Original Paper

THE ASSESSMENT OF SENSITIVITY OF BIOTIC INDICES IN DETERMINING SYMPTOMS AND LEVELS OF DISTURBANCE USING MACROBENTHIC ASSEMBLAGE DATA

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ABSTRACT

Marine macrobenthic dynamics have been intensively studied in response to environmental disturbances, and a large number of techniques, including a variety of indices, have been proposed and developed for assessment. Structural and functional variability have been summarized through both univariate and multimetric indices as integrative indicators in environmental monitoring programs. In general, multimetric indices are considered to be sensitive, stable, and robust, thus offer a promising approach for ecological assessment. This study is aimed to assess the degree of sensitivity for several indices in terms of their ability to determine environmental changes. Based on macrofaunal data, several univariate and multimetric indices were used to assess and compare the level of disturbance at fallowed farm and reference (control) sites. The two multimetric indices, the AZTI's Marine Biotic Indices (AMBI) and Ecological Quality Ratio (EQR), were used as integrative indicators to assess the categorisation of each sampled site. The results showed that a combination of multimetric index and univariate indices provide a better assessment. The categories determined by multimetric indices seem to be in accordance with level of disturbance expressed by the trophic analysis, multivariate and graphical analyses used in this study. The AMBI has the ability to detect large scale differences among sites. However, AMBI was unable to discriminate slight changes in the macrobenthic assemblages between sites, as have been exposed by EQR.

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INTRODUCTION

Changes in the diversity, biomass, abundance, and distribution of the macrobenthic fauna has been recognized as a sensitive indicator of marine environmental changes, whether these are natural or anthropogenic (Reiss and Kroncke; 2005; Quintino *et al.*, 2006). Consequently, marine macrobenthic dynamics in response to environmental disturbances have been intensively studied, and a large number of techniques, including a variety of indices, have been proposed and developed for assessment (Reiss and Kroncke, 2005; Quintino *et al*., 2006). Structural and functional variability have been summarized through both univariate and multimetric indices as integrative indicators in environmental monitoring programs. The variability of fauna structure can thus be easily detected by assessing composition and distribution of opportunistic species, and summarized as an index.

A broad range of methods in assessing and mapping marine benthic habitats have been developed, particularly the use of multimetric indices (Diaz *et al*., 2004). Despite the need for further evaluations, in general, multimetric indices are considered to be sensitive, stable, and robust (Diaz *et al*., 2004), and thus offer a promising approach for ecological assessment (Reiss and Kroncke, 2005). Although some information may be lost as a result of the use of biotic indices, it is a simple way to present

complex data to potential users (Rosenberg *et al.*, 2004).

Because the concept of species diversity involves combining the number of species and their relative abundance, diversity indices utilize two features of sample information: the species richness and their equitability or evenness (Clarke and Warwick, 2001). The indices commonly used to assess 'community' structure include total number of species (S) and Margalef index (d), Shannon-Wiener index (H') and Hurlbert Index (ES_n)), Pielou's evenness index (J'), and Simpson index ($1 - \lambda'$). By categorizing species into ecological groups of species or taxa (trophic groups, sensitive/tolerant/opportunist, etc.), which characterize different stages of altered structure according to their sensitivity to stress, several multimetric indices have also been developed. These multimetric indices include the Biotic Index (BI), the AZTI's Marine Biotic Indices (AMBI) (Borja, 2004), the Infaunal Trophic Index (ITI) (Cromey *et al.*, 2002), the Biotic Index (BENTIX) (Simboura, 2004; Simboura *et al.*, 2006; Zenetos *et al.*, 2004), the Benthic Index of Biotic Integrity, the Biotic Quality Index (BQI) (Rosenberg, *et al.*, 2004), and Ecological Quality Ratio (EQR) (Quintino *et al.*, 2006).

Although the Shannon-Wiener index (H') depends on sample size, it is the most commonly used in benthic ecology. The Hurlbert index (ES_n) is less dependent on sample size than H' index and is based on the rarefaction technique of Sanders modified by Hurlbert. The SEP compares H' biomass and H' numbers (abundance). The AMBI uses the ecological strategies of the r, k and T proposed by Pianka and the progressive steps of successional stages in stressed environments as the theoretical basis (Borja, 2004). The AMBI has been tested on various benthic data representing a large variety of combinations of environments and disturbance sources, and is considered to be capable of detecting several environmental impact sources, such as dredging, engineering works, sewerage plans and the dumping of polluted waters (Borja *et al.,* 2000). The EQR is a multimetric index developed for the European Water Framework Directive (WFD), combining AMBI, Simpson's index, abundance and number of species/taxa in one cumulative index (Borja *et al.*, 2003; Reiss and Kroncke, 2005). Because the index is

highly dependent on the Pearson-Rosenberg succession model for organic enrichment it is still being developed and requires further testing (Quintino *et al.*, 2006).

