



# Comparison of the performances of two biotic indices based on the MacroBen database

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**ABSTRACT:** The pan-European MacroBen database was used to compare the AZTI Marine Biotic Index (AMBI) and the Benthic Quality Index (BQI<sub>ES</sub>), 2 biotic indices which rely on 2 distinct assessments of species sensitivity/tolerance (i.e. AMBI EG and BQI E[S<sub>50</sub>]<sub>0.05</sub>) and which up to now have only been compared on restricted data sets. A total of 12 409 stations were selected from the database. This subset (indicator database) was later divided into 4 marine and 1 estuarine subareas. We computed E(S<sub>50</sub>)<sub>0.05</sub> in 643 taxa, which accounted for 91.8% of the total abundances in the whole marine indicator database. AMBI EG and E(S<sub>50</sub>)<sub>0.05</sub> correlated poorly. Marked heterogeneities in E(S<sub>50</sub>)<sub>0.05</sub> between the marine and estuarine North Sea and between the 4 marine subareas suggest that sensitivity/tolerance levels vary among geographical areas. High values of AMBI were always associated with low values of BQI<sub>ES</sub>, which underlines the coherence of these 2 indices in identifying stations with a bad ecological status (ES). Conversely, low values of AMBI were sometimes associated with low values of BQI<sub>ES</sub> resulting in the attribution of a good ES by AMBI and a bad ES by BQI<sub>ES</sub>. This was caused by the dominance of species classified as sensitive by AMBI and tolerant by BQI<sub>ES</sub>. Some of these species are known to be sensitive to natural disturbance, which highlights the tendency of BQI<sub>ES</sub> to automatically classify dominant species as tolerant. Both indices thus present weaknesses in their way of assessing sensitivity/tolerance levels (i.e. existence of a single sensitivity/tolerance list for AMBI and the tight relationship between dominance and tolerance for BQI<sub>ES</sub>). Future studies should focus on the (1) clarification of the sensitivity/tolerance levels of the species identified as problematic, and (2) assessment of the relationships between AMBI EG and E(S<sub>50</sub>)<sub>0.05</sub> within and between combinations of geographical areas and habitats.

**KEY WORDS:** AZTI Marine Biotic Index · Benthic Quality Index · Macrozoobenthos · Water framework directive

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## INTRODUCTION

The European Water Framework Directive (WFD) establishes a basis for the protection of ground, continental, transitional and coastal waters. It aims at achieving a good ecological status (ES) for all European water bodies by 2015. The first step consists of assessing the current ES of these water bodies, which

is based on a large variety of hydromorphological, physicochemical and biological parameters. In order to unravel natural and man-induced changes, ES values are derived from ecological quality ratios (EQR), which correspond to the ratio of the value of the considered parameter at each sampled station divided by the value of the same parameter at a reference (i.e. non-impacted) station (Wallin et al. 2003).

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Macrozoobenthos is one of the biological compartments considered by the WFD (Borja et al. 2004a, Borja 2005) and a large variety of biotic indices use its composition to infer ES (Grall & Glémarec 1997, Borja et al. 2000, Gomez Gesteira & Dauvin 2000, Rosenberg et al. 2004). In spite of their diversity, most of these indices are based on the same paradigm: disturbances are generating secondary successions during which tolerant species are at first dominant and then progressively replaced by sensitive species (Pearson & Rosenberg 1978). There is, thus, more need for testing and unifying the existing benthic biotic indices than for producing new ones (Diaz et al. 2004). Two of the main indices introduced in view of the implementation of the WFD are (1) the AZTI Marine Biotic Index (AMBI; Borja et al. 2000), and (2) the Benthic Quality Index (BQI; Rosenberg et al. 2004). Although these 2 indices rely on the same concept, they differ in (1) their ways of assessing species sensitivity/tolerance levels, (2) the consideration of species richness, and (3) the procedures used to convert computed indices of ES.

In AMBI, sensitivity/tolerance levels are assessed based on the compilation of expert knowledge and its translation into ecological groups (AMBI EG). This results in a single sensitivity/tolerance per species that is used for all data sets irrespective of geographic location (Borja et al. 2000, Borja et al. 2003, Salas et al. 2004, Muxika et al. 2005). Conversely, for BQI, Rosenberg et al. (2004) assume that species sensitivity/tolerance levels vary according to geographical location. The assessment of sensitivity/tolerance within BQI is based on the concept of  $E(S_{50})_{0.05}$  (see 'Data and methods' for definition) (Rosenberg et al. 2004). The availability of  $E(S_{50})_{0.05}$  constitutes a severe limitation to the computation of BQI, which is either restricted to large data sets (Rosenberg et al. 2004, Labruno et al. 2006, Dauvin et al. 2007, Zettler et al. 2007) or to areas where a list of  $E(S_{50})_{0.05}$  is available (Reiss & Kröncke 2005).

The computation of AMBI is based on the sole sensitivity/tolerance concept (Borja et al. 2000), which makes it largely sampling effort-independent (Fleischer et al. 2007, Muxika et al. 2007b). Conversely, BQI also takes into account species richness ( $S$ ) through a  $\log(S + 1)$  term (Rosenberg et al. 2004), which makes it sampling effort-dependent when computed on lumped data (Fleischer et al. 2007) and/or on individual samples collected with different gears. This constitutes another restriction to its use since large databases are (1) often constituted of several surveys with different sampling strategies (see Table 1 for the present study), and (2) often comprised of a significant proportion of lumped data (i.e. 96.3% of all stations during the present study). Fleischer et al. (2007) proposed to overcome this difficulty by replacing  $\log(S + 1)$  by  $\log(E[S_{50}] + 1)$  and proved that the so-modified BQI (i.e.  $BQI_{ES}$ ) is indepen-

dent of sampling effort and correlates tightly with BQI.

AMBI uses a single scale to infer ES (Borja et al. 2004a), whereas BQI assumes that for each habitat the station with the highest BQI constitutes a valid reference for the computation of EQR. The stations with an EQR higher than 0.6 are then considered to at least be in a good ES (Rosenberg et al. 2004).

Multivariate AMBI (M-AMBI) was recently introduced as a refinement of AMBI (Borja et al. 2004b, Borja et al. 2007, Muxika et al. 2007a). Its computation involves a factorial correspondence analysis (FCA) based on AMBI, species richness and the Shannon-Wiener diversity index,  $H'$ . FCAs are carried out for each habitat and 2 bad and good reference stations are included. The coordinates of the projection of the stations along the axis linking the bad and good reference stations in the first plane of the FCA constitute EQR, which are transformed into ES using an appropriate conversion scale (Wallin et al. 2003). M-AMBI is much more similar to BQI than AMBI since it accounts for species richness and uses several scales to infer ES. BQI and M-AMBI, however, still largely differ in their assessments of species sensitivity/tolerance.

Both AMBI and BQI were initially proposed and tested based on individual data sets (Borja et al. 2000, Rosenberg et al. 2004). AMBI has, since then, been tested on a large variety of other (but still mostly individual) data sets (Borja et al. 2000, 2003, Salas et al. 2004, Marin-Guirao et al. 2005, Muniz et al. 2005, Muxika et al. 2005, Bigot et al. 2008, Blanchet et al. 2008), BQI has been tested on a much smaller number of datasets due to the difficulty in computing  $E(S_{50})_{0.05}$ . AMBI and BQI have recently been compared in the North Sea (Reiss & Kröncke 2005), the Gulf of Lions (Labruno et al. 2006), the Seine estuary (Dauvin et al. 2007) and the Baltic Sea (Zettler et al. 2007). All comparisons have shown major discrepancies but have largely ignored their potential causes. The adequacy of the use of a single sensitivity/tolerance list by AMBI as opposed to BQI is, for example, yet to be tested partly due to the lack of any comprehensive database at the pan-European level. The Network of Excellence Marine Biodiversity and Ecosystem Functioning (MarBEF) has recently filled this gap for soft-bottom macrozoobenthos by creating the MacroBen database. The aim of the present study is to use this new tool to (1) promote the use of  $BQI_{ES}$  by providing lists of  $E(S_{50})_{0.05}$  both at the pan-European level and within distinct geographic subareas, (2) compare AMBI EG and  $E(S_{50})_{0.05}$ , (3) assess the validity of the use of a single list of sensitivity/tolerance levels by comparing  $E(S_{50})_{0.05}$  between subareas, (4) assess the relationships between AMBI and  $BQI_{ES}$  and (5) compare the ES assessments derived from AMBI and  $BQI_{ES}$ .

## DATA AND METHODS

**MacroBen database.** The main characteristics of MacroBen are described in Vanden Berghe et al. (2009, this Theme Section) and will not be repeated here. The filtering procedure used during the present study consisted of selecting (1) quantitative data, (2) adult animal taxa, (3) organisms identified to the species level, (4) non-colonial organisms and (5) samples collected after 1980. Baltic Sea samples were excluded because an extensive comparison between AMBI and BQI has recently been carried out in this area (Zettler et al. 2007), and Black Sea samples were excluded because they were too few. The data set was further reduced by considering only the most recent sampling date for each station. This reduced indicator database was com-

posed of 29 individual data sets and contained a total of 12 409 stations (Fig. 1, Table 1). It was later divided into 4 subareas based on the Large Marine Ecosystem classification ([www.edc.uri.edu/lme/intro.htm](http://www.edc.uri.edu/lme/intro.htm)), namely: (1) the Celtic-Biscay Shelf (115 stations), (2) the Mediterranean (426 stations), (3) the North Sea (11 664 stations), and (4) the Norwegian and Barents Seas (204 stations). Because of the importance of the ni data set (10 251 stations), North Sea data were divided in an estuarine (i.e. ni) and a marine (1413 stations) data set. The ranges of  $E(S_{50})$  (see 'Data and methods—Computation of AMBI and BQI<sub>ES</sub>' for definitions) in each marine subarea were: 1.95 to 33.53, 2.86 to 34.61, 1.35 to 39.59 and 1.00 to 33.19 in the Celtic-Biscay Shelf, the Norwegian and Barents Seas, the Mediterranean and the marine North Sea, respectively.

