8.332
Multiple *R*-squared	0.226	0.252	0.413	0.451	0.594
p-value	<0.001	<0.01	<0.001	<0.001	<0.001

variance explained by abundance, richness, diversity, AMBI and ITI was 22, 25, 41, 45 and 59%, respectively (Table 4). These regression equations enable us to predict (Figs. 7 and 8) average responses of macrobenthos, in terms of AMBI and ITI, respectively, to different scenarios of farms (production of 100, 800 and 1500 tons yr^{-1}), hydrographical conditions (current speeds of 3, 8, and 14 cm s^{-1}), and water depths (15, 40 and 60 m water depth), at increasing distances from cages. Thus, for example, AMBI values higher than 3.3 (indicating moderate perturbation, see Discussion) are predicted within a radius of *ca.* 120 m from the net cages, when average current speed is 3 cm s^{-1}, depth is 15 m, and annual production is 1500 tons (Fig. 7). In the case of ITI, the above scenario yielded a prediction of degraded situations (ITI values < 30) within a radius of 150 m from the farm (Fig. 8).

The outstanding feature of these analyses is that, when considering parameters for site selection (or site suitability, in the case of existing farms), an increase in water depth in these scenarios seems to play a minor role at a given current velocity; however, an increase in current speed seems to play a much bigger role in reducing the AMBI or increasing the ITI values, and consequently predicting improved benthic conditions (Figs. 7 and 8).

4. Discussion

Benthic indicators have been used extensively in the assessment of aquaculture impacts (Gowen and Bradbury, 1987; Gyllenhammar and Hakanson, 2005; Buschmann et al., 2006; Kalantzi and Karakassis, 2006). When investigating spatial variability of macrobenthos parameters (e.g. abundance, biomass, etc.), in relation to increasing distance from aquaculture farms, a general pattern was found among ECASA sites of increasing abundance, diversity, richness, and ITI, and decreasing biomass and AMBI. However, this pattern was not present at several of the locations (e.g. Abornes, Garrucha, Bisceglie).

The Pearson and Rosenberg (1978) paradigm predicts that benthic species richness or diversity should decrease with an increase in organic enrichment, above a certain threshold level. However, sometimes, as found at some of the ECASA sites (this study) and elsewhere, the responses were different from what would be expected, as described by Edgar et al. (2005), Sanz-Lázaro and Marín (2006), Aguado-Giménez et al. (2007) and Gibbs (2007). In the specific case of mussel farms, recent studies have shown impact gradients when using benthic indices, but when these were compared with results from the reference sites the differences were not significant (Callier et al., 2008). The wide range in benthic responses to aquaculture enrichment has therefore led to the conclusion that, aside from extreme cases, it is not trivial to anticipate how the benthic community will be affected (Karakassis et al., 2000; Pereira et al., 2004; Buschmann et al., 2006; Klaoudatos et al., 2006).

It is generally accepted that the main factors controlling the extent of benthic organic enrichment are: farm size, husbandry methods and hydrographic conditions (Hartstein and Rowden, 2004; Mente et al., 2006; Giles, 2008). However, very few studies have included a quantitative assessment of the relationships of impact parameters with site and farm characteristics, i.e. an assessment of benthic impacts from site and farm parameters (Giles, 2008). Hence, most of the abovementioned studies, which reported contradictory results in farm impacts, have not included hydrodynamics or husbandry practices in their analyses (e.g. Stenton-Dozey et al., 1999; Danovaro et al., 2004; Pereira et al., 2004; Buschmann et al., 2006), or they have included information which was not used in the analysis (e.g. Gowen and Bradbury, 1987; Aguado-Giménez et al., 2007). Although the data in our study were collected from a wide array of environments, encompassing