In this study, several univariate and multimetric indices are used to assess and compare the level of disturbance at fallowed and control sites. The degree of sensitivity for each index in terms of its ability to determine environmental changes is discussed. The relative ability of multimetric and univariate indices, to generate comprehensive assessment of the benthic structure is also discussed. The two multimetric indices, AMBI and EQR, were used as integrative indicators to assess the categorisation of each sampled site. It was hypothesized that the distribution of macrobenthic abundance among trophic groups, expressed by the selected biotic indices, would be more diverse at the control sites than at fallowed pontoon sites. As EQR includes AMBI in its calculation, it was also hypothesized that the two integrative indicators would give relatively similar values of assessment for each sampling site.

MATERIALS AND METHODS

Sampling sites

Data of abundance and biomass of macrobenthic assemblages used for the computation of biotic indices were generated from sampling sites. The sites were located between 135° 58.25' to 135° 59.82' E and 34° 35.41' to 34° 42.43' S, in Southern Spencer Gulf, South Australia, where farming of southern bluefin tuna (*Thunnus maccoyii*) takes place. Fallowed farm sites were sampled after all fish and pontoons were removed and their coordinates were recorded. Reference (control) sites were at least 1 km from any leased site. Samples were subsequently collected five times during the period from October 2002 to October 2003. Sediment samples were taken using a HAPS bottom corer equipped with a corer of 67 mm in diameter and 315 mm in length, operated from the research vessel RV Ngerin.

Environmental assessment

Among various indices proposed in the literature, six multimetric indices were selected

and evaluated for environmental assessment. These were: Shannon-Wiener Evenness Proportion (SEP), Azti's Marine Biological Index (AMBI) (Borja *et al.*, 2000) and its reciprocal (1/AMBI) (Quintino *et al.*, 2006), Infaunal Trophic Index (ITI), Ecological Quality Ratio (EQR) (the UK MBITT Multimetric Approach in Quintino *et al.,* 2006), W statistics and Index of Multivariate Dispersion (IMD). Quantitative variables and univariate indices were also measured for comparison and descriptive purposes against multimetic indices. They were: total abundance (A), total species richness (S), total biomass (B), A/S (abundance : species ratio), B/A (biomass : abundance ratio), Shannon–Wiener index (H′), Margalef index (d), Pielou's evenness index (J'), Hurlbert Index (ES_n), Simpson's Index (1−D). Calculations of univariate and multimetric indices used in this study are shown in **Table 1.**

Table 1. Quantitative variables, univariated and multimetric indices used for this study.

Variable	Calculation/ Equation	References
Quantitative variables:		
S	Number of species/taxa	-
A	Number of individuals m^{-2}	-
B	Biomass	-
Univariate indices:		
A/S	Abundance : species ratio	Pearson *et al.* (1982)
B/A	Biomass : abundance ratio	
H'	$$-\sum_i p_i \log(p_i)$$	Krebs (1989); Clarke & Warwick (2001
d	$$\frac{(S-1)}{\log N}$$	
J'	$$\frac{H'}{\log N}$$	
1-λ'	$$\frac{1-\sum_i N_i(N_i-1)\}}{\{N(N-1)\}}$$	Hurlbert (1971) Rosenberg et al. (2004)
ES(50)	$$\sum_{i=1}^{S}\left[1-\frac{(N-N_i)!(N-50)}{(N-N_i-50)!\,N!}\right]$$	
Multimetric indices:		
SEP	H' biomass / H' numbers	McManus & Pauly (1990)
AMBI and its reciprocal	{(0x%GI)+(1.5x%GII)+(3x%GIII)+ (4.5x%GIV)+(6+%GV)}/100	Borja *et al.* (2000); Quintino *et al.* (2006)
ITI	$$100-\left\{33.33\left[\frac{(0n_1+n_2+2n_3+3n_4)}{(n_1+n_2+n_3+n_4)}\right]\right\}$$	Cromey *et al.* (1998)
EQR	$$\frac{\left(\left(2\times\left(1-\left(\frac{AMBI}{7}\right)\right)\right)+(1-\lambda)\right)}{3}\times$$ $$\frac{\left(\left(1-\left(\frac{1}{A}\right)\right)+\left(1-\left(\frac{1}{S}\right)\right)\right)}{2}$$	Quintino *et al.* (2006)
Wstatistic	$$\sum_{i=1}^{S}\frac{(B_i-A_i)}{[50(S-1)]}$$	Warwick (1986);; Warwick & Clarke (1994)
IMD	$$\frac{2(\bar{r}_c-\bar{r}_t)}{(N_c+N_t)}$$	Warwick & Clarke (1993)

Cluster analysis

Hierarchical cluster analyses of the (Un-weighted Pair Group Mean Average algorithm (UPGMA) and Non-metric Multi-Dimensional Scaling (NMDS) plots were used to assess the relationships between the indices. The resemblance matrix of Spearman rank correlation coefficients was created from the squared-root transformed data of biotic indices. All multivariate analyses were performed using the software PRIMER, version 6.1.5 (Clarke and Gorley, 2006). The increasing level of variability among samples/sites was assessed using Index of Multivariate Dispersion (*IMD*) by quantifying the differences in relative faunal variability between the fallowed and the control sites.

