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Testing the efficiency of temperate benthic biotic indices in assessing the ecological status of a tropical ecosystem

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ABSTRACT

The objectives of the present study were to evaluate the ecological status of tropical coastal waters using the temperate benthic indices and examine the effect of seasonal variability on the performance of benthic indices. Macrobenthic samples were collected from northwest to southeast coast of India during 2003–2012 and we tested different univariate indices, ecological strategies, indicator species and multimetric indices. AMBI and multimetric indices performed satisfactorily in evaluating the ecological status. Seasonal variability on the biotic indices was observed during the southwest monsoon and fall intermonsoon period due to recruitment. Therefore, we recommended the non-monsoon period (January–May) as a suitable time of the year to use the indices for effective assessment of the Indian coastal waters. Results show that, the temperate benthic indices are efficient in assessing the tropical environmental status. However, complementary use of different indices is suggested for accurate assessment of the environmental status.

1. Introduction

The coastal waters are subjected to a variety of changes resulting from anthropogenic and natural processes. Anthropogenic activities, such as habitat alteration, sewage discharge and overexploitation of resources have a deleterious effect on the coastal ecosystem (Halpern et al., 2008; Seitz et al., 2013). In addition to the anthropogenic stressors, the coastal ecosystems are also threatened by global climate change (Watson and Albritton, 2001). One of the main problems affecting these areas is the shift in the benthic community (Pearson and Rosenberg, 1978; Weis et al., 2004; Villnäs et al., 2012). Undisturbed systems are often dominated by *K*-selected species (large-body size, long lifespan, slow-growing) while *r*-selected species characterised by small body size, short lifespan, fast growth and variable population size represent a disturbed community (Pianka, 1970; Warwick, 1986). Changes in the community structure at any trophic level will have a cascading effect that, in turn, affects the ecosystem functioning (e.g. Hooper et al., 2005; Frank et al., 2011).

Given the increasing threat to the coastal environment, many countries worldwide have implemented legislation to assess the ecological quality of estuarine and coastal waters (Borja et al., 2008, 2015). Therefore, several methods and indices have been established to assess the impact of increasing pollution in the marine ecosystem (Borja et al., 2015). Benthic fauna have some features that make them potential candidates for evaluating the health of the coastal environment (Pearson and Rosenberg, 1978; McLusky and Elliott, 2004; Dauvin et al., 2010). Further, they play diverse roles in marine ecosystem functioning by being a primary food source for demersal fish and other benthic fauna. They also actively contribute to the marine biogeochemical cycling (Quintino et al., 2006). However, the macrobenthic community shows a spatio-temporal variation, a result of the natural functioning of the aquatic system. The marked change in the macrobenthic community pattern is reflected in the benthic indices that have often been the main criticism of its use (Reiss and Kröncke, 2005; Kröncke and Reiss, 2010). Therefore improving our understanding of the natural dynamics of indices is of primary importance when considering the use of indices for environmental monitoring and assessment (Kröncke and Reiss, 2010).

In this study, we focussed on the Indian coast, one of the fastest developing regions of the world. India, with ~8000 km long coastline, has 1,641,514 km^2 Exclusive Economic Zone (EEZ). Like the other coastal parts of the world, the Indian coast is also affected by natural and anthropogenic pressures. Major environmental stressors in the Indian coastal environment include heavy floods, sedimentation and eutrophication resulting in increase of coastal hypoxic zones. The main anthropogenic pressures affecting the coast are municipal sewage, industrial and aquaculture effluents, ports and harbours, ship building and breaking yards,

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fish processing industries, salt pans, tourist resorts/beaches, solid waste dumping and overexploitation of resources (Central Pollution Control Board, Govt. of India) (http://www.cpcb.nic.in/).

Despite the considerable importance of the coastal system and threats imposed by increasing pollution, few studies have been carried out to assess the ecological status of the coastal waters of India. Some of the indices used are the Polychaete: Amphipod ratio and Abundance Biomass Comparison (ABC) curve (Ingole et al., 2006), Geometric Abundance Class and ABC curve (Ingole et al., 2009); Benthic Opportunistic Polychaete Amphipoda Index (BOPA) (Sukumaran et al., 2014), AMBI (AZTI-Marine Biotic Index) and M-AMBI (multivariate AMBI) (Ganesh et al., 2014; Khan et al., 2014; Sigamani et al., 2014, 2015). Also, like most tropical regions, the Indian coast is influenced by the monsoon system. The monsoon system leads to semi-annual reversal of ocean circulation resulting in two seasonal phytoplankton blooms followed by an organic enrichment of the benthic system (Prasannakumar et al., 2010; Ingole et al., 2014). During the Northeast monsoon (December–February), the convective mixing triggers the phytoplankton bloom while coastal upwelling characterises the Southwest monsoon (June–August); the increased runoff during the monsoon affects the physico-chemical characters of the estuaries. The macrobenthic community showed a marked temporal variation under the influence of rainfall associated changes (Alongi, 1990; Gaonkar et al., 2013; Ingole et al., 2014). In this context, the aim of the present paper is to test the applicability of different benthic indices to assess the ecological quality of the coastal waters of India. Given the strong seasonality in the region, we also examined the effect of seasonal variability on the performance of benthic indices and to identify the most suitable time of year for effective assessment of the ecological status of tropical coastal waters.