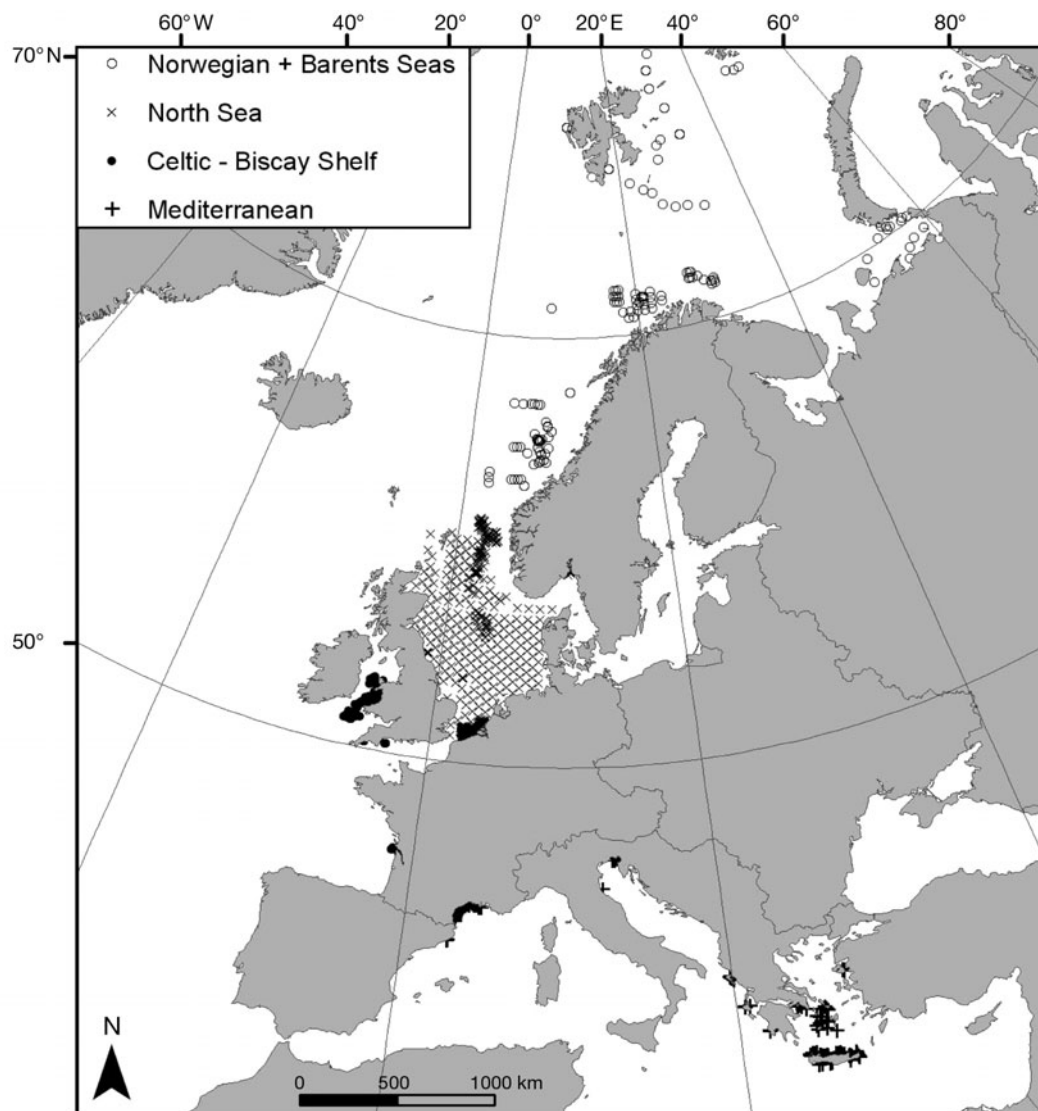


Fig. 1. Location of the stations in the indicator database delimiting of the 4 geographical marine subareas considered during the present study

Table 1. Composition of the indicator data set with information regarding the location and the number of stations in the 4 subareas and in each individual data set. Depth range, sampling gear, sample replication and total sampled area is also provided for each individual data set

Data set and subarea	Location	Depth range (m)	No. stations	Sample gear	No. replicates	Total sampled area (m <sup>2</sup> )
<b>Norwegian and Barents Seas</b>						
ar	Svalbard	75–335	22	Box corer	1	0.1
hs	Hornsund	25–203	34	Van Veen grab	1	0.1
ko	Kongsfjorden/Spitsbergen	5–30	6	Box corer	1	–
o2	Northern Barents Sea	–	10	Van Veen grab	4–5	0.4–0.5
o4 <sub>NB</sub>	Norwegian Sea	71–1520	55	Van Veen grab	1–5	0.1–0.5
o6	Finmark	160–374	53	Van Veen grab	5	0.5
o7	Pechoran Sea	7–207	15	Van Veen grab	3–5	0.3–0.5
o8	Franz Josef Land	52–312	9	Van Veen grab	5	0.5
<b>North Sea</b>						
ni	Dutch Delta area	0–57	10251	–	1	–
np <sub>NS</sub>	North Sea	35–70	20	–	1	0.1
ns	Belgian part of the North Sea	0–150	231	Van Veen grab	–	–
o3	Staffjord, Oseberg, Ekosfisk	65–91	30	Van Veen grab	1–5	0.1–0.5
o4 <sub>NS</sub>	Norwegian coast	71–1520	128	Van Veen grab	–	–
of	Oslo Fjord	19–356	57	Van Veen grab	1	0.1
ug	North Sea	0–40	947	–	3	0.09–0.27
<b>Celtic-Biscay Shelf</b>						
np <sub>CS</sub>	English Channel, Irish Sea	50–96	20	–	1	0.1
o5	Southern Irish Sea	7–130	51	Van Veen grab	1	–
pl	Plymouth Sound	15	44	SCUBA diving	1	0.008
<b>Mediterranean</b>						
bl	Bay of Blanes	–	2	Van Veen grab	5	0.3
do	Continental Cretan Shelf	10–60	56	Smith McIntyre grab	1	0.1
gr	Gulf of Lions	10–50	92	Van Veen grab	2–4	0.2–0.4
ka	Cretan Shelf	10–190	199	–	–	0.1
lm	Gulf of Trieste, Adriatic	4–25	28	Van Veen grab	1	–
M0	Gialova Lagoon, Ionian Sea	–	7	Van Veen grab	5	0.25
M2	Gulf of Geras, Aegean Sea	–	9	Ponar grab	1	0.045
M3	Saronikos Gulf	–	6	Ponar grab	2–5	0.1–0.25
M7	Kerkyra, Ionian Sea	–	12	Van Veen grab	1	0.2
M8	Kyklades, Aegean Sea	–	14	Smith McIntyre grab	3–5	0.3–0.5
oc	Northern Adriatic	12	1	Van Veen grab	1	0.06

**Computation of AMBI and BQI<sub>ES</sub>.** AMBI was computed as:

$$\text{AMBI} = [(0 \times \% \text{GI}) + (1.5 \times \% \text{GII}) + (3 \times \% \text{GIII}) + (4.5 \times \% \text{GIV}) + (6 \times \% \text{GV})] / 100 \quad (1)$$

where %GI is the relative abundance of disturbance-sensitive species, %GII is the relative abundance of disturbance-indifferent species, %GIII is the relative abundance of disturbance-tolerant species, %GIV is the relative abundance of second-order opportunistic species and %GV is the relative abundance of first-order opportunistic species (Borja et al. 2000). AMBI was computed as recommended by Borja & Muxika (2005) using a specific function implemented in MacroBen and based on the species reference list available at www.azti.es in July 2006. We used a single fixed scale to infer ES from AMBI (Borja et al. 2004a).

$E(S_{50})_{0.05}$  is defined as the  $E(S_{50})$  (Hurlbert 1971) corresponding to the 5 lowest percentiles of the total

abundance of the considered species within the studied area (Rosenberg et al. 2004).  $E(S_{50})_{0.05}$  values were computed for the whole marine indicator data set and each subarea.

BQI<sub>ES</sub> was then computed as:

$$\text{BQI}_{\text{ES}} = \left\{ \sum_{i=1}^s \left[ \frac{A_i}{A_{\text{Tot}}} \times E(S_{50})_{0.05i} \right] \right\} \times \log_{10} [E(S_{50}) + 1] \quad (2)$$

where  $A_i$  is the abundance of the  $i$ th species at the considered station,  $E(S_{50})_{0.05i}$  is the  $E(S_{50})_{0.05}$  of species  $i$  in the considered subarea,  $A_{\text{Tot}}$  is the total abundance of the individuals belonging to the species for which  $E(S_{50})_{0.05}$  can be computed and  $E(S_{50})$  is the expected number of species in a sample of 50 individuals taken at the considered station (Fleischer et al. 2007).  $E(S_{50})_{0.05}$  and BQI<sub>ES</sub> were computed on lumped data using a specific function implemented in MacroBen.  $E(S_{50})_{0.05}$  values were not computed for species present at less than 20 stations. We used several conversion

scales to infer ES from  $BQI_{ES}$ . Homogeneous habitats were defined based on multi-dimensional scaling and cluster analyses of macrozoobenthos composition carried out on the whole subarea data set (Celtic-Biscay Shelf and Norwegian and Barents Seas) or on each major individual data set (i.e. ka, gr and do, see Table 1) in the Mediterranean and the North Sea. The highest value of  $BQI_{ES}$  in each homogeneous habitat was used to compute an EQR. Each scale was then obtained by dividing these maximal values into 5 equal classes (Rosenberg et al. 2004).