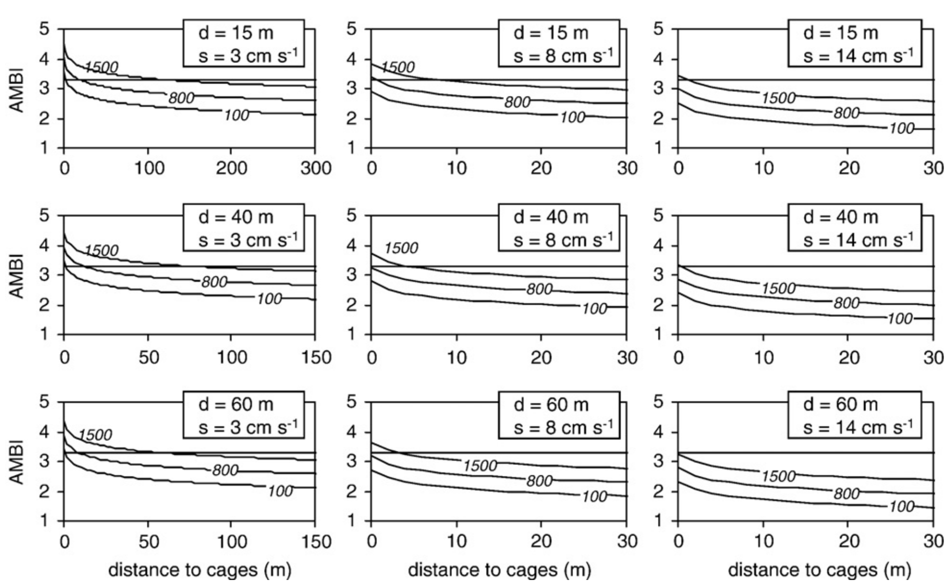

Fig. 7. Spatial variability of AMBI (AZTI's Marine Biotic Index) predicted by the multiple linear regression given in Table 4, for three annual production rates: 100, 800 and 1500 tons yr^{-1} (indicated with numbers in italics), at different combinations of depth and currents. Key: d — depth, s — average current speed.

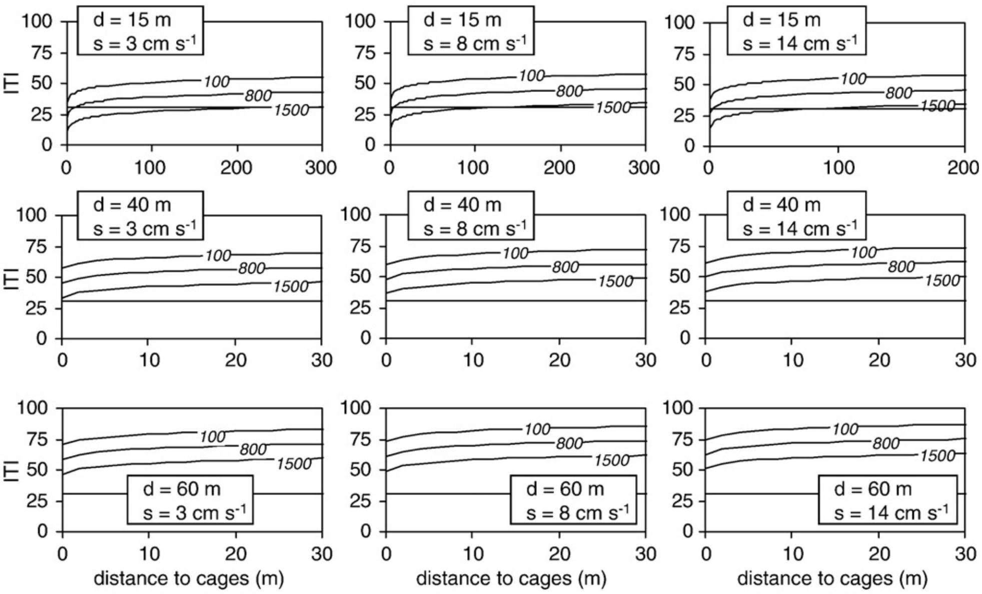

Fig. 8. Spatial variability of ITI (Infaunal Trophic Index) predicted by the multiple linear regression given in Table 4, for three annual production rates: 100, 800 and 1500 tons yr^{-1} (indicated with numbers in italics), at different combinations of depth and currents. Key: d — depth, s — average current speed.

a range of latitudes (from 37 to 69° N) and different farmed species, most of the benthic indices examined were significantly correlated with average current speed, farm production level, and number of years of farm activity. It is interesting to note therefore that, although Kalantzi and Karakassis (2006) observed latitudinal differences in farm impacts, this was not found in our study.

The statistical analyses of our data indicate that the major sources of variance are: total production and years of activity (15.2% of the variability explained), and water depth and current velocity (11.5% of the variability explained), together with the interactions between these factors. It is noteworthy that sediment characteristics, such as redox potential and sediment grain size were considerably less important (5.4% of the variability explained). This pattern may explain why some authors do not find strong relationships between benthic parameters and husbandry practices (see, for example, Miron et al., 2005), as they did not account for the "dynamic characteristics" (hydrodynamics) of the studied sites. This may also be the underlying reason behind the observation that despite the differences among locations the physico-chemical and biological structure of the surface sediments surrounding the farms was generally quite similar (see Chamberlain et al., 2001; Gyllenhammar and Hakanson, 2005; Kutti et al., 2007).