2. Methods

2.1. Study area and sampling

2.1.1. Southeast coast

Sampling in the southeast coast of India was carried out at Gulf of Mannar (GoM) and Palk Bay (PB) during March 2010 (Fig. 1 and Table 1). The GoM is located between the southern tip of India and northwest of Sri Lanka and is the first marine biosphere reserve of India. It is a large and relatively deep gulf of the Arabian Sea. On the other hand, the PB is a shallow and enclosed basin with an average depth of 5 m. In the GoM, the main sediment type consists of sand while muddy sediment dominated in the PB. Sampling was carried out at 30 stations distributed along ten transects and three depths (5, 10, 25 m) with 15 locations each in the GoM and PB (Fig. 1 and Table 1).

2.1.2. Southwest coast

Sampling was carried out in the coastal waters of Kochi (KO) along a single transect at six depths (10, 20, 30, 40, 50 and 100 m) from March–December 2012. The samples from 10, 20 30 m stations were pooled as the inner shelf, stations 40 and 50 m as mid shelf stations and the outer shelf was represented by 100 m station. Seasonal sampling was carried out at Mangalore (MG) along a single transect with six stations (10, 20, 30, 40 and 50 m). Sandy substrate dominated the southwest coast stations.

2.1.3. Northwest coast

The northwest coast of India was sampled at three different locations: (1) Goa (GO): Sampling at Goa was conducted in two major estuaries, the Mandovi (MD) and Zuari (ZU) and a single transect in the coastal region. The Mandovi-Zuari estuarine system is considered the life-line of Goa. The samples in the Mandovi estuary (MD) were collected fortnightly in 2007–2009 at a single station (Gaonkar et al., 2013). The Zuari River has a length of ~50 km and the Mormugao port is located on the southern bank of the river mouth. The Mormugao port is one of the oldest and finest major ports along the west coast of India. In the ZU, samples were collected from seven stations during 2003–2004. The first four stations were located outside the harbour (ZU estuary) and three stations (Stn 5–7) in the harbour (ZU harbour). The third sampling location in Goa was a single transect, in the coastal waters sampled during November 2009. Samples were collected from six depths (10–60 m) (2) Ratnagiri: Kalbadevi Bay (KB) is a potential placer mineral (Ilmenite) mining site along the northwest coast of India and is a relatively undisturbed area. Samples were collected from two transects with three stations (2, 5 and 10 m) each during January, May and September 2006 (Sivadas et al., 2013). Coastal samples were collected from off Ratnagiri (RT coast) during November 2009 and March 2011 at a depth of 10–50 m (five stations) (Ingole et al., 2014). (3) Mumbai (MU): The northernmost area included in the present study was the coastal region of Mumbai (Ingole et al., 2014). Sampling was carried out during November 2009 along a single transect with five stations at depth of 10–50 m. In addition, data from Ratnagiri (RT) and Karwar (KR) harbour were also used (Ingole et al., 2009). All the stations at Goa, Ratnagiri and Mumbai, were pooled based on depth as mentioned for Kochi. Clayey-silt dominated the sediment type in the northwest stations, except for KB and the ZU estuary which were dominated by sand.

2.2. Environmental data

Surface and bottom water samples were collected for temperature, salinity, pH, dissolved oxygen, biological oxygen demand, total suspended solids, nutrients, Petroleum Hydrocarbon Carbon (PHC), metals (Fe, Mn, Pb, Hg, and Cd), chlorophyll and phaeopigments. Sediment was collected for sediment texture, organic carbon, carbohydrates, proteins, lipids, PHC, metals (Fe, Mn, Pb, Hg, and Cd), chlorophyll and phaeopigments.

2.3. Biological variables

The uneven distribution of stations and seasons at each location was according to the objectives of the different projects during which the samples were collected. All the sediment samples were collected from the subtidal region using a Van Veen grab (0.04 or 0.1 m^2). Samples were sieved on a 500 μm sieve and preserved in 4% buffered formaldehyde with Rose Bengal solution. In the laboratory, the macrofaunal samples were analysed for abundance (ind m^{-2}), biomass (wet weight; g m^{-2}) and species composition. We identified the species to the lowest possible taxa. The samples were pooled to obtain a sample size of 0.1 m^2 for stations where the grab size was <0.1 m^2 (Borja et al., 2007).