## RESULTS

### Computation of $E(S_{50})_{0.05}$ between subareas and with AMBI EG

We computed the  $E(S_{50})_{0.05}$  of 76 species in the Celtic-Biscay Shelf, 246 in the Mediterranean, 165 in the Norwegian and Barents Seas, 337 in the marine North Sea and 158 in the estuarine North Sea. The corresponding lists are available at: [www.marbef.org/documents/data/theme1/es50\\_005.xls](http://www.marbef.org/documents/data/theme1/es50_005.xls). The proportions of species and/or individuals — which are attributed sensitivity/tolerance levels, essential for a sound assessment of ES using either AMBI and  $BQI_{ES}$  — with an  $E(S_{50})_{0.05}$  were between 16.0 (Celtic-Biscay Shelf) and 54.7% (estuarine North Sea), much lower than for AMBI EG (91.8 and 92.4%, respectively) (Fig. 2A). Differences between the 2 indices were lower when considering the number of individuals. The proportions of individuals with an  $E(S_{50})_{0.05}$  were between 69.9% (Norwegian and Barents Seas) and 99.8% (estuarine North Sea), which were still lower than for AMBI EG (88.7 and 99.9%, respectively) (Fig. 2B). When considering the marine indicator data set as a whole, 643 species (46.7%) corresponding to 91.8% of individuals were attributed an  $E(S_{50})_{0.05}$  (versus 97.1% of individuals for AMBI EG).

*Dipolydora quadrilobata*, *Microdeutopus gryllotalpa*, *Boccardiella ligERICA*, *Streblospio shrubsolii*, *Spio armata*, *Corophium volutator* and *Hydrobia ulvae* were the most dominant (rank < 97) species in the marine indicator data set lacking an  $E(S_{50})_{0.05}$  (Table 2). *Dacrydium vitreum*, *Potamides conicus*, *Eudorellopsis deformis*, *Micronephthys maryae* and *Crenella decussata* were the most dominant (rank < 141) species in the marine indicator data set lacking an AMBI EG (Table 2).

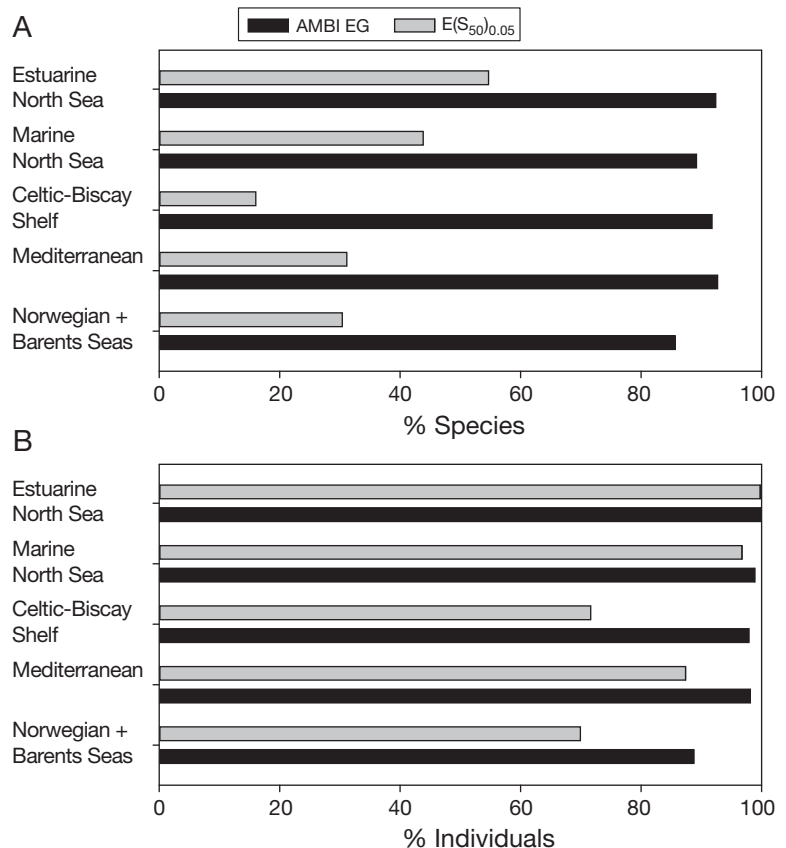


Fig. 2. Proportions of the number of (A) species and (B) individuals with an AMBI EG or an  $E(S_{50})_{0.05}$  value in the different subareas

Table 2. Most dominant (ranks based on decreasing abundances) species in the whole marine indicator data set which are still lacking an  $E(S_{50})_{0.05}$  and/or an AMBI EG value

Species	Rank	$E(S_{50})_{0.05}$	AMBI EG
<i>Dipolydora quadrilobata</i>	16	–	IV
<i>Microdeutopus gryllotalpa</i>	33	–	III
<i>Boccardiella ligERICA</i>	39	–	III
<i>Streblospio shrubsolii</i>	43	–	III
<i>Spio armata</i>	56	–	III
<i>Dacrydium vitreum</i>	67	9.82	–
<i>Corophium volutator</i>	91	–	III
<i>Hydrobia ulvae</i>	96	–	III
<i>Langerhansia heterochaeta</i>	102	–	II
<i>Potamides conicus</i>	122	–	–
<i>Eudorellopsis deformis</i>	127	12.27	–
<i>Micronephthys maryae</i>	139	13.25	–
<i>Crenella decussata</i>	140	–	–
<i>Aricidea fragilis mediterranea</i>	163	–	I
<i>Microphthalmus similis</i>	167	–	II
<i>Malacoceros fuliginosus</i>	169	–	V
<i>Ophelina abranchiata</i>	173	17.88	–
<i>Pectinaria belgica</i>	179	–	I
<i>Dendrodoa grossularia</i>	180	–	I
<i>Axinopsida orbiculata</i>	184	–	–
<i>Octobranchus floriceps</i>	195	23.43	–



$E(S_{50})_{0.05}$  values were between 1.00 and 10.48, 1.96 and 24.14, 5.64 and 25.77, 1.35 and 28.36, and 2.86 and 27.85 in the estuarine North Sea, marine North Sea, Celtic-Biscay Shelf, Mediterranean and Norwegian and Barents Seas, respectively. When considering the whole marine indicator data set, there was a significant

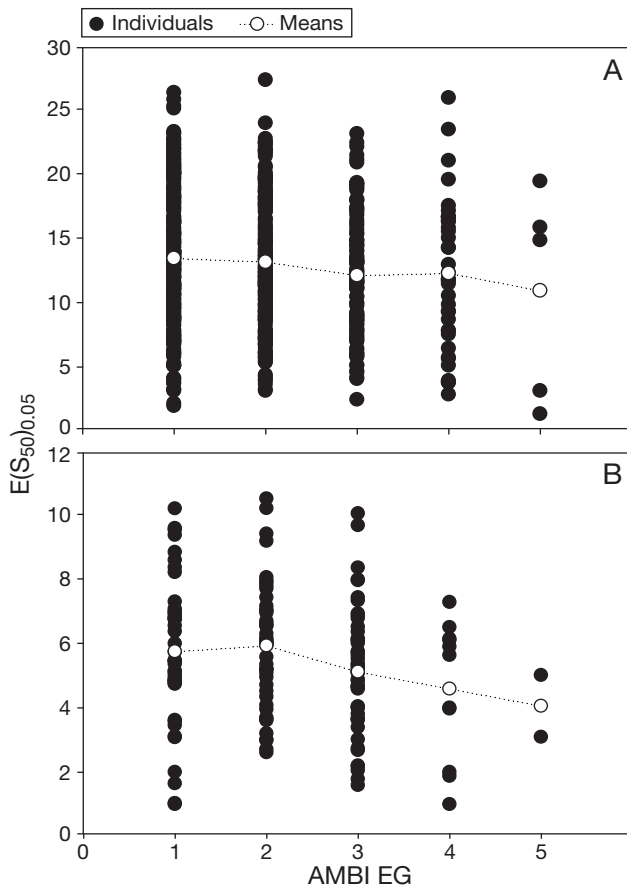


Fig. 3. Relationship between AMBI EG and  $E(S_{50})_{0.05}$  in the (A) whole marine indicator data set and (B) estuarine North Sea. Closed symbols refer to individual stations and open symbols to the mean value of  $E(S_{50})_{0.05}$  for each AMBI EG

negative correlation between AMBI EG and  $E(S_{50})_{0.05}$  (Fig. 3, Table 3), even though the explicative power of the corresponding linear regression model was low. There were significant (but still weak) negative correlations between these 2 parameters in the marine and estuarine North Sea and in the Norwegian and Barents

Table 3. Main characteristics of the simple linear regression models linking AMBI EG and  $E(S_{50})_{0.05}$  in the whole marine data set and within each subarea. Significant ( $p < 0.05$ ) negative correlations are in bold

Subarea	N	r	p	Intercept	Slope
Marine indicator data set	669	<b>-0.150</b>	<b>&lt;0.0001</b>	14.86	<b>-1.32</b>
Celtic-Biscay Shelf	75	0.022	0.848	-	-
Mediterranean	240	0.037	0.572	-	-
Marine North Sea	95	<b>-0.324</b>	<b>0.001</b>	17.82	<b>-1.64</b>
Norwegian and Barents Seas	143	<b>-0.217</b>	<b>0.009</b>	19.75	<b>-1.38</b>
Estuarine North Sea	152	<b>-0.185</b>	<b>0.023</b>	6.350	<b>-0.385</b>