The results of our study are in agreement with some of the recent aquaculture reviews that consider different latitudinal, husbandry and hydrographic environments; although 47% of the variability in macro-faunal structure was not explained by the investigated factors. For example, Kalantzi and Karakassis (2006) reviewed 41 papers on the benthic effects of fish farming, and performed a meta-analysis of 120 variables. They found that most biological and geochemical impacts are determined by a combination of distance from the farm, bathymetry and/or latitude. They concluded that the complex interactions between variables and the lack of data, such as current speed, result in difficulties in setting common or uniform environmental quality standards for benthic effects of fish farming. Moreover, they recommend that environmental quality standards should take into account the differences between geographic regions, depth zones and sediment types. Giles (2008) reviewed 64 publications on fish farm benthic impacts and proposed a Bayesian network for the quantitative assessment of the relationships between impact parameters and site and farm characteristics. Furthermore, Giles showed that benthic impact was a function of fish density, farm volume, food conversion ratio, water depth, current strength and sediment mud content. From our study, and the reviews mentioned here, it seems that benthic communities show relatively general patterns of response to aquaculture pressure. Thus benthic indices are useful in environmental impact studies, provided they are combined with other tools (e.g. stable isotopes studies, as recommended in Sarà (2004)) and essential information (e.g. farm husbandry and hydrography).

These findings shed important light on aquaculture management considerations, particularly with regard to site selection and monitoring schemes for existing farms (Borja, 2002). Essential variables that should be considered when selecting a new location for aquaculture, include water depth and current speed. Borja (2002) recommended the following: (i) 'good' sites have >30 m water depth and current speed >15 cm s^{-1}; (ii) 'moderate' sites have between 15 and 30 m water depth and current speed between 5 and 15 cm s^{-1}; and (iii) 'bad' sites have <15 m water depth and current speed <5 cm s^{-1}. In Figs. 7 and 8, it is clear that the conditions prevailing in Borja's 'bad' sites would produce substantially negative impacts in terms of AMBI and ITI values (level of impact determined also by the total annual production of the farm), whilst conditions prevailing in 'good' sites are unlikely yield "bad" AMBI or ITI impact scores. Thus, the use of some critical boundaries in biotic indices, such as the limit between 'slightly' and 'moderate' disturbance in AMBI (established as a value of 3.3, in Borja et al., 2000) or the limit between 'degraded' and 'changed' in ITI (established as a value of 30, in Word, 1979), allows the prediction of the level of impact when using multiple regression analysis (see Figs. 7 and 8).

This approach can also be used in the assessment and evaluation of cumulative impacts of marine farms (see King and Pushchak, 2008). Current guidelines entail impact assessment on a site-by-site, but this creates an incomplete picture of the range of potential impacts associated with new farms or added production on-site. Gyllenhammar and Hakanson (2005) proposed a general 'rule of thumb' whereby a 50 ton fish farm has a footprint (impacted benthos) the size of a 'football field'; however, Figs. 7 and 8 demonstrate that this rule is rather inaccurate as impacts are a composite result of multiple factors and variables that may also interact among themselves.

Among the key variables that determine aquaculture impacts on the seafloor, water depth and hydrodynamics are positively correlated with dispersion of farm wastes, and inversely correlated with benthic impacts. Nonetheless, Sarà et al. (2006) recently suggested that the relative area of influence of the impacts of fish farms increases proportionally to increasing current velocities. Moreover, they proposed that the distribution of wastes from the cages is likely to be dependent on movements at the bottom of the water column, suggesting that resuspension (described by Cromey et al., 2002b) is a key factor. Similarly, Kutti et al. (2007) found that, at deepwater sites, organic waste affected the benthic community on a much larger spatial scale than at shallow water sites. Thus, the benthos is affected by hydrodynamic conditions that govern the sediments' natural assimilative capacity (Macleod et al., 2007).