2.4. Data analysis

We grouped the data into monsoon and intermonsoon seasons relevant to the Indian Ocean, i.e., December to February (Northeast Monsoon; NEM), March to May (Spring Intermonsoon; SIM), June–August (Southwest Monsoon; SWM) and September to November (Fall Intermonsoon; FIM).

2.4.1. Indices based on diversity

The classical univariate indices such as species abundance (N), species number (S), Margalef's species richness (d), Pielou's species evenness (J), Shannon species diversity (H′ log2) and Simpson's Evenness Index (1-λ′) were calculated. We also used the Hurlbert Index (ESn) which is based on the rarefaction technique of Sanders (1968) and modified by Hurlbert (1971). The Hurlbert Index is less sample size dependent compared to the evenness and species diversity. The Hurlbert Index calculates the expected number of species (ES) among a certain number of individuals. In the present study, we estimated ES for 50 individuals (ES50). The indices based on relatedness such as quantitative taxonomic diversity (Δ), taxonomic distinctness (Δ^*), qualitative Average Taxonomic Distinctness (Δ^+) and Variation Taxonomic distinctness (Λ^+) were also calculated. We calculated the indices for all the replicates from each station and season using Primer v 6.1.10 software (Clarke and Gorley, 2006).

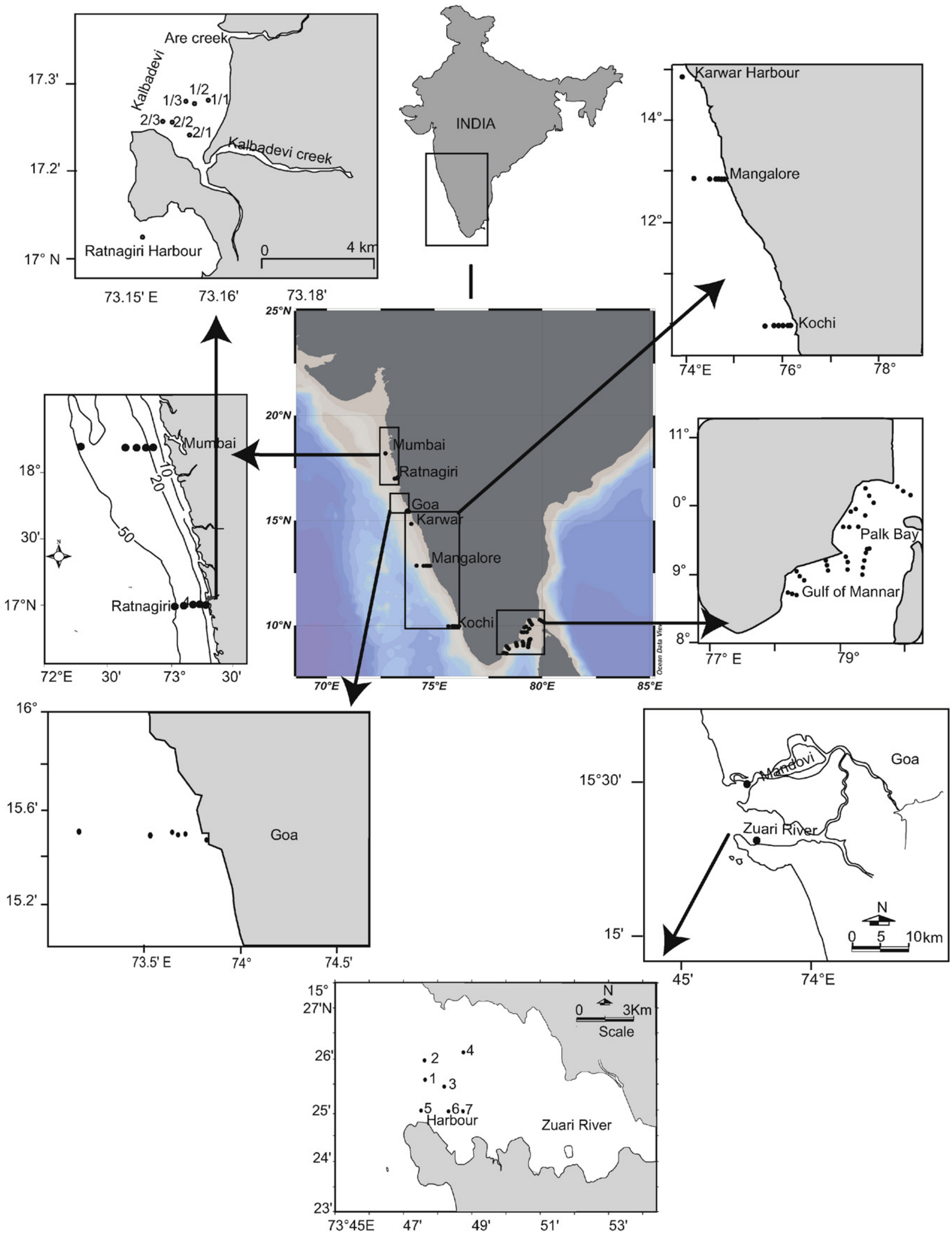

Fig. 1. Map of the study area showing location of the sampling stations.