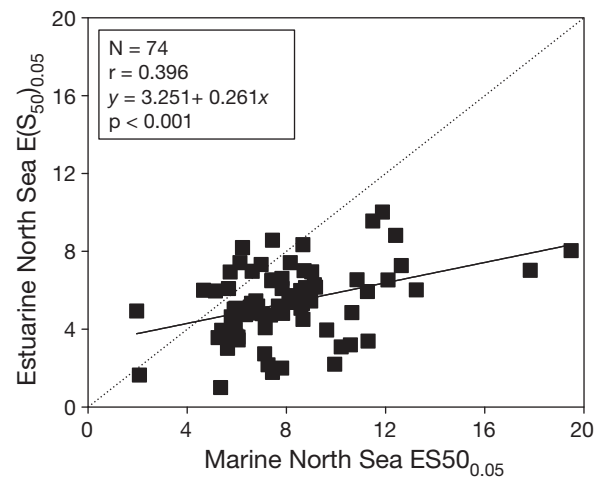


Fig. 4. Relationship between  $E(S_{50})_{0.05}$  in the marine and estuarine North Sea. Solid line: linear regression; dotted line:  $y = x$

Table 4.  $E(S_{50})_{0.05}$  of the 11 species for which they could be computed in all 4 marine subareas. VC: variation coefficient computed for the 4 marine subareas

Species	Celtic-Biscay Shelf	Mediterranean	Marine North Sea	Norwegian and Barents Seas	Marine indicator data set	VC (%)
<i>Heteromastus filiformis</i>	5.64	2.56	7.81	16.81	16.81	74.6
<i>Goniada maculata</i>	9.21	18.98	11.16	22.34	22.34	40.5
<i>Scoloplos armiger</i>	9.34	18.51	7.26	11.24	11.24	42.2
<i>Myriochele oculata</i>	11.90	7.16	6.95	13.99	13.99	35.1
<i>Owenia fusiformis</i>	10.36	6.18	13.24	9.82	9.82	29.3
<i>Aricidea catherinae</i>	17.49	18.56	17.10	15.71	15.71	6.8
<i>Paradoneis lyra</i>	17.54	18.93	18.28	19.43	19.43	4.4
<i>Scalibregma inflatum</i>	9.34	21.78	11.88	9.94	9.94	43.8
<i>Prionospio cirrifera</i>	17.99	10.55	13.28	12.01	12.01	23.9
<i>Spiophanes kroyeri</i>	17.50	18.13	12.06	16.09	16.09	17.1
<i>Terebellides stroemii</i>	16.73	19.46	17.81	9.82	9.82	26.6

Seas (Table 3). This correlation was not significant in the Mediterranean or in the Celtic-Biscay Shelf, where AMBI was initially developed.

There was a weak but significant positive correlation between  $E(S_{50})_{0.05}$  in the marine and estuarine North Sea (Fig. 4). However,  $E(S_{50})_{0.05}$  tended to be lower in the estuarine than in marine North Sea (Wilcoxon signed-rank test,  $p < 0.001$ ). There were only 11 species for which we were able to compute  $E(S_{50})_{0.05}$  in all

4 marine subareas (Table 4). Overall there were marked changes in  $E(S_{50})_{0.05}$  between subareas as indicated by variation coefficients between 4.4% (*Paradoxeis lyra*) and 74.6% (*Heteromastus filiformis*). When comparing the  $E(S_{50})_{0.05}$  of species occurring in any combination of 2 subareas, we found significant positive correlations between the marine North Sea and both the Celtic-Biscay Shelf and the Norwegian and Barents Seas (Fig. 5). Here again, the explicative pow-

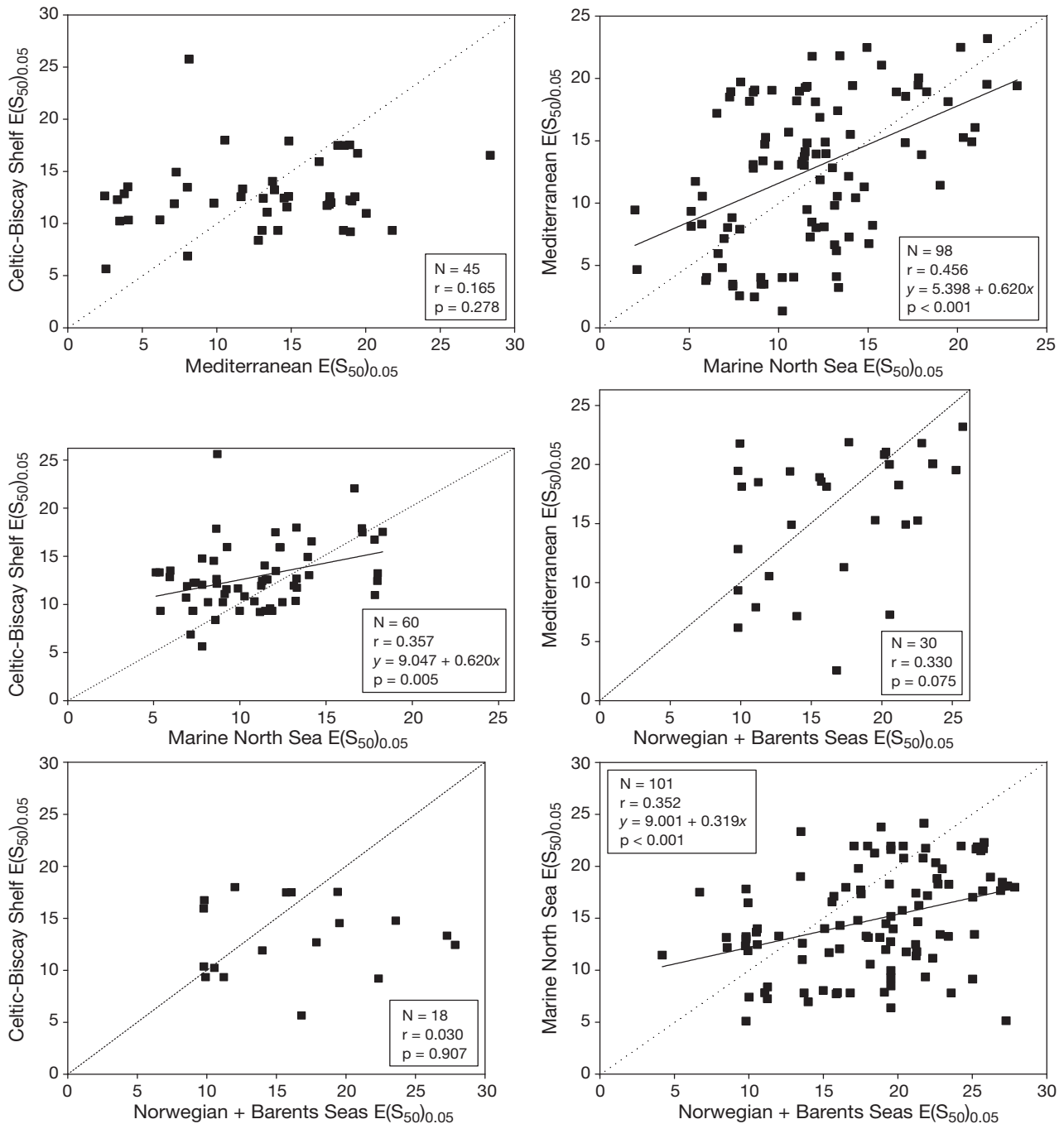


Fig. 5. Relationships between  $E(S_{50})_{0.05}$  in the 4 marine subareas. Solid line: linear regression, dotted line:  $y = x$

Table 5. Significance of the Wilcoxon signed-rank tests used to compare the  $E(S_{50})_{0.05}$  computed within different marine subareas. N: number of species for which  $E(S_{50})_{0.05}$  could be computed in the 2 considered subareas. Significant ( $p < 0.05$ ) differences are in bold

	Celtic-Biscay Shelf		Mediterranean		Marine North Sea	
	p	N	p	N	p	N
Celtic-Biscay Shelf	–					
Mediterranean	0.505	45	–			
Marine North Sea	<b>&lt;0.001</b>	60	0.184	98	–	
Norwegian and Barents Seas	0.099	18	0.508	30	<b>&lt;0.001</b>	101

Table 6. Main characteristics of the simple linear regression models linking AMBI and  $BQI_{ES}$  in the different subareas and individual data sets. Significant ( $p < 0.05$ ) negative correlations are in bold