RESULTS

Correlation and variability between indices

The general trends of macrobenthic structure expressed by quantitative variables (total abundance, total species richness, total biomass), and univariate indices (Shannon–Wiener diversity index, Margalef index, Pielou's evenness index, and Simpson's Index) have been discussed. **Fig. 1.** shows the results of a cluster analysis and ordination technique derived from Pearson correlation matrices of all biotic indices used in this study. The EQR is mainly influenced by 1/AMBI, with a strong correlation between the two indices (Spearman rho = 0.96, p = 0.01), whereas AMBI does not seem to be influenced by any index nor quantitative variable. Other correlations, such as between Simpson index and Shannon-Wiener index (Spearman rho = 0.94), and between the two indices and Pielou's evenness index (Spearman rho = 0.87) were also observed.

Table 2 shows the average scores of each sampling site assessed by AMBI, ranging from 1.711 to 2.252. These scores classify all the sites as 'slightly polluted'. However, a slight temporal variation was observed within a site. The sites that varied differently over time by AMBI were P05, P06, and RC5, particularly owing to the changes of the proportion of their ecological groups during the study period, as shown in **Table 3** and **Fig. 2.**

As it can be seen in **Fig. 2**, a high temporal variability between ecological groups for the selected sites is obvious. At site RC5, the relative abundance of group I ('disturbance-sensitive') was the highest, whilst the relative abundance of group III ('disturbance-tolerant') and group V ('first-order opportunistic') were the lowest in October 2003. Therefore, AMBI classified this site as 'undisturbed' (**Table 3**). Similarly, at site P05 was classifying as 'undisturbed' in October 2003. A site P06, however, the relative abundance of group V was the highest in October 2002, compared to the other sampling times. This leads to the site being classified as 'moderately disturbed'. This classification was mainly due to the occurrence of higher numbers of capitellid polychaetes at site P06.

Ecological classification of the sites

The environmental classification for each site and sampling time based on AMBI and EQR are shown in **Fig.3.** Both indices classify control and fallowed sites into a different ecological status, giving the ranges of habitat quality from 'unpolluted' to 'moderately polluted', based on AMBI, and from 'high' to 'moderate', based on EQR. The AMBI classifies both control and fallowed sites mainly as slightly polluted over the study period, except for P06A that was classified as moderately polluted, and RC5E as unpolluted. Meanwhile, the classification of EQR showed higher variability between sites and times, compared to AMBI.
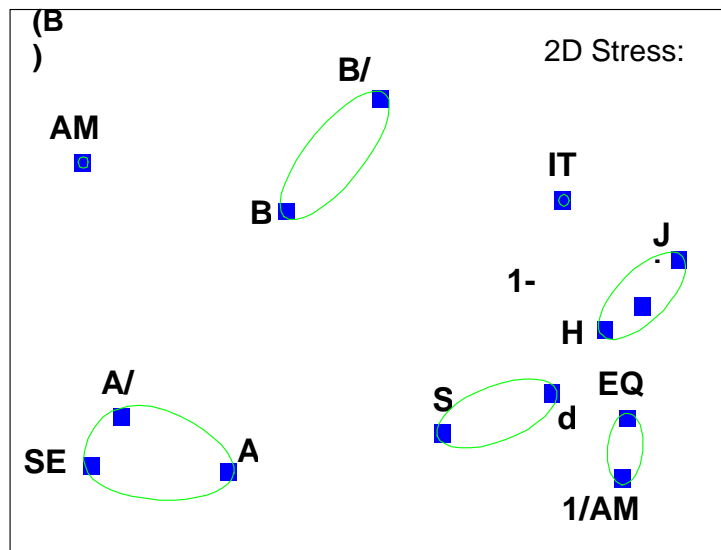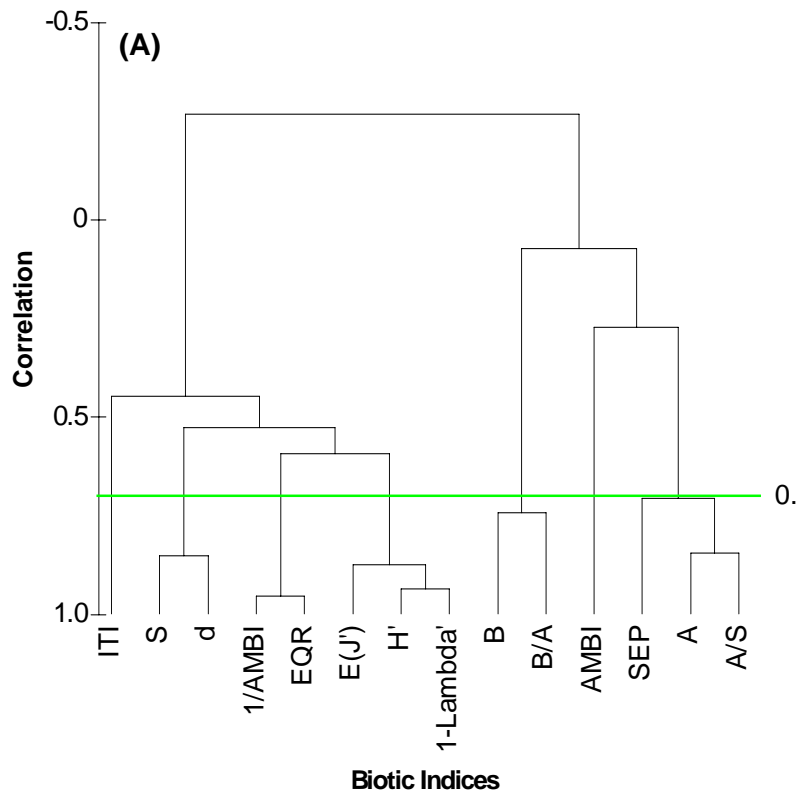
Fig. 1. Dendrograms (A) and MDS plots in 2 dimensions (B) of biotic indices derived from Spearman rank correlation coefficient matrix of square root transformed data. Green circles derived from cluster analysis, indicate an arbitrary correlation level of 0.7.