Table 1
Sampling details in the study area. NEM Northeast Monsoon; SIM Spring Intermonsoon; SWM Southwest Monsoon; FIM Fall Intermonsoon.

Sr. no.	Location	No. of transect/station	Depth (m)	Replicates (n)	Sampling period	References
1	Gulf of Mannar (GoM)	5/15	5–25	2–3	SIM (2010)	Present study
2	Palk Bay (PB)	5/15	5–25	2–3	SIM (2010)	Present study
3	Kochi (KO)	1/6	10–100	1–2	Monthly (Mar-Dec. 2012)	Present study
4	Mangalore (MG)	1/6	10–100	1–2	SIM, FIM & NEM (2012)	Present study
5	Karwar harbour (KR)	2 stns.		5	NEM (2006)	Ingole et al., 2009
6	Goa (GO)	1/5	10–60	4–6	FIM (2009)	Present study
7	Mandovi estuary (MD)	1 stns	8	10	Fortnightly (2007–09)	Gaonkar et al., 2013
8	ZUARI (ZU)				Monthly (2003–04)	Sivadas et al., 2011
	Zuari estuary	4 stns	8–15	2–3		
	Zuari harbour	3 stns	8–15	2–3		
9	Kalbadevi Bay (KB)	3/3	2–10	2–3	NEM, SIM & FIM (2006)	Sivadas et al., 2013
10	Ratnagiri (RT coast)	1/5	10–50	3–6	FIM (2009) & SIM (2011)	Ingole et al. 2014
11	Ratnagiri harbour (RT)	2 stns		10–15	NEM (2006)	Ingole et al., 2009
12	Mumbai (MU)	1/5	10–50	3	FIM (2009)	Ingole et al. 2014

2.4.2. Indices based on species abundance and biomass

We tested two indices based on the abundance and biomass in the present study. The geometric abundance curve is based on the distribution of abundance within different classes. Species represented by a single individual represent (class 1), 2–3 individuals in (class 2), 4–7 (class 3), 8–15 (class 4) and so on. In the unpolluted situation, the community is represented by rare species resulting in a very steep curve as species extend over very few abundance classes. In polluted conditions, the curve becomes flatter and extends across many abundance classes as few species dominated the community. ABC (Abundance Biomass Comparison) curves proposed by Warwick (1986) were used to investigate community perturbation. The W statistic involves summing the differences between the ranked cumulative percentages of abundance and biomass values (Warwick and Clarke, 1994). In this statistic, W takes values in the range (−1, 1), +1 value for even abundance across species, but biomass dominated by a single species, which is an undisturbed community, and −1 in the converse case. The ABC curve and Geometric class abundance analyses were carried out using the PRIMER v 6.1.10 software.

2.4.3. Indices based on ecological strategies

We used the benthic opportunistic polychaete amphipod index (BOPA; Dauvin and Ruellet, 2007) and polychaete amphipod ratio (PA; Gesteira and Dauvin, 2000) in the present study. The Infaunal Trophic Index (ITI) is based on the feeding strategies (Word, 1978), where ITI values near 100 mean that the macrobenthic community is dominated by suspension feeders indicating an undisturbed condition. A value near 0 indicates the dominance of subsurface feeders and that the environment has been strongly disturbed.

2.4.4. Index based on indicator species

The AZTI-Marine Biotic Index (AMBI) is based on the distribution of the abundance of benthic communities into five ecological groups (Grall and Glémarec, 1997). The species were classified into ecological groups (EG) based on their sensitivity along an increasing gradient of organic enrichment (Pearson and Rosenberg, 1978; Grall and Glémarec, 1997):

- EG I: species very sensitive to organic enrichment and present in unpolluted conditions, which include specialist carnivores and some deposit-feeding tubiculous polychaetes;
- EG II: species which are indifferent to organic enrichment and always present in low densities without non-significant variation with time. These include suspension feeders, less selective carnivores and scavengers;
- EG III: species which are tolerant to excess organic matter enrichment and may occur under normal conditions, but their populations are stimulated under increasing organic loads in sediments. This group includes some surface deposit feeding species;
- EG IV: second-order opportunistic species which have a small body size, short life cycle and proliferate in reduced sediments. They are subsurface deposit feeders;
- EG V: first-order opportunistic species which are deposit feeders abundant in sediments which are close to anoxic.