Data set	N	r	p	Intercept	Slope
Norwegian and Barents Seas	204		<b>&lt;0.001</b>	31.267	–5.991
ar	22	–0.308	0.164	–	–
hs	31	–0.911	<b>&lt;0.001</b>	9.557	–1.397
ko	6	–0.667	0.148	–	–
o2	10	–0.366	0.298	–	–
o4 <sub>NB</sub>	57	–0.745	<b>&lt;0.001</b>	40.930	–8.476
o6	54	0.220	0.110	–	–
o7	15	–0.083	0.769	–	–
o8	9	0.355	0.349	–	–
Marine North Sea	850	0.013	0.715	–	–
np <sub>NS</sub>	14	–0.530	0.051	–	–
ns	224	0.315	<b>&lt;0.001</b>	10.606	1.812
o3	30	–0.913	<b>&lt;0.001</b>	29.347	–7.603
o4 <sub>NS</sub>	128	–0.416	<b>&lt;0.001</b>	28.632	–6.140
of	57	–0.800	<b>&lt;0.001</b>	20.181	–3.141
ug	357	0.261	<b>&lt;0.001</b>	4.343	0.506
Estuarine North Sea	3889	–0.040	<b>0.017</b>	4.120	–0.051
Celtic-Biscay Shelf	115	–0.602	<b>&lt;0.001</b>	20.402	–2.489
np <sub>CS</sub>	20	–0.276	0.239	–	–
o5	51	–0.212	0.136	–	–
pl	44	–0.160	0.299	–	–
Mediterranean	394	–0.250	<b>&lt;0.001</b>	19.620	–1.803
bl	2	–	–	–	–
do	49	0.291	0.042	17.437	4.196
gr	47	0.720	<b>&lt;0.001</b>	4.097	6.391
ka	190	–0.587	<b>&lt;0.001</b>	25.389	–3.893
lm	28	–0.480	<b>0.010</b>	22.373	–3.665
M0	7	0.254	0.582	–	–
M2	9	–0.727	<b>0.026</b>	31.935	–4.807
M3	6	–0.989	<b>&lt;0.001</b>	38.864	–7.583
M7	4	–0.371	0.629	–	–
M8	8	0.395	0.333	–	–
oc	1	–	12	–	–

ers of corresponding simple linear regression models always remained low, and these models differed clearly from the  $y = x$  equation.  $E(S_{50})_{0.05}$  tended to be lower in the marine North Sea than in the Celtic-Biscay Shelf and the Norwegian and Barents Seas (see Table 5 for the significance of corresponding Wilcoxon signed-rank tests).

**Comparisons between AMBI and  $BQI_{ES}$**

AMBI and  $BQI_{ES}$  correlated negatively in all 4 marine subareas and in the estuarine North Sea (Table 6, Figs. 6–10). However, in most cases these correlations were weak and found in only a few individual data sets.

The Celtic-Biscay Shelf was the only subarea where the use of a simple linear regression model seemed appropriate to account for the general negative relationship between AMBI and  $BQI_{ES}$  (Fig. 6, Table 6). However, there was no significant negative correlation between AMBI and  $BQI_{ES}$  in any individual data set within this subarea (Table 6).

A simple linear regression model did not seem appropriate to account for the relationship between AMBI and  $BQI_{ES}$  in the Norwegian and Barents Seas (Fig. 7). AMBI and  $BQI_{ES}$  correlated negatively in only 2 individual data sets (i.e. hs and o4<sub>NB</sub>, Table 6), and the slopes and the intercepts of the corresponding linear regression models differed significantly (ANCOVA,  $p < 0.001$  in both cases). Moreover, low values of AMBI sometimes also corresponded to low values of  $BQI_{ES}$  (stations in the shaded area in Fig. 7).

Negative correlations between AMBI and  $BQI_{ES}$  were found in only 4 Mediterranean individual data sets (i.e. ka, lm, M2 and M3) (Fig. 8, Table 6). The

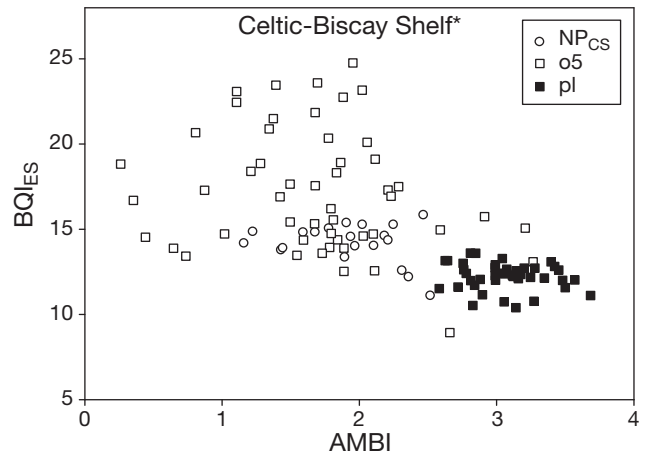


Fig. 6. Relationships between AMBI and  $BQI_{ES}$  in the Celtic-Biscay Shelf. Symbols refer to individual data sets (see Table 1). \*Significant negative correlation (for the subarea or the individual data sets) between AMBI and  $BQI_{ES}$



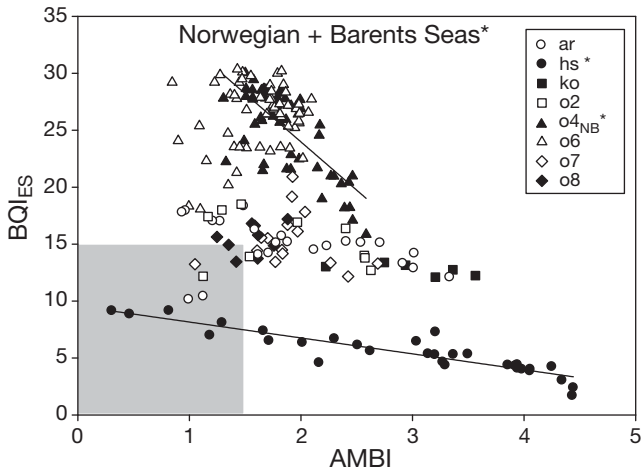


Fig. 7. Relationships between AMBI and BQIES in the Norwegian and Barents Seas. Symbols refer to individual data sets (see Table 1). \*Significant ( $p < 0.05$ ) negative correlation (for the subarea or the individual data sets) between AMBI and BQIES. Shaded rectangle in the bottom left delimits stations with a low AMBI (<1.5) and BQIES (<15) (see 'Results-Comparisons between AMBI and BQIES' for details)

slopes of corresponding linear regression models did not differ significantly (ANCOVA,  $p = 0.473$ ), whereas intercepts did ( $p = 0.027$ ). Both ka and gr contained stations characterized by low values of AMBI and BQIES (shaded area in Fig. 8, all data), which weakens the use of simple linear regression models to infer the relationships between the 2 indices for the whole Mediterranean.

In the marine North Sea (Fig. 9), high values of AMBI were also always associated with low values of BQIES. Conversely, very low values of AMBI tended to be associated with very low values of BQIES (shaded area in Fig. 9, marine North Sea). Intermediate values of AMBI were associated with a very large range (i.e. from very high to very low) of BQIES values. The analysis of individual data sets showed the occurrence of significant negative relationships between AMBI and BQIES in o3 (Fig. 9), o4<sub>NS</sub> (data not shown) and 'of' (Fig. 9). The slopes and the intercepts of corresponding linear regression models differed significantly (ANCOVA,  $p < 0.001$  and  $p = 0.007$ , respectively). Con-

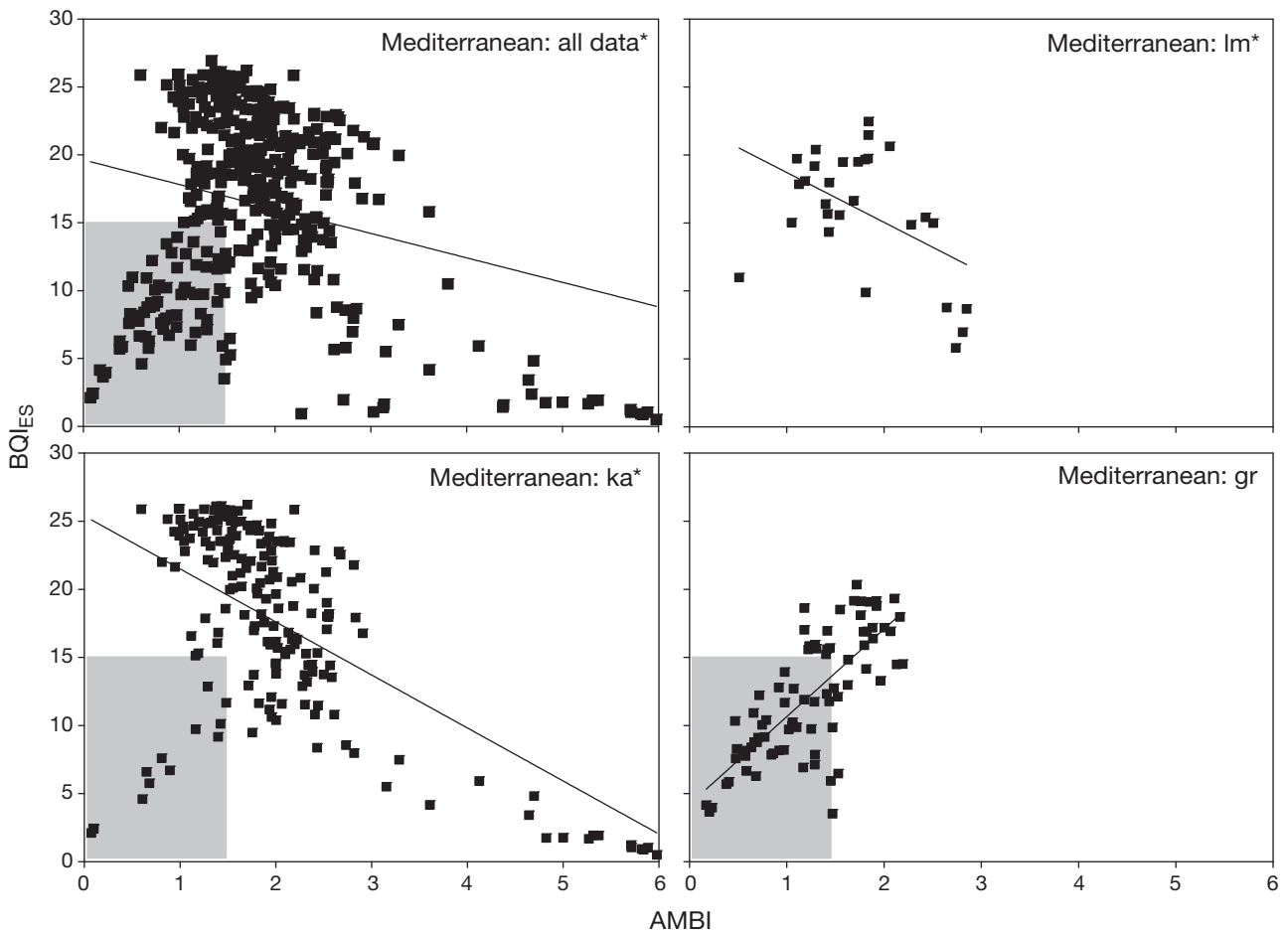


Fig. 8. Relationships between AMBI and BQIES in the Mediterranean. Data are provided for the whole Mediterranean and 3 individual data sets (see Table 1). \*Significant ( $p < 0.05$ ) negative correlation (for the subarea or the individual data sets) between AMBI and BQIES. Shaded rectangles in the bottom left of the Mediterranean, ka and gr graphs delimit stations with a low AMBI (<1.5) and BQIES (<15) (see 'Results-Comparisons between AMBI and BQIES' for details)