Finally, as observed by Miron et al. (2005), the wide range of aquaculture impacts reported in the literature may largely be the result of individual use of ecological indices. Certain diversity indices may be misleading if not complemented with other ecological or statistical tools, as shown in our contribution. Hence, although ITI and AMBI have proven to be fairly reliable at indicating impacts in most of the circumstances examined, we have shown that the consideration of additional factors, such as hydrography and husbandry, increases the reliability of the impact assessment or prediction.

5. Conclusions

Assessment of the response of benthic communities to organic enrichment from aquaculture may be improved by using a suite of benthic indicators (rather than a single indicator), and considering variables that are unique to the studied location, e.g. water depth, hydrodynamics, years of farm activity, and total annual production. Assessments that do not consider these factors could lead to an incorrect interpretation of benthic response. Moreover, these factors should be taken into account when studying cumulative effects of existing farms, and when designing monitoring programmes of aquaculture impacts. Lessons learned from such multi-factorial approaches can also be applied to the process of aquaculture site selection.

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References

Aguado-Giménez, F., Marín, A., Montoya, S., Marín-Guirao, L., Piedecausa, 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.

Apostolaki, E.T., Tsagaraki, T., Tsapakis, M., Karakassis, I., 2007. Fish farming impact on sediments and macrofauna associated with seagrass meadows in the Mediterranean. Estuarine, Coastal and Shelf Science 75, 408–416.

Black, K.D. (Ed.), 2001. Environmental Impacts of Aquaculture. Sheffield Academic Press. 214 pp.

Bocard, D., Legendre, P., Drapeau, P., 1992. Partialling out the spatial component of ecological variation. Ecology 73 (3), 1045–1055.

Borja, A., 2002. Los impactos ambientales de la acuicultura y la sostenibilidad de esta actividad. Boletín del Instituto Español de Oceanografía 18, 41–49.

Borja, A., Muxika, I., 2005. Guidelines for the use of AMBI (AZTI's marine biotic index) in the assessment of the benthic ecological quality. Marine Pollution Bulletin 50, 787–789.

Borja, A., Dauer, D.M., 2008. Assessing the environmental quality status in estuarine and coastal systems: Comparing methodologies and indices. Ecological Indicators 8, 331–337.

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. Marine Pollution Bulletin 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. Marine Pollution Bulletin 46, 835–845.

Borja, A., Bricker, S.B., Dauer, D.M., Demetriades, N.T., Ferreira, J.G., Forbes, A.T., Hutchings, P., Jia, X., Kenchington, R., Marques, J.C., Zhu, C., 2008. Overview of integrative tools and methods in assessing ecological integrity in estuarine and coastal systems worldwide. Marine Pollution Bulletin 56, 1519–1537.

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 Pertuis Charentais (SW France): a multi-index approach. Marine Pollution Bulletin 56, 1898–1912.

Buschmann, A.H., Riquelme, V.A., Hernández-González, M.C., Varela, D., Jiménez, J.E., Henríquez, L.A., Vergara, P.A., Guinez, R., Filun, L., 2006. A review of the impacts of salmonid farming on marine coastal ecosystems in the southeast Pacific. ICES Journal of Marine Science 63, 1338–1345.

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.

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. Marine Pollution Bulletin 52, 1573–1584.

Chamberlain, J., Fernandes, T.F., Read, P., Nickell, T.D., Davies, I.M., 2001. Impacts of biodeposits from suspended mussel (*Mytilus edulis* L.) culture on the surrounding surficial sediments. ICES Journal of Marine Science 58, 411–416.

Cromey, C.J., Nickell, T.D., Black, K.D., 2002a. DEPOMOD-modelling the deposition and biological effects of waste solids from marine cage farms. Aquaculture 214 (1-4), 211–239.

Cromey, C.J., Nickell, T.D., Black, K.D., Provost, P.G., Griffiths, C.R., 2002b. Validation of a fish farm waste resuspension model by use of a particulate tracer discharged from a point source in a coastal environment. Estuaries 25, 916–929.

Danovaro, R., Gambi, C., Luna, G.M., Mirto, S., 2004. Sustainable impact of mussel farming in the Adriatic Sea (Mediterranean Sea): evidence from biochemical, microbial and meiofaunal indicators. Marine Pollution Bulletin 49, 325–333.

Edgar, G.J., Macleod, C.K., Mawbey, R.B., Shields, D., 2005. Broad-scale effects of marine salmonid aquaculture on macrobenthos and the sediment environment in south-eastern Tasmania. Journal of Experimental Marine Biology and Ecology 327, 70–90.