The AMBI was calculated using the AMBI software available on the AZTI site (http://www.azti.es) (Borja et al., 2000). For taxa not determined to species level, the information was deduced from closely related species. Species, for which the classification was not in accordance with the AZTI species list, were re-assigned to an EG based on expert advice, and from our own experience. About ~5–10% of the taxa could not be assigned to any category and these species contributed to <1% of the total abundance.

2.4.5. Multimetric indices

For the multivariate AMBI (M-AMBI), the AMBI, Shannon diversity and species richness are combined and used in a factor analysis (Muxika et al., 2007). Here, we used two methods that determined the reference condition for evaluating the M-AMBI index; (1) the lowest AMBI and highest species richness and diversity values of all the replicates, and (2) increasing the highest species richness and diversity of all replicates by 15% (Borja et al., 2012; Cai et al., 2014). We also used the Danish Index (DKI), Benthic Ecosystem Quality Index 2 (BEQI 2), Norwegian Quality Index (NQI), and the Infaunal Quality Index (IQI) (Borja et al., 2007; Perus et al., 2007; Kröncke and Reiss, 2010; van Loon et al., 2015). We used the highest values of species richness and diversity within the entire dataset as a reference condition (Borja et al., 2011) for each location, except for the harbours. For ZU harbour, the ZU estuary was considered as the reference. While the reference values for KR and RT harbour were obtained from the inner shelf regions of GO to MU as the environmental conditions were comparable.

2.5. Statistical analyses

The assessment of the performance of the biotic indices was tested using the intraclass correlation (ICC) (McGraw and Wong, 1996). The ICC was analysed using a two-way random, consistency, average-measures and used the cut-offs provided by Cicchetti (1994) for qualitative ratings of the agreement. Pearson's correlation was used to assess the relation between the indices and environmental variables. The correlation was carried out at each sampling site and for the overall data. However, when $N > 20$, the α (0.05) value was modified after Bonferroni correction to α' (i.e., α/N) (Seber, 1984). The statistical analyses were carried using the SPSS software package. Sediment composition, grain size, and sorting was analysed using Gradistat 5 (Blott and Pye, 2001).

3. Results

3.1. Indices based on diversity

The macrofaunal species number ranged from 30 to 204 taxa, recorded at KR harbour and KO, respectively (Fig. 2a). The macrofaunal abundance was highest in the Northwest stations and ranged from 667 to 46,329 ind m^{-2} (Table 2). The Shannon diversity indices (H') were lowest in the stations located on the northwest coast (ZU, RT and MU) while highest values were recorded at KO (mid and outer shelf; Fig. 2b). The Hurlbert Index (ES50) followed a trend very similar to the Shannon diversity index, showing highest values in the southern area of KO, MG, GoM, and KB (Fig. 2c). The lowest values (<10) were observed on the GO, RT coast, MU and the harbours (RT, ZU and KR). The average Pielou's and Simpson's indices showed values >0.5 in all cases, except for MU (0.11–0.48).

The range value of Δ and Δ^* in the study are given in Table 2. The comparison of samples with the master list showed that the average taxonomic distinctness (Δ^+) for most of the stations were within the 95% confidence funnel, except for Stn 7 at the ZU harbour. In the same way, the variation taxonomic distinctness (Λ^+) of the stations was within the funnel, but below the mean value. In general, Δ^+ and $\Lambda\Lambda+$ were within the 95% confidence funnel for most stations; however, most values were below the expected mean.

3.2. Indices based species abundance and biomass

The ABC and W statistics indicated that almost the entire study area was moderately disturbed. The W statistic varied very close to zero for most of the study area. Although the biomass line was above the abundance line for some locations, the W-statistic was low, indicating a moderate disturbance. The geometric abundance and size class ranged from 7 to 15. The lowest classes were observed in the GoM, PB and outer shelf region of KO and MG. The highest class were seen for the inner and middle shelf region of RT (14 classes) and MU (15 classes), respectively.