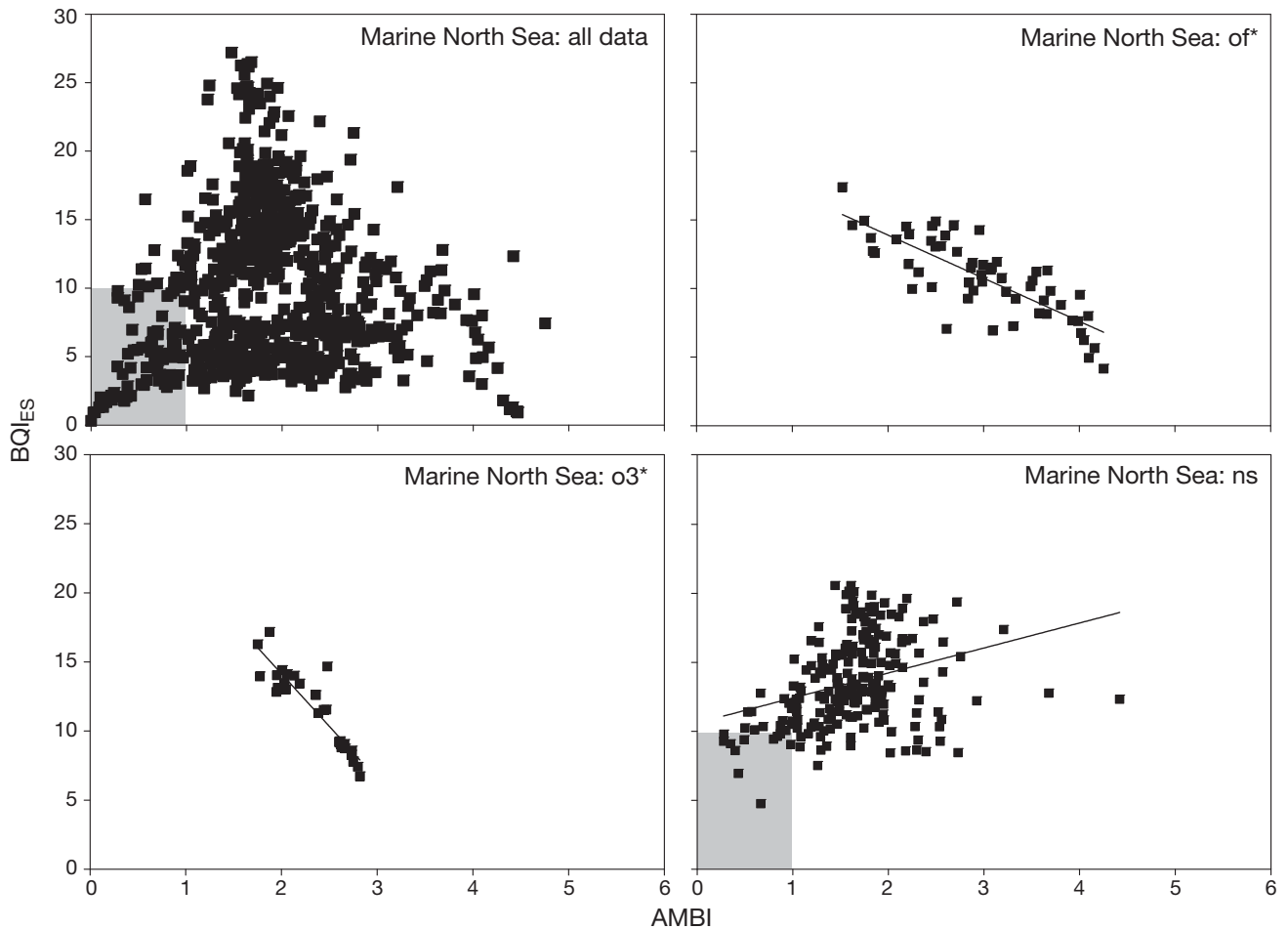


Fig. 9. Relationships between AMBI and  $BQ_{ES}$  in the marine North Sea. Data are provided for the whole marine North Sea and 3 individual data sets (see Table 1). \*Significant ( $p < 0.05$ ) negative correlation (for the subarea or the individual data sets) between AMBI and  $BQ_{ES}$ . Shaded rectangles in the bottom left of the marine North Sea and ns graphs delimit stations with a low AMBI ( $< 1$ ) and  $BQ_{ES}$  ( $< 10$ ) (see 'Results-Comparisons between AMBI and  $BQ_{ES}$ ' for details)

versely, AMBI and  $BQ_{ES}$  correlated positively in ns (Fig. 9) and ug (data not shown). The relationship between AMBI and  $BQ_{ES}$  in the estuarine North Sea (Fig. 10) was very similar to that observed in the marine North Sea.

The  $E(S_{50})_{0.05}$  and the AMBI EG of the most dominant species for each station characterized by low AMBI and  $BQ_{ES}$  (shaded areas in Figs. 7–9) are listed in Table 7. In most cases  $E(S_{50})_{0.05}$  were lower than expected from the AMBI EG values. This mismatch was especially clear for the most dominant species in the Norwegian and Barents Seas (*Maldane sarsi*), the Mediterranean (*Ditrupa arietina*, *M. glebifex*, *Turritella communis* and *Owenia fusiformis*) and the marine North Sea (*Magelona mirabilis*, *Modiolus modiolus* and *Spisula subtruncata*). Moreover, these species tended to be more dominant at the stations characterized by low AMBI and  $BQ_{ES}$  than in the whole subareas.

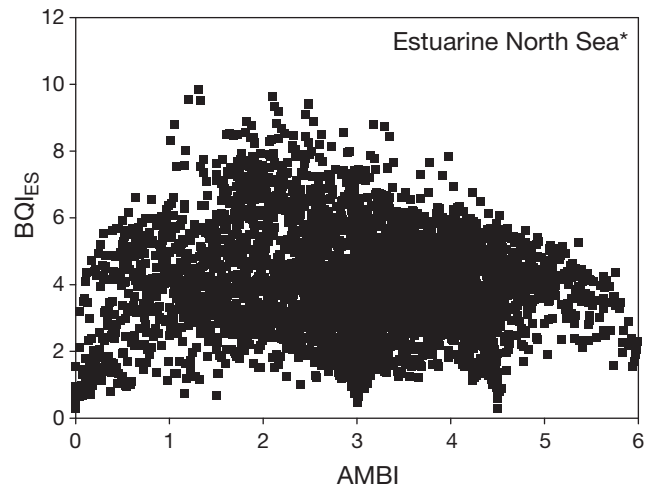


Fig. 10. Relationships between AMBI and  $BQ_{ES}$  in the estuarine North Sea. \*Significant ( $p < 0.05$ ) negative correlation (for the subarea or the individual data sets) between AMBI and  $BQ_{ES}$

Table 7. Comparison of the  $E(S_{50})_{0.05}$  and the AMBI EG of the most dominant species at each of the stations characterized by low AMBI and  $BQI_{ES}$  (shaded areas in Figs. 7–9). Species in **bold** are those for which (1) there is a clear mismatch between  $E(S_{50})_{0.05}$  and AMBI EG, and (2) dominance is higher in the corresponding shaded area. The ranges of  $E(S_{50})_{0.05}$  in each subarea are given for comparison