Table 2. The average scores of AMBI for each control and fallowed site over the study period, with also show relative abundance of ecological groups: (I) disturbance sensitive, (II) disturbance-indifferent, (III) disturbance-tolerant, (IV) second-order opportunistic, and (V) first-order opportunistic.

SITE	Groups of macrobenthic animals					Mean AMBI
	I(%)	II(%)	III(%)	IV(%)	V(%)	
Control sites						
BC4	30.2	36.1	15.7	13.3	4.7	**1.924**
BC5	25.9	38.7	20.7	8.3	6.4	**1.982**
BC7	21.3	34.2	33.8	6.5	4.2	**1.995**
BC8	23.2	45.8	21.1	4.4	5.6	**1.867**
RC1	37	32	18.4	6	6.7	**1.711**
RC3	30.8	39.9	20.6	3.3	5.9	**1.694**
RC5	37.3	29.9	24.7	3.5	4.6	**1.731**
RC7	34.1	30	24.6	5.9	5.4	**1.776**
Fallowed sites						
P01	10.7	38.4	44.5	2.5	3.9	**2.164**
P02	18.1	32.8	37.3	5	6.9	**2.252**
P03	20.5	25.3	38.9	2.4	12.8	**2.456**
P04	30.4	37.4	14.1	13.4	4.8	**1.864**
P05	30.6	31.7	22.5	9.8	5.4	**1.862**
P06	22.1	44.7	13.8	4.6	14.8	**1.994**
P07	28	44.4	17.2	6.6	3.8	**1.692**
P08	17.6	37.5	32.9	7.5	4.6	**2.095**

Table 3. The ecological status of the selected sites according to the AMBI assessed using relative abundance of ecological groups summarized by Grall and Glemarec (1997): (I) disturbance sensitive, (II) disturbance-indifferent, (III) disturbance-tolerant, (IV) second-order opportunistic, and (V) first-order opportunistic.