3.3. Indices based on ecological strategies

The PA ratio ranged from 0.78 to 3.9 (Table 2). High values of BOPA were observed in the northwest coast locations (GO, RT, MU) both the estuaries (MD and ZU) and the three harbours (Fig. 2d). Low values

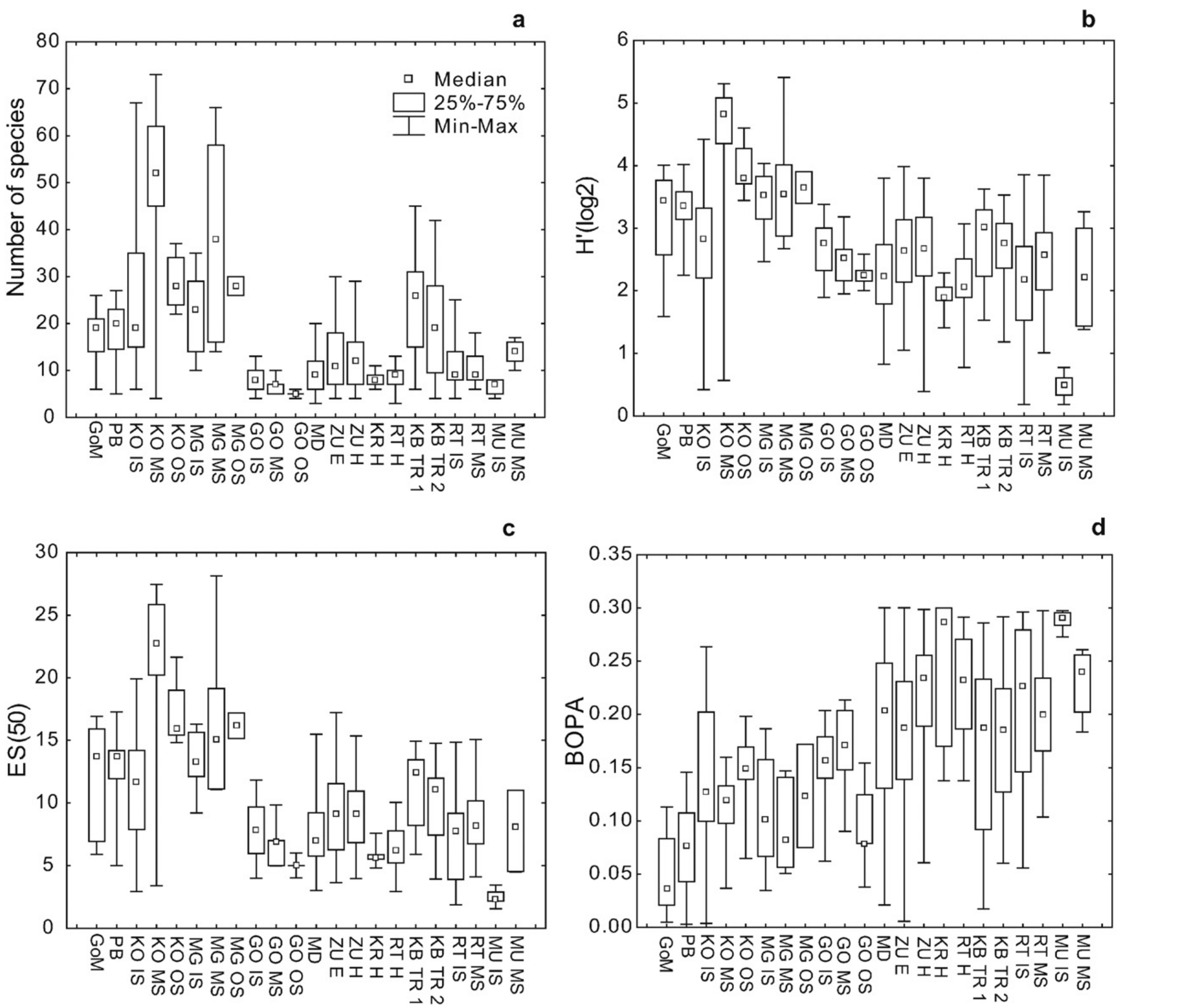

Fig. 2. Box-whisker plot for (a) number of species (b) Shannon diversity index H′ (c) ES (50) (d) BOPA. (GoM Gulf of Mannar; PB Palk Bay; KO Kochi; Mangalore MG; GO Goa; MD Mandovi; ZU E Zuari estuary; ZU H Zuari harbour; Karwar harbour KR H; Ratnagiri harbour RT H; KB Kalbadevi; MU Mumbai; IS inner shelf; MS mid shelf; OS outer shelf; TR Transect).

Table 2
Range of the univariate and ecological strategy indices in the study area (GoM Gulf of Mannar; PB Palk Bay; KO Kochi; MG Mangalore; GO Goa; MD Mandovi; ZU E Zuari estuary; ZU H Zuari harbour; KA H Karwar harbour; RT H Ratnagiri harbour; KB TR Kabadevi transect; MU Mumbai; IS Inner shelf; MS mid shelf; OS outer shelf. N abundance; d species richness; J′ species evenness; Simpson's Evenness Index (1-λ′) quantitative taxonomic diversity (Δ), taxonomic distinctness (Δ*), qualitative Average Taxonomic Distinctness (Δ+) and Variation Taxonomic distinctness (Λ+).