Eleftheriou, A., McIntyre, A., 2005. Methods for the study of marine benthos, 3d ed., Wiley-Blackwell Publishing, Oxford, 418 p.

FAO, 2007. The state of world fisheries and aquaculture 2006. FAO Fisheries and Aquaculture Department; Food and Agriculture Organization of the United Nations, Rome. 180 pp. http://www.fao.org/docrep/009/a0699e/A0699E00.HTM.

Fernandes, T.F., Eleftheriou, A., Ackefors, H., Eleftheriou, M., Ervik, A., Sanchez-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. Journal of Applied Ichthyology 17 (4), 181–193.

Ferreira, J.G., Hawkins, A.J.S., Monteiro, P., Moore, H., Service, M., Pascoe, P.L., Ramos, L., Sequeira, A., 2008. Integrated assessment of ecosystem-scale carrying capacity in shellfish growing areas. Aquaculture 275, 138–151.

Fox, J., 2005. The R commander: a basic-statistics graphical user interface to R. Journal of Statistical Software 14 (9), 42.

Garcia, S.M., Zerbi, A., Aliaume, C., Do Chi, T., Lasserre, G., 2003. The ecosystem approach to fisheries. Issues, terminology, principles, institutional foundations, implementation and outlook. FAO Fisheries Technical Paper. No. 443. FAO, Rome. 71 pp.

Gibbs, M.T., 2007. Sustainability performance indicators for suspended bivalve aquaculture activities. Ecological Indicators 7, 94–107.

Giles, H., 2008. Using Bayesian networks to examine consistent trends in fish farm benthic impact studies. Aquaculture 274, 181–195.

Gowen, R.J., Bradbury, N.B., 1987. The ecological impact of salmonid farming in coastal waters: a review. Oceanography and Marine Biology: An Annual Review 25, 563–575.

Gyllenhammar, A., Hakanson, L., 2005. Environmental consequence analyses of fish farm emissions related to different scales and exemplified by data from the Baltic — a review. Marine Environmental Research 60, 211–243.

Hargrave, B.T.E. (Ed.), 2004. Environmental Studies for Sustainable Aquaculture (ESSA), 2004 Symposium Report. Canadian Technical Reports of Fishery Aquatic Science, vol. 2542. vi + 81 pp.

Hartstein, N.D., Rowden, A.A., 2004. Effect of biodeposits from mussel culture on macroinvertebrate assemblages at sites of different hydrodynamic regime. Marine Environmental Research 57, 339–357.

Holmer, M., Argyrou, M., Dalsgaard, T., Danovaro, R., Diaz-Almela, E., Duarte, C.M., Frederiksen, M., Grau, A., Karakassis, I., Marbà, N., Mirto, S., Pérez, M., Pusceddu, A., Tsapakis, M., 2008. Effects of fish farm waste on Posidonia oceanica meadows: Synthesis and provision of monitoring and management tools. Marine Pollution Bulletin 56, 1618–1629.

Jones, J., 2002. Development of indicators on the quality of the marine environment. Proceedings of the Seabed disturbance indicators workshop, Five Lakes, Tolleshunt Knights, DEFRA & CEFAS. 72 pp.

Kalantzi, I., Karakassis, I., 2006. Benthic impacts of fish farming: meta-analysis of community and geochemical data. Marine Pollution Bulletin 52, 484–493.

Karakassis, I., Tsapakis, M., Hatziyanni, E., Papadopoulou, K.N., Plaiti, W., 2000. Impact of cage farming of fish on the seabed in three Mediterranean coastal areas. ICES Journal of Marine Science 57, 1462–1471.

King, S.C., Pushchak, R., 2008. Incorporating cumulative effects into environmental assessments of mariculture: limitations and failures of current siting methods. Environmental Impact Assessment Review 28, 572–586.

Klaoudatos, S.D., Klaoudatos, D.S., Smith, J., Bogdanos, K., Papageorgiou, E., 2006. Assessment of site specific benthic impact of floating cage farming in the eastern Hios island, Eastern Aegean Sea, Greece. Journal of Experimental Marine Biology and Ecology 338, 96–111.

Kutti, T., Hansen, P.K., Ervik, A., Hoisaeter, T., Johannessen, P., 2007. Effects of organic effluents from a salmon farm on a fjord system. II. Temporal and spatial patterns in infauna community composition. Aquaculture 262, 355–366.