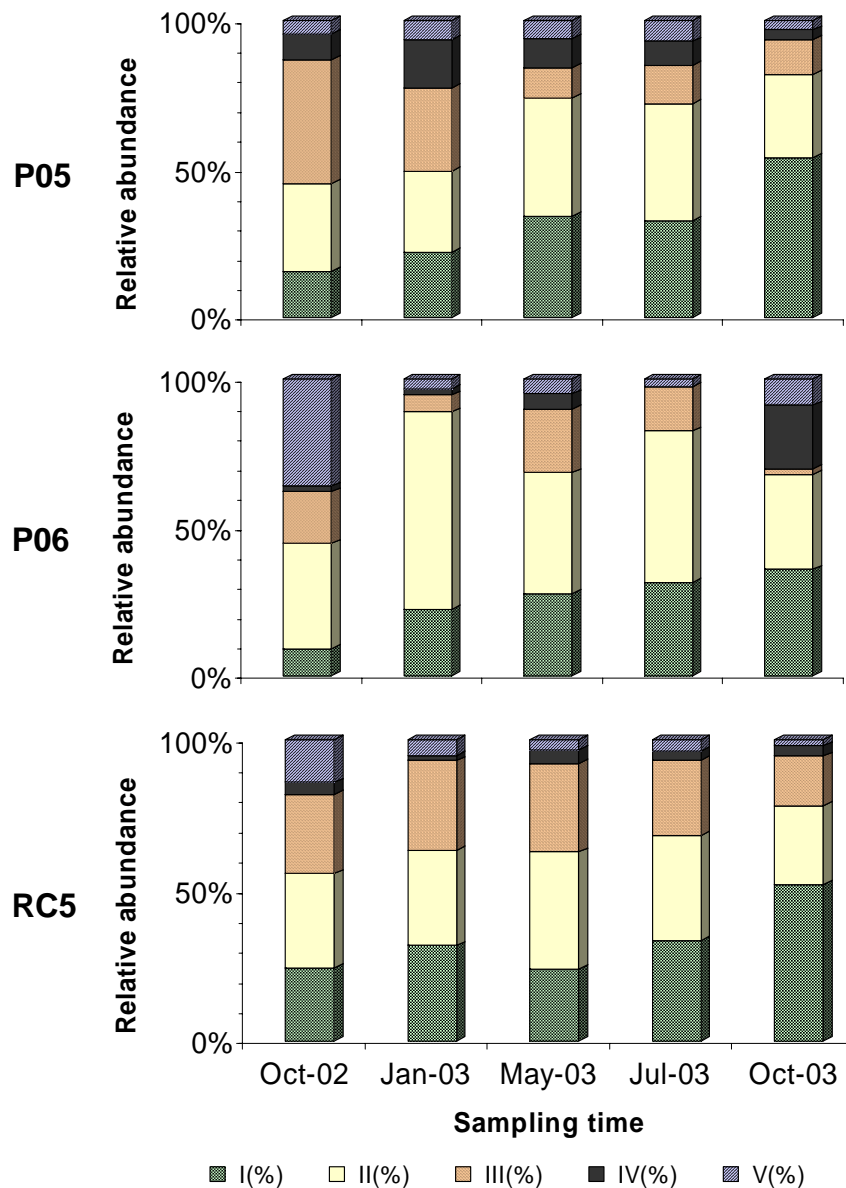| SITE	Groups of macrobenthic animals					Mean AMBI	BI	Disturbance Classification
RC5	I(%)	II(%)	III(%)	IV(%)	V(%)			
Oct-02	23.1	29.7	25	3.8	13.5	**2.25**	**2**	**Slightly disturbed**
Jan-03	27.3	27.3	25.8	1.5	4.5	**1.727**	**2**	**Slightly disturbed**
May-03	24.4	40	30.2	4.7	3.5	**1.884**	**2**	**Slightly disturbed**
Jul-03	37.9	39.5	28.4	3.4	4.3	**1.655**	**2**	**Slightly disturbed**
Oct-03	55.1	28	17.4	3.6	2.2	**1.141**	**1**	**Undistrubed**
P05								
Oct-02	15.4	29.7	41.8	8.8	4.4	**2.357**	**2**	**Slightly disturbed**
Jan-03	21.9	27.3	28.1	16.4	6.3	**2.367**	**2**	**Slightly disturbed**
May-03	34	40	10	10	6	**1.71**	**2**	**Slightly disturbed**
Jul-03	32.6	39.5	12.8	8.1	7	**1.762**	**2**	**Slightly disturbed**
Oct-03	53.8	28	11.8	3.2	3.2	**1.113**	**1**	**Undistrubed**
P06								
Oct-02	9	35.5	17.4	1.9	36.1	**3.31**	**3**	**Moderately distrubed**
Jan-03	22.2	66.7	5.6	2.2	3.3	**1.467**	**2**	**Slightly disturbed**
May-03	27.6	40.8	21.4	5.1	5.1	**0.791**	**2**	**Slightly disturbed**
Jul-03	31.3	51	15	0	2.5	**1.369**	**2**	**Slightly disturbed**
Oct-03	35.7	32.1	1.8	21.4	8.9	**2.036**	**2**	**Slightly disturbed**