Species	AMBI EG	$E(S_{50})_{0.05}$	Mean dominance in shaded area (%)	Mean dominance in whole subarea (%)
<b>Norwegian and Barents Seas</b>				
<b><i>Maldane sarsi</i></b>	<b>I</b>	<b>8.5</b>	<b>29.7</b>	<b>9.7</b>
<i>Dacrydium vitreum</i>		9.8	26.2	4.4
<b><i>Lumbrineris mixochaeta</i></b>	<b>II</b>	<b>5.6</b>	<b>25.4</b>	<b>16.1</b>
<i>Lumbriclymene minor</i>	III	13.5	13.9	1.8
<i>Ophiura robusta</i>	II	11.5	9.5	4.2
<i>Chone duneri</i>	II	9.8	7.8	2.9
Range		2.9–28.3		
<b>Mediterranean</b>				
<b><i>Ditrupa arietina</i></b>	<b>I</b>	<b>3.2</b>	<b>37.8</b>	<b>17.0</b>
<b><i>Maldane glebifex</i></b>	<b>I</b>	<b>9.1</b>	<b>23.4</b>	<b>5.6</b>
<b><i>Turritella communis</i></b>	<b>II</b>	<b>4.4</b>	<b>16.4</b>	<b>8.9</b>
<b><i>Owenia fusiformis</i></b>	<b>II</b>	<b>6.2</b>	<b>13.9</b>	<b>8.0</b>
<i>Nucula nucleus</i>	I	12.1	12.0	3.0
<i>Paradoneis armata</i>	III	11.6	8.7	4.3
<i>Spisula subtruncata</i>	I	4.7	8.6	7.6
Range		1.3–28.3		
<b>Marine North Sea</b>				
<b><i>Magelona mirabilis</i></b>	<b>I</b>	<b>2.0</b>	<b>51.5</b>	<b>15.1</b>
<b><i>Modiolus modiolus</i></b>	<b>I</b>	<b>5.9</b>	<b>18.2</b>	<b>7.2</b>
<i>Urothoe brevicornis</i>	I	6.1	12.2	16.5
<b><i>Spisula subtruncata</i></b>	<b>I</b>	<b>2.1</b>	<b>8.9</b>	<b>6.8</b>
Range		1.0–24.1		

### Comparison between ES derived from AMBI and $BQI_{ES}$

The frequency distributions of the ES derived from AMBI and  $BQI_{ES}$  in the 4 marine subareas are shown in Fig. 11. In all cases there were clear discrepancies. In the Celtic-Biscay Shelf and in the Mediterranean, both indices resulted in the classification of a large majority of stations as high and good. The main differences between indices were (1) the dominance of stations classified as good by AMBI versus high for  $BQI_{ES}$  and (2) the occurrence of a larger proportion of stations classified as moderate, poor and bad by  $BQI_{ES}$  than by AMBI. Discrepancies between the indices were much larger in the Norwegian and Barents Seas and in the marine North Sea, where the majority of stations were classified as good by AMBI versus moderate, poor and bad by  $BQI_{ES}$ . In the estuarine North Sea, AMBI classified most of the stations as moderate and good versus moderate and poor for  $BQI_{ES}$  (Fig. 12). The differences in the proportions of the stations classified as high and good versus moderate, poor and bad were 15.6, 34.8, 29.3, 51.5 and 46.1% in the Celtic-Biscay Shelf, the Norwegian and Barents Seas, the Mediterranean and the marine and estuarine North Sea, respectively.

### DISCUSSION

To our knowledge, the largest comparison between EQR derived from macrozoobenthos composition in European waters was based on a database encompassing data from ca. 192 stations located in the Celtic-Biscay Shelf, the North Sea and the Kattegat (Borja et al. 2007). Three of the 4 procedures compared were based on the use of AMBI and the last one was based on the Indicator Species Index (ISI index), which is an equivalent. It was therefore not surprising that EQR computed using these procedures correlated tightly. The present study is the first to be performed at a pan-European scale (12 409 stations, including 2158 marine stations located in the Celtic-Biscay Shelf, the Mediterranean, the North Sea and the Norway and Barents Seas). Moreover, it compares AMBI and  $BQI_{ES}$ , 2 indices which show major differences in their way of assessing the sensitivity/tolerance level of individual species, and which have been shown to locally result in different ES assessments (Labruno et al. 2006, Dauvin et al. 2007, Zettler et al. 2007).

#### Facilitation of the use of $BQI_{ES}$

One of the major limitations to the spread of the use of  $BQI_{ES}$  is the difficulty in deriving  $E(S_{50})_{0.05}$ , which

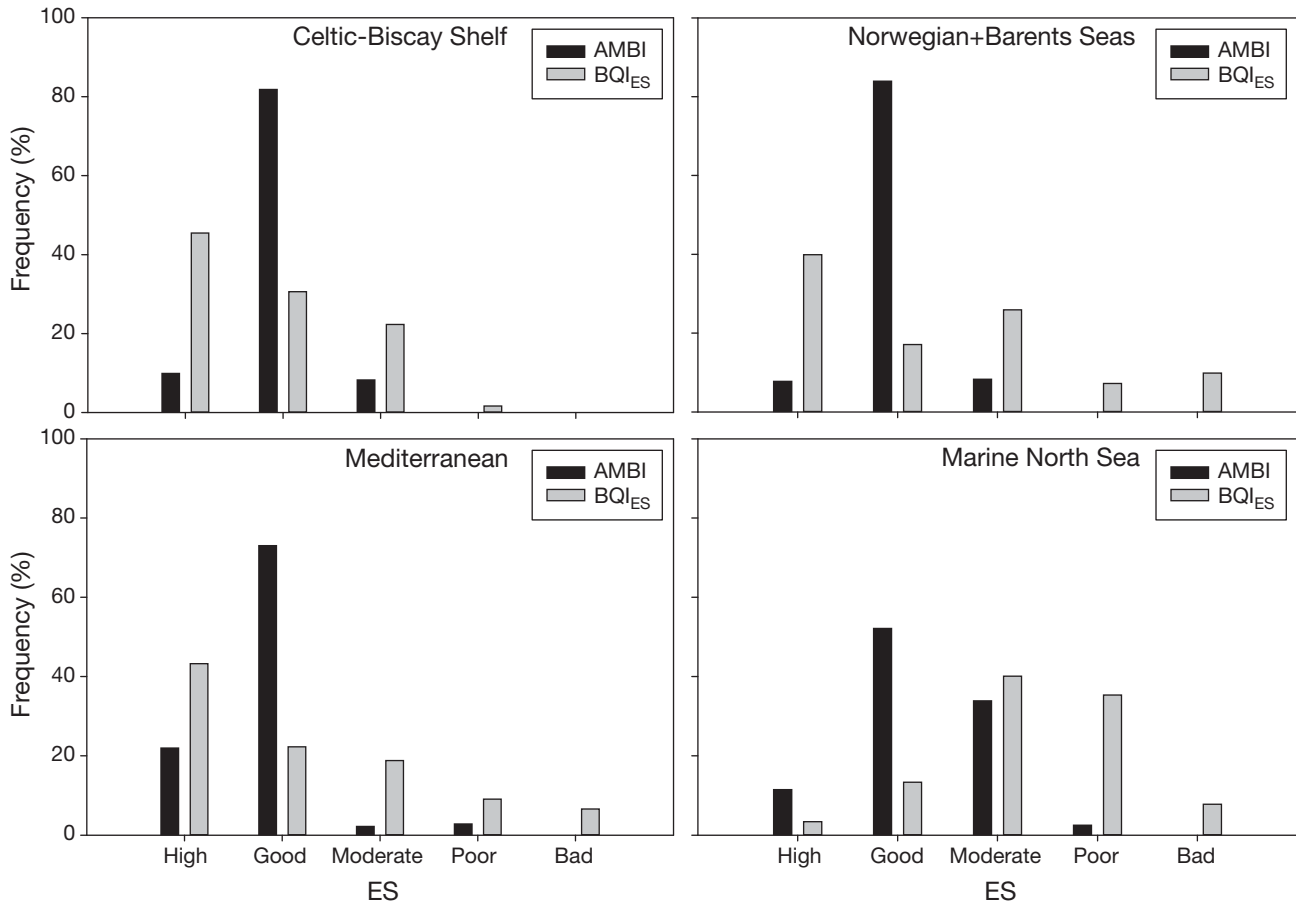


Fig. 11. Frequency distributions of ES derived from AMBI and BQI<sub>ES</sub> in the 4 marine subareas: the Celtic-Biscay Shelf, the Norwegian and Barents Seas, the Mediterranean and the marine North Sea

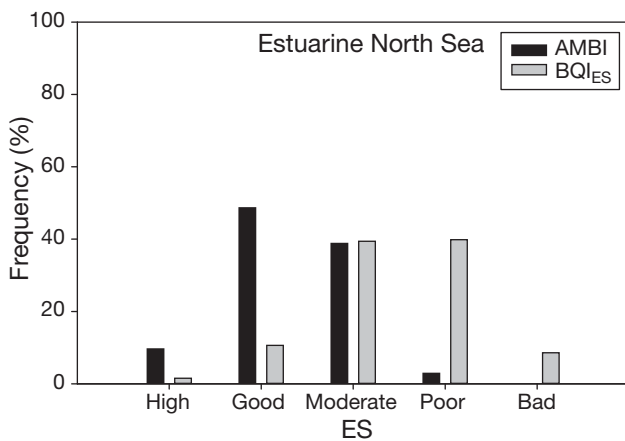


Fig. 12. Frequency distributions of ES derived from AMBI and BQI<sub>ES</sub> in the estuarine North Sea

requires the species to be present in at least 20 samples (Rosenberg et al. 2004). To our knowledge,  $E(S_{50})_{0.05}$  lists have only been compiled for the Swedish West Coast (Rosenberg et al. 2004), the Gulf of Lions (Labruno et al. 2006), the Southern Baltic (Zettler et al. 2007), the Seine estuary (Dauvin et al. 2007) and the

Marennes Oléron and Arcachon Bays (Blanchet et al. 2008). All lists are limited regarding species numbers and are not available online except for Rosenberg et al. (2004). The present study resulted in the computation of  $E(S_{50})_{0.05}$  for 643 species in the whole marine indicator data set, 76 species in the Celtic-Biscay Shelf, 246 species in the Mediterranean, 337 species in the marine North Sea and 158 species in the Norwegian and Barents Seas. The proportions of species with an  $E(S_{50})_{0.05}$  increased with the number of stations within each subarea, which simply corresponded to the increase of the proportions of species present at more than 20 stations. In spite of the size of our data sets, the proportions of species with an  $E(S_{50})_{0.05}$  were always lower than for AMBI EG, which further underlines practical difficulty in computing  $E(S_{50})_{0.05}$  and thus BQI<sub>ES</sub>. AMBI should be interpreted with caution when the proportion of non-assigned taxa is higher than 20% (Borja & Muxika 2005). To our knowledge, no such recommendation is yet available for BQI<sub>ES</sub>. Due to the strong analogy in the formula used to compute the sensitivity/tolerance terms in both AMBI and BQI<sub>ES</sub>, this figure can nevertheless also probably be used for

$BQI_{ES}$ . In this sense, it is important to note that although  $E(S_{50})_{0.05}$  were available for 91.8% of the individuals in the whole marine indicator data set, these proportions were lower than 80% both in the Norwegian and Barents Seas and in the Celtic-Biscay Shelf.