	N	d	J′	ES(50)	H′(log2)	1-λ′	Δ	Δ*	Δ^+	Λ^+	ITI	PA ratio
GoM	104–1258	1.07–3.5	0.39–0.91	6–17	1.58–4.0	0.52–0.92	42–79	60–95	73–85	344–768	14–86	0.95–2.6
PB	30–1445	1.18–4	0.66–0.97	5–17	2.25–4	0.75–0.91	55–76	73–89	78–87	281–437	2–70	0.41–2.9
KO IS	260–17,570	0.86–7	0.09–0.93	2.9–20	0.42–4.42	0.08–0.94	18–78	75–98	73–86	334–572	55–100	0.70–4.2
KO MS	340–5130	0.51–8.6	0.28–0.89	3.4–27	0.57–5.3	0.16–0.96	59–78	72–99	79–91	54–572	54.5–100	0.29–1.53
KO OS	545–2320	3.17–5	0.73–0.9	15–21	3.4–4.6	0.82–0.94	20–75	38–93	38–84	371–500	57–100	1.24–3.12
MG IS	26–12,270	1.6–3.6	0.69–0.89	9–16	2.4–4	0.71–0.93	15–53	38–100	43–100	78–555	44–89	0.59–3.2
MG MS	450–7510	2.12–8.3	0.59–0.89	11–28	2.67–5.4	0.7–0.96	13–72	25–100	25–100	555	55–100	0.15–2.5
MG OS	760–1040	3.7–4.17	0.72–0.79	15–17	3.39–3.9	0.80–0.89	28–55	75–81	75–86	391	77–99	1.83–1.9
GO IS	278–1667	0.5–1.62	0.85–1	4–12	1.89–3.37	0.72–0.88	49.5–72	63–85	64.2–86	86.8–559	37–77	1–3.14
GO MS	278–889	0.6–1.35	0.82–1	5–10	1.95–3.18	0.69–0.87	46–74	58–86	57.5–88	113–446	28–81	1.07–2.8
GO OS	222–444	0.5–0.86	0.92–1	4–6	2–2.58	0.75–0.83	66–72	85–90	85–90	150–250	45–67	2.6–2.64
MD	88–6950	0.41–2.5	0.28–1	3.7–16	0.82–3.8	0.23–0.9	10–70	25–100	25–100	139–1406	16–100	0.30–3.8
ZU E	88–14,447	0.46–3.5	0.42–1	3.6–17	1.05–4	0.37–0.91	25–76	50–96	50–92	131–872	3.34–100	2.33–0.86
ZU H	110–26,751	0.53–2.8	0.41–0.97	3.9–15	0.39–3.8	0.56–0.91	75–48	57–93	67–92	87–572	11–100	0.89–4.37
KA H	2797–8880	0.61–1.2	0.47–0.79	4.8–7.6	1.40–2.2	0.48–0.73	34–54	70–80	77–86	332–539	101–100	1.4–3.5
RT H	888–21,578	0.35–1.5	0.43–0.9	3.8–10	1.09–3.07	0.39–0.85	31–69	72–85	80–88	156–517	54–89	0.77–4.33
KB TR 1	800–35,100	0.7–4.22	0.32–0.86	5.8–15	1.53–3.63	0.4–0.47	15–54	72–87	67–92	139–619	23–72	1.29–5.5
KB TR 2	800–32,500	0.44–3.9	0.47–0.9	4–15	1.2–3.5	0.4–0.88	20–68	67–91	58–92	139–555	33–100	1.5–4.38
RT IS	944–30,386	0.3–2.54	0.09–0.92	1.88–15	0.18–3.86	0.04–0.92	1–72	50–89	50–87	38–589	63–99	0.72–4.48
RT MS	667–7388	0.8–2.14	0.34–0.95	4.11–15	1–3.8	0.33–0.91	24–74	63–87	68–85	106–545	63–92	0.72–3.8
MU IS	7610–46,329	0.3–0.66	0.04–0.28	1.5–3.4	0.18–0.78	0.02–0.22	1.58–16	52–76	70–86	100–367	67–74	1.54–5
MU MS	5499–13,999	097–0.88	0.21–0.82	4.4–11	1.3–3.27	0.18–0.85	13.8–60	63–79	79–84	327–466	54–64	1.4–4

were observed in GoM and PB. On average, the ITI value ranged from 11 to 94 with the lowest value recorded at ZU harbour.