Fig. 2. Relative abundance of the ecological groups of three selected sites, based on the AMBI, over the study period.
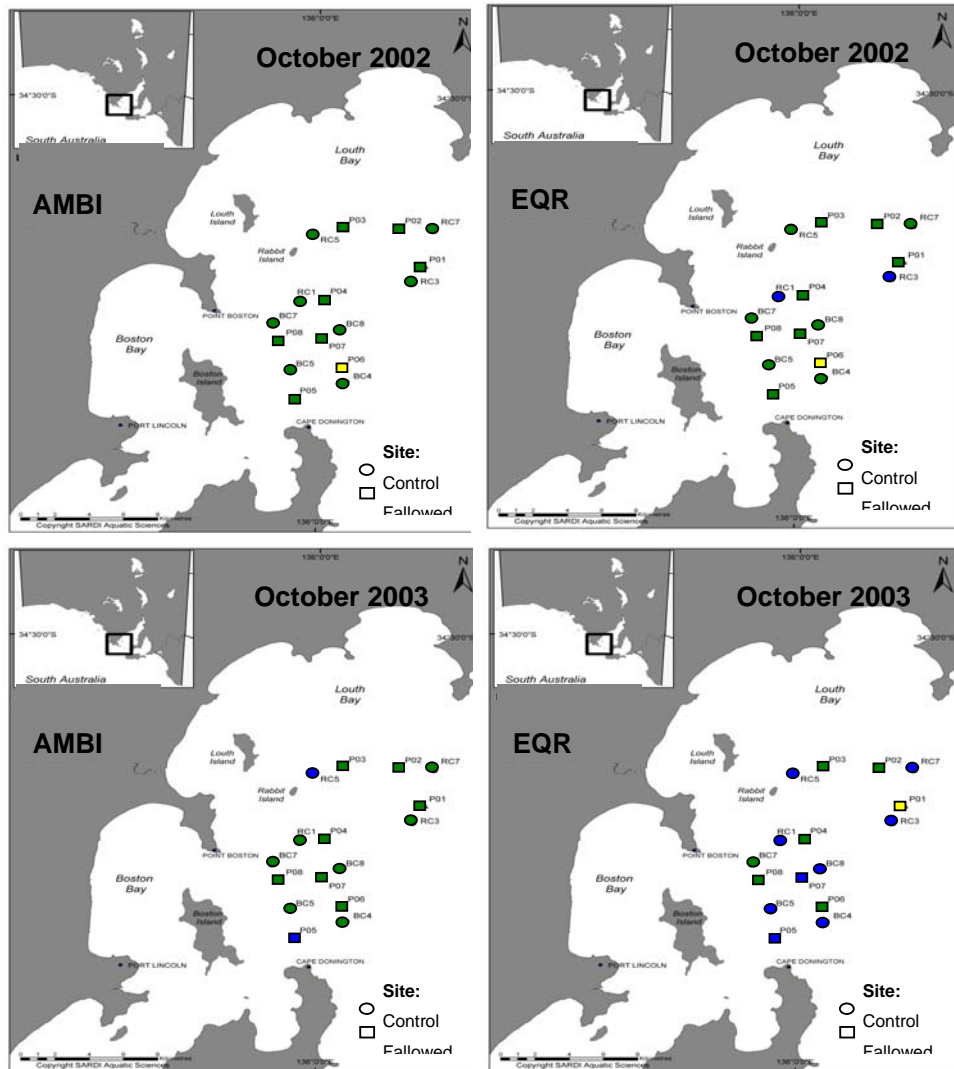
For both the control or fallowed sites, AMBI classifies 97.5% of the sites as 'slightly polluted', 1.25% of the sites as 'moderately polluted', and 1.25% of the sites as 'unpolluted' over the study period. In contrast, EQR classifies 70% of the sites as 'good', 27.5% of the sites as 'high', and 2.5% of the sites as 'moderate'. The sites that have different classification based on the two indices are listed in **Table 4.** The only site that was classified as 'moderate' or 'moderately polluted' by both indices, is site P06A . This classification is in accordance with Shannon-Wiener diversity index (H') , SEP, and W-statistic, which categorize the site as 'moderate' by H' (2.3), 'disturbed' by SEP (1.23), and 'moderately polluted' by the W-statisitic (+0.03) (**Table 5**). Meanwhile, site P01E is classified as 'moderately polluted' only by The EQR, which is also supported by the values of univariate

indices, such as diversity, evenness, and richness indices (**Table 4**).

The values of all univariate indices were the lowest for P01E, suggesting that this site has the lowest taxa richness, evenness, and diversity (and intrinsically high dominance) compared to the other sites in October 2003's samples (**Table 4**). These conditions are suggested as an indication of more disturbed area than any other sites. Considering the scores by other indices, AMBI failed to classify site P01E accurately, because this site was classified as 'slightly polluted' according to AMBI, while it was classified as 'moderate' by EQR and diversity indices.