Our  $E(S_{50})_{0.05}$  lists clearly could be improved and we want to stress that other data sets could be aggregated to MacroBen to refine estimates of  $E(S_{50})_{0.05}$  in each subarea. This will facilitate the use of  $BQI_{ES}$  on small individual data sets and allow further testing of the response of  $BQI_{ES}$  to disturbances. In this sense, the present study will contribute to further testing of  $BQI_{ES}$  and/or to more specific comparative studies between AMBI and  $BQI_{ES}$ . We have also identified a list of the most dominant species in the marine indicator data set which are still either lacking an AMBI EG or an  $E(S_{50})_{0.05}$ . Effort should now be preferentially focussed on the assessment of their sensitivity/tolerance levels to further improve the use of both indices in European waters.

#### Comparison between AMBI EG and $E(S_{50})_{0.05}$

One would expect a strong negative correlation between AMBI EG and  $E(S_{50})_{0.05}$  in the case of a similar assessment of species sensitivity/tolerance levels using these 2 parameters. We indeed reported negative correlations in the whole marine indicator data set, the Norwegian and Barents Seas, and the marine and estuarine North Sea. However, the explanatory powers of the corresponding linear regression models always remained limited and we found no significant negative correlation in both the Celtic-Biscay Shelf and the Mediterranean. Our overall conclusion is that there is no good agreement between AMBI EG and  $E(S_{50})_{0.05}$ , and in this sense our results support those already collected in more restricted areas such as the Gulf of Lions (Labruno et al. 2006) or in other subareas such as the Baltic Sea (Zettler et al. 2007).

#### Assessment of the validity of the use of a single list of sensitivity/tolerance levels

Bustos-Baez & Frid (2003) showed that the response of potential indicator species to organic enrichment differed between locations, and Rosenberg et al. (2004) found that AMBI EG may vary between geographical areas. It was, therefore, interesting to compare  $E(S_{50})_{0.05}$  between subareas; the poor agreement probably did not result from differences in anthropogenic pressures.  $E(S_{50})_{0.05}$  values are mostly dependent on the  $E(S_{50})$  of stations with low species richness. For  $E(S_{50})_{0.05}$  to be comparable, it is thus not necessary for

the levels of anthropogenic pressures to be strictly equivalent between subareas, but rather that a wide range from disturbed to undisturbed stations is present in all subareas. Unfortunately, there is no comprehensive information available on the level of disturbance experienced by each station in MacroBen. However, the Pearson & Rosenberg (1978) model states that species richness decreases with disturbance. The large ranges of  $E(S_{50})$  recorded within each subarea therefore suggest that both disturbed and undisturbed stations were indeed present in each subarea. This was further confirmed by the large ranges of  $E(S_{50})_{0.05}$  found within each marine subarea (see Table 7). Our results thus support those of Labruno et al. (2006) in showing that there are heterogeneities in  $E(S_{50})_{0.05}$  computed for different subareas. This does not support the use of a single list of species sensitivity/tolerance levels at the pan-European scale.

Overall, the relationships (1) between AMBI EG and  $E(S_{50})_{0.05}$  and (2) of  $E(S_{50})_{0.05}$  between subareas were rather noisy. If sensitivity/tolerance levels indeed vary between geographical areas, they also probably vary between habitats within a single geographic area, which may be partly responsible for the noise observed during the present study. Up to now (and the present study is no exception), AMBI EG and even  $E(S_{50})_{0.05}$  have never been assessed at the habitat level. Interesting lines for future research would thus consist of comparing  $E(S_{50})_{0.05}$  (1) within the same subarea but between habitats and (2) within the same habitat but between subareas. In both cases, this will require the construction of large and comprehensive databases and we suggest that this exercise should first focus on a restricted set of well-studied habitats.

#### Unravelling the causes of discrepancies between the 2 indices

The negative correlation between AMBI and  $BQI_{ES}$  was satisfactory only in the Celtic-Biscay Shelf. Interestingly, there was no significant negative correlation between AMBI EG and  $E(S_{50})_{0.05}$  in this subarea, which suggests that the agreement between the values of the 2 indices is not necessarily reliant on the general correlation between their assessments of sensitivity/tolerance levels. In all other subareas, AMBI and  $BQI_{ES}$  correlated only poorly. Overall, stations with high AMBI also tended to have low  $BQI_{ES}$ . Conversely, some of the stations with low AMBI also featured low  $BQI_{ES}$ . The present study shows that this mostly resulted from strong dominance by species classified as sensitive by AMBI but with a low  $E(S_{50})_{0.05}$ . Labruno et al. (2006) reported a positive correlation between AMBI and BQI in the Gulf of Lions and attributed this result to the



strong dominance of the serpulid polychaete *Ditrupa arietina* (Grémare et al. 1998, Labruno et al. 2007a), which was classified as sensitive by AMBI and had a low  $E(S_{50})_{0.05}$ . Our results support this interpretation and generalize it to other geographical areas (e.g. the Cretan Shelf) and to other species. The present study provides the first lists of the most dominant species within each marine subarea for which there are important discrepancies between AMBI EG and  $E(S_{50})_{0.05}$ . All were classified in AMBI EG I or II. However, some of them are known to be influenced by natural sources of disturbance such as sediment instability (*D. arietina*, Grémare et al. 1998 and *Magelona mirabilis*, Rayment 2007) or climatic anomalies (*Maldane glebifex*, Glémarec et al. 1986) and cycles (*D. arietina*, Labruno et al. 2007b). These observations are indicative of the tendency of  $E(S_{50})_{0.05}$  to automatically classify dominant species as tolerant and its inability to differentiate between natural and anthropogenic sources of disturbance (Labruno et al. 2006, 2007b). Further autoecological studies are nevertheless clearly needed to better unravel the actual sensitivity/tolerance levels of the species highlighted in Table 7.

#### Comparison of ES assessments derived from AMBI and $BQI_{ES}$

Given the discrepancies between AMBI and  $BQI_{ES}$ , it was not surprising that the frequency distributions of ES derived from these 2 indices differed in most subareas. In the Norwegian and Barents Seas and both the marine and estuarine North Sea, these discrepancies were also apparent when distinguishing stations with a high or good ES from those with a moderate, poor or bad ES as recommended by the WFD.  $BQI_{ES}$  resulted in overall poorer ES than AMBI, which supports preliminary results in the Gulf of Lions (Labruno et al. 2006), the Southern Baltic (Zettler et al. 2007) the Bay of Seine (Dauvin et al. 2007) and to a lesser extent the North Sea (Reiss & Kröncke 2005).

It should be underlined that all the above-mentioned studies plus the present one have used a fixed conversion scale to infer ES from AMBI. One of the characteristics of the recently introduced M-AMBI is that it is using a different conversion scale for each homogeneous habitat as does  $BQI_{ES}$  (Borja et al. 2007, Muxika et al. 2007a). In both cases, this requires the existence of valid references (i.e. a single high reference in the case of  $BQI_{ES}$ , and both a bad and a high reference in the case of M-AMBI). The computation of M-AMBI was not integrated in the MacroBen tool and we did not use this procedure to infer ES during the present study.

## CONCLUSIONS

AMBI and  $BQI_{ES}$  both ultimately rely on species sensitivity/tolerance levels, which they respectively assess through AMBI EG and  $E(S_{50})_{0.05}$ . We identified the most dominant species in marine European waters still lacking an AMBI EG or an  $E(S_{50})_{0.05}$ . Our results support those of previous studies, obtained at much smaller geographical scales, in showing that AMBI EG and  $E(S_{50})_{0.05}$  poorly agree. They suggest that the use of a single sensitivity/tolerance list in different geographical areas (such as in AMBI EG) is not appropriate. Discrepancies between the values of the 2 indices are due to the dominance of species characterized as sensitive by AMBI and tolerant by  $BQI_{ES}$ . These species were identified and some of them are known to be influenced by natural disturbance, which highlights the tendency of  $BQI_{ES}$  to classify dominant species as tolerant and thus to be inefficient in distinguishing anthropogenic from natural disturbances. AMBI and  $BQI_{ES}$  thus both present weaknesses relative to the assessment of sensitivity/tolerance. Both indices have been subject to several recent refinements regarding their computation and their procedures to infer ES, which are now quite comparable. However, all these steps are posterior (and thus dependent on) a sound assessment of species sensitivity/tolerance. Changes in the scales used to convert indices to ES can only partially compensate for changes in sensitivity/tolerance levels among geographical areas and/or habitats. Preferential attention should thus now be paid to this particular issue. Future studies should focus on (1) the clarification of the sensitivity/tolerance levels of the species identified as problematic during the present study, and (2) the assessment of the relationships between AMBI EG and  $E(S_{50})_{0.05}$  within and between combinations of geographical areas and habitats.

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