3.4. Index based on indicator species

A wide range of values were observed for AMBI reflecting 'good' to 'poor' ecological status for the coastal and estuarine waters of India. The AMBI values were high in the harbours, Mandovi estuary and coastal waters of RT and MU (Fig. 3a-c). Based on the index, the undisturbed locations (GoM, PB and KB) had the lowest values (Fig. 3a). In GO, no clear spatial gradient was observed and showed 'good' status in all the stations. The AMBI value classified MU as having a 'moderate' status, with inner shelf (IS) having higher values than the mid shelf (Fig. 3a). Kochi and MG maintained the same status (good) throughout the sampling period (Fig. 3b). Similarly, no temporal variation in the AMBI indices was observed in KB (Fig. 3b). The seasonal pattern of AMBI values in the MD estuary fluctuated from 'good' to 'moderate' with highest value recorded during FIM 2007 (Fig. 3b). The AMBI value in the coastal stations of RT showed 'good' status during SIM and the inner shelf was classified as 'moderate' during FIM (Fig. 3b). When the AMBI values were compared between the three harbours, the highest value was related to the highest activity of the harbour i.e. ZU > KR > RT. The ZU estuary was categorised as 'good' by AMBI during all the seasons. For the ZU harbour, the AMBI index showed 'good' status for all the three stations during FIM and SWM (Stn 6; Fig. 3c). During rest of the seasons, they were classified as 'moderate' with the maximum AMBI value found in Stn 5, located in the approach channel (Fig. 3c).

The most abundant species and the ecological group are presented in Table suppl. 1. The EG I and II dominated the community at GoM and PB (Fig. 4a). The EG IV dominated at RT and KR harbour and MU (Fig. 4a). The EG I and III dominated KO during different seasons. Further, EG V showed an increase from inner shelf to outer shelf with highest values during NEM (Fig. 4b). A similar trend of dominance of EG I and III, and, the spatial gradient in EG V was observed in MG. In MD and RT, the community was dominated by EG III and IV (Fig. 4b). The EG II and III dominated in KB during NEM and SIM, and in FIM, the community was represented by EG IV (Fig. 4b). The ZU harbour was dominated by EG IV during NEM to SWM (Fig. 4c). At ZU estuary, the indifferent species (EG II) and pollution tolerant species (EG III) were important. The EG III was represented by *Magelona* sp. in MG and KO while the EG IV (*Paraprionospio* sp.) was the most dominant species from GO to MU. The macrofaunal community in KB was dominated by *Jasmineira* sp. (EG II) and *Mediomastus* sp. (EG III).

3.5. Multimetric indices

Two methods were used for setting the reference condition for M-AMBI for each sampling site, as mentioned in the methods section. Although the M-AMBI values obtained from the two methods were different, they were within the same ecological status. The pattern of M-AMBI was similar to that of AMBI but differed concerning the classification of the ecological status in some stations. The AMBI classified some stations as 'high'/'moderate' when M-AMBI classified those stations as 'good'/'poor'. In terms of ecological status, the M-AMBI classified GoM, PB, KO (mid shelf), GO, KB as 'good' status (Fig. 5a-c). The inner and outer shelf of KO, MG IS, and OS (NEM), ZU estuary, RT coast KB (FIM) were classified as 'moderate' (Fig. 5b) when AMBI indicated a 'good' ecological status (Fig. 3b). The MD estuary in 2009, RT and Karwar harbours were categorised as 'poor' by M-AMBI, and 'moderate' by AMBI (Fig. 3b). The ZU estuary was classified as 'good' by AMBI (Fig. 3a); however, M-AMBI indicated a 'moderate' to 'poor' ecological status (Fig. 5c). The effect of seasonality on the M-AMBI index was seen in KO (IS), MG (OS), MD estuary (2009) ZU estuary and harbour and RT (IS; Fig. 5b).

On the other hand, IQI, DKI, NQI and BEQI2 were similar but differed with regard to the classification of the ecological status at some stations (Figs. 5a and 6a–c). These indices agreed in assigning a similar ecological status ('good') to GoM, PB, KO (mid and outer shelf) and GO and a 'moderate' status to MD estuary and harbours (ZU, KR and the RT). The IQI index classified KB as 'moderate' (SIM and NEM) and 'poor' during FIM (Fig. 5b), when other indices showed a 'moderate' status only during FIM. Similarly, MU was categorised as 'good' (IS) by IQI (Fig. 5a), and 'moderate' by other multimetric indices. The ZU estuary was classified as 'moderate' by IQI, DKI and NQI during SWM. The classification of the ZU estuary by BEQI2 was similar to M-AMBI and showed a 'moderate' to 'poor' ecological status. The southwestern stations (KO and MG) were classified by AMBI as undisturbed (Fig. 3a), however, mutlimetric indices indicates a moderately disturbed status for the inner shelf during some seasons (Figs. 5b, 6a and c, 7a and c).

In general, the effect of seasonality on the multimetric indices was observed for the estuarine locations (ZU and MD) and the coastal