Langmuir, D., 1971. Eh-pH determination. In: Carver, R.E. (Ed.), Sedimentary petrology. John Wiley & Sons, New York, pp. 597–634.

Legendre, P., Bocard, D., Peres-Neto, P.R., 2005. Analyzing beta diversity: Partitioning the spatial variation of community composition data. Ecological Monographs 75 (4), 435–450.

Macleod, C.K., Moltschaniwskyj, N.A., Crawford, C.M., Forbes, S.E., 2007. Biological recovery from organic enrichment: Some systems cope better than others. Marine Ecology. Progress Series 342, 41–53.
Maurer, D., Nguyen, H., Robertson, G., Gerlinger, T., 1999. The Infaunal Trophic Index (ITI): its suitability for marine environmental monitoring. Ecological Applications 9 (2), 699–713.
McKindsey, C.W., Thetmeyer, H., Landry, T., Silvert, W., 2006. Review of recent carrying capacity models for bivalve culture and recommendations for research and management. Aquaculture 261, 451–462.
Mente, E., Pierce, G.J., Santos, M.B., Neofitou, C., 2006. Effect of feed and feeding in the culture of salmonids on the marine aquatic environment: a synthesis for European aquaculture. Aquaculture International 14, 499–522.
Michler-Cieluch, T., Kodeih, S., 2008. Mussel and seaweed cultivation in offshore wind farms: an opinion survey. Coastal Management 36, 392–411.
Miron, G., Landry, T., Archambault, P., Frenette, B., 2005. Effects of mussel culture husbandry practices on various benthic characteristics. Aquaculture 250, 138–154.
Muxika, I., Borja, A., Bonne, W., 2005. The suitability of the marine biotic index (AMBI) to new impact sources along European coasts. Ecological Indicators 5, 19–31.
Pacyna, J.M., Barrett, K., Namiesnik, J., 2005. The EU European contribution to global coastal zone research: an ELOISE (European Land-Ocean Interaction Studies) project. Estuarine, Coastal and Shelf Science 62, 387–389.
Pearson, T., Rosenberg, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. Oceanography and Marine Biology Annual Review 16, 229–311.
Pereira, P.M.F., Black, K.D., McLusky, D.S., Nickell, T.D., 2004. Recovery of sediments after cessation of marine fish farm production. Aquaculture 235, 315–330.
Pergent-Martini, C., Boudouresque, C.F., Pasqualini, V., Pergent, G., 2006. Impact of fish farming facilities on Posidonia oceanica meadows: a review. Marine Ecology 27, 310–319.
Sanz-Lázaro, C., Marín, A., 2006. Benthic recovery during open sea fish farming abatement in Western Mediterranean, Spain. Marine Environmental Research 62, 374–387.
Sarà, G., 2004. Effects of fish fanning waste to sedimentary and particulate organic matter in a southern Mediterranean area (Gulf of Castellammare, Sicily): a multiple stable isotope study (delta C-13 and delta N-15). Aquaculture 234, 199–213.
Sarà, G., Scilipoti, D., Milazzo, M., Modica, A., 2006. Use of stable isotopes to investigate dispersal of waste from fish farms as a function of hydrodynamics. Marine Ecology. Progress Series 313, 261–270.
Sequeira, A., Ferreira, J.G., Hawkins, A.J.S., Nobre, A., Zhang, X.L., Yan, X., Nickell, T., 2008. Trade-offs between shellfish aquaculture and benthic biodiversity: a modelling approach for sustainable management. Aquaculture 274, 313–328.
Stenton-Dozey, J.M.E., Jackson, L.F., Busby, A.J., 1999. Impact of mussel culture on macrobenthic community structure in Saldanha Bay, South Africa. Marine Pollution Bulletin 39, 357–366.
Stigebrandt, A., Aure, J., Ervik, A., Hansen, P.K., 2004. Regulating the local environmental impact of intensive marine fish farming III. A model for estimation of the holding capacity in the Modelling – Ongrowing fish farms – Monitoring system. Aquaculture 234, 239–261.
ter Braak, C.J.F., Smilauer, P., 2002. Canoco reference manual and CanoDraw for Windows user's guide: software for canonical community ordination (version 4.5). Microcomputer Power, Ithaca, New York. 252 pp.
Word, J.Q., 1979. The infaunal trophic index. Souththern Californian Coastal Water Research Project Annual Report, El Segundo, California, pp. 19–39.