Ecological Classification for AMBI:

■ Unpolluted   ■ Slightly polluted   ■ Moderately polluted   ■ Polluted   ■ Heavily polluted

Ecological Classification for EQR:

■ High   ■ Good   ■ Moderate   ■ Poor   ■ Bad

Fig. 3. Map showing the ecological classification of 8 control and 8 fallowed sites over a full year period of sampling times based on AMBI and EQR values.

Table 4. The sites with different classifications according to the AMBI and EQR over the study period. The other univariate and multimetric indices are shown for comparisons.

Site	Univariate indices				Multimetric indices			
	H'	d	J'	1 - λ'	SEP	Wstat	AMBI	EQR
Oct 2002								
RC1A	2.98	5.94	0.94	0.96	0.92	0.31	1.71	0.80
RC3A	2.91	5.73	0.91	0.94	1.00	0.19	1.55	0.81
PO6A	2.34	4.16	0.76	0.84	1.23	0.03	3.31	0.61
Jan 2003								
RC1B	2.75	5.41	0.84	0.91	1.11	0.07	1.55	0.80
RC3B	2.96	5.66	0.90	0.94	1.05	0.11	1.45	0.82
PO6B	2.61	5.89	0.78	0.87	1.12	0.23	1.47	0.80
May 2003								
BC4C	2.99	6.43	0.89	0.94	1.00	0.26	1.62	0.81
RC1C	2.88	5.32	0.89	0.93	1.04	0.16	1.36	0.83
RC3C	2.79	5.10	0.86	0.92	1.11	0.05	1.57	0.80
RC7C	2.92	5.69	0.90	0.94	1.01	0.23	1.68	0.80
PO6C	3.15	7.31	0.89	0.95	1.03	0.22	1.79	0.80
Jul 2003								
PO4D	3.07	6.33	0.89	0.95	1.04	0.13	1.65	0.81
PO5D	3.05	6.46	0.90	0.95	0.99	0.25	1.76	0.80
PO6D	3.06	6.70	0.89	0.94	1.01	0.24	1.37	0.83
PO7D	2.85	5.48	0.89	0.94	1.02	0.21	1.56	0.81
Oct 2003								
BC4E	3.01	6.31	0.90	0.95	1.00	0.23	1.74	0.80
BC5E	3.01	5.99	0.94	0.95	0.94	0.33	1.69	0.80
BC8E	2.90	5.66	0.88	0.94	1.06	0.13	1.61	0.81
RC5E	2.91	5.87	0.86	0.94	1.05	0.12	1.14	0.85
RC7E	2.95	5.44	0.89	0.94	1.01	0.19	1.75	0.80
PO1E	1.73	3.73	0.56	0.66	1.54	0.10	2.48	0.64
PO5E	2.71	5.02	0.85	0.91	1.08	0.15	1.11	0.84
PO7E	2.83	5.94	0.85	1.23	1.04	0.23	0.84	0.23

DISCUSSION

A broad range of methods for assessing and mapping marine benthic habitats have been developed. Consequently, interest in benthic indicators has increased during recent years, particularly the use of multimetric indices (Diaz, 2004). The successional pattern of macrobenthic structure in response to environmental disturbance proposed by Pearson & Rosenberg has been used in developing most recent biotic indices (Dauvin *et al*., 2007; Quintino *et al*., 2006; Borja, 2004; Labrune *et al.*, 2006). Along an increasing gradient of organic enrichment, macrofauna communities tend to change in diversity, abundance, and species composition depending on their tolerance to disturbance. In terms of secondary succession following a disturbance, species richness tends to increase, while their dominance tends to decrease. The changes of dominant species from polluted-tolerant to polluted-sensitive species indicate improvement of habitat quality.

The results from multimetric index analyses, used to categorise the study sites, showed little variability between sites and times. Most of the categories were in accordance with level of disturbance expressed

by trophic, multivariate and graphical analyses used in this study. The multimetric indices tend to be less influenced by natural seasonal changes compared to the univariate indices, as has been observed by Reiss & Kroncke (2005). The discrepancy in assessing categories between the two indices, may be due to differences in sensitivity in detecting variability of macrobenthic structure over time. For example, while EQR classified the studied sites as 'good'(70%) and 'high'(27.5%), AMBI detected both the control and fallowed sites mostly as 'slightly disturbed', with the exception of P06A (October 2002), RC5E, and P05E (October 2003). Higher categories at the control sites than at the fallowed sites were also observed (assessed by EQR). Considering the scores from other biotic indices, AMBI has the ability to detect large scale differences among sites; however, AMBI is unable to discriminate slight changes in the macrobenthic assemblages between sites. Some 'high' sites, assessed by EQR, are underestimated as 'slightly polluted' sites by AMBI (see **Fig. 3.** and **Table 4**). Because EQR combines species richness, abundance, Simpson's index, and AMBI in one cumulative index (Quintino, *et al.,* 2006), the index seems to be more sensitive than AMBI. Given that macrobenthic abundance significantly varied over time, it is likely that AMBI is not influenced by the changes of taxa abundance as has been reported by Salas (2004). Results from the MDS plot (**Fig. 1.**), suggests that AMBI was not influenced by any biotic index or quantitative variables (abundance, taxa richness, and biomass).

The implication of these results is that the true classification of a site cannot be easily established by a single index. None of the existing indices are suitable for assessing all types of impact, as has been suggested by Dauvin *et al*. (2007) and Marin-Guirao (2005). Ideally, if an index has been tested successfully at areas with different types of pollution, as with the AMBI, it is always possible to choose a single index to categorise a site (Muniz *et al.*, 2005). The AMBI has been successfully verified in a large set of environmental impacts sources, including effects of fish farming (Borja *et al.,* 2003). However, one of the difficulties in assessing environmental disturbance is the inconsistency between biotic indices, as in this study for AMBI and EQR. To justify a biotic index as an estimate of the environmental

impact of a site correctly, other indices, such as univariate indices, are needed for comparisons. For that reason, Borja (2004) recommended the use of a combination of a multimetric index (such as AMBI) and univariate indices (such as species richness and diversity), to obtain a comprehensive assessment. Other difficulties in assessing environmental impact using biotic indices that have been a concern for several researchers include: an appropriate index, sensitivity to dominance species, practical problems in classifying species as indicators, differing seasonal response of biotic indices caused by natural and human-induced disturbance, and applicability to different biogeographic regions, different level of biodiversity, and different source of pollution (Dauvin *et al*., 2007; Simboura, 2004; Dale, 2001; Marin-Guirao *et al*., 2005; Reiss and Kroncke, 2005). Furthermore, Quintino *et al*. (2006) suggested that because integrative indicators are highly dependent on the Pearson–Rosenberg model for organic enrichment, further testing and validation using other stressors, such as physical disturbance and chemical pollution, are needed. Despite the need for further evaluations, in general, multimetric indices seem to be sensitive, stable, and robust (Diaz, 2004), and thus seem to be a promising approach for ecological impact assessment (Reiss and Kroncke, 2005).

Based on the increased variability among samples, the Index of Multivariate Dispersion (IMD) is one of the indicative features of disturbed communities. The index is an expression of multivariate variance among samples that quantifies the differences in relative faunal variability between two groups of samples (impacted and control samples) (Wlodarska-Kowalczuk, 2005; Clarke and Warwick, 2001).

The result for IMD showed that the dispersion between the control and fallowed sites at both Boston Island and Rabbit Island was generally low, ranging from -0.46 to +0.48, as shown in **Table 5**. This implies that the differences in relative macrobenthic variability between the two sites were small. Dispersion among samples between sites at Rabbit Island was slightly higher compared to Boston Island, except in January 2003, indicating higher variability at both the control and fallowed sites. It is not surprising that the result from IMD showed little difference between the

control and fallowed sites, suggesting no major difference in relative macrobenthic variability, as has been assessed by other methods, particularly the AMBI values and ABC curves. However, the dispersion of sites located at

Rabbit Island assessed by IMD was slightly higher than at Boston Island, indicating a slight increase in variability among samples and thus higher level of disturbance.

Table 5. Index of Multivariate Dispersion (IMD) comparing average ranks of similarities among samples of the fallowed and control sites over the study period.

Sampling time	Zone	IMD (Fallowed compared to Control)
Oct-2002	Boston Island	-0.39
	Rabbit Island	+0.45
Jan-2003	Boston Island	+0.39
	Rabbit Island	-0.05
May-2003	Boston Island	+0.29
	Rabbit Island	+0.36
Jul-2003	Boston Island	-0.46
	Rabbit Island	+0.48
Oct-2003	Boston Island	-0.11
	Rabbit Island	+0.20

CONCLUSION

The environmental categories for the control and fallowed sites of the southern Spencer Gulf SBT farms varied with the biotic indices. In particular, site P06A (October 2002) was classified as 'moderate' or 'moderately polluted' by both AMBI and EQR. This is in accordance with the other biotic indices. Site P01E (October 2003) was classified as 'moderately polluted' only by EQR in concordance with the scores of univariate indices, such as diversity, evenness, and richness indices. Considering the variability between biotic indices in categorising each site, it is considered that a combination of multimetric index and univariate indices provide a better assessment. The categories determined by multimetric indices seem to be in accordance with level of disturbance expressed by the trophic analysis, multivariate and graphical analyses used in this study. The AMBI has the ability to detect large scale differences among sites. However, AMBI was unable to discriminate slight changes in the macrobenthic assemblages between sites, as have been exposed by EQR.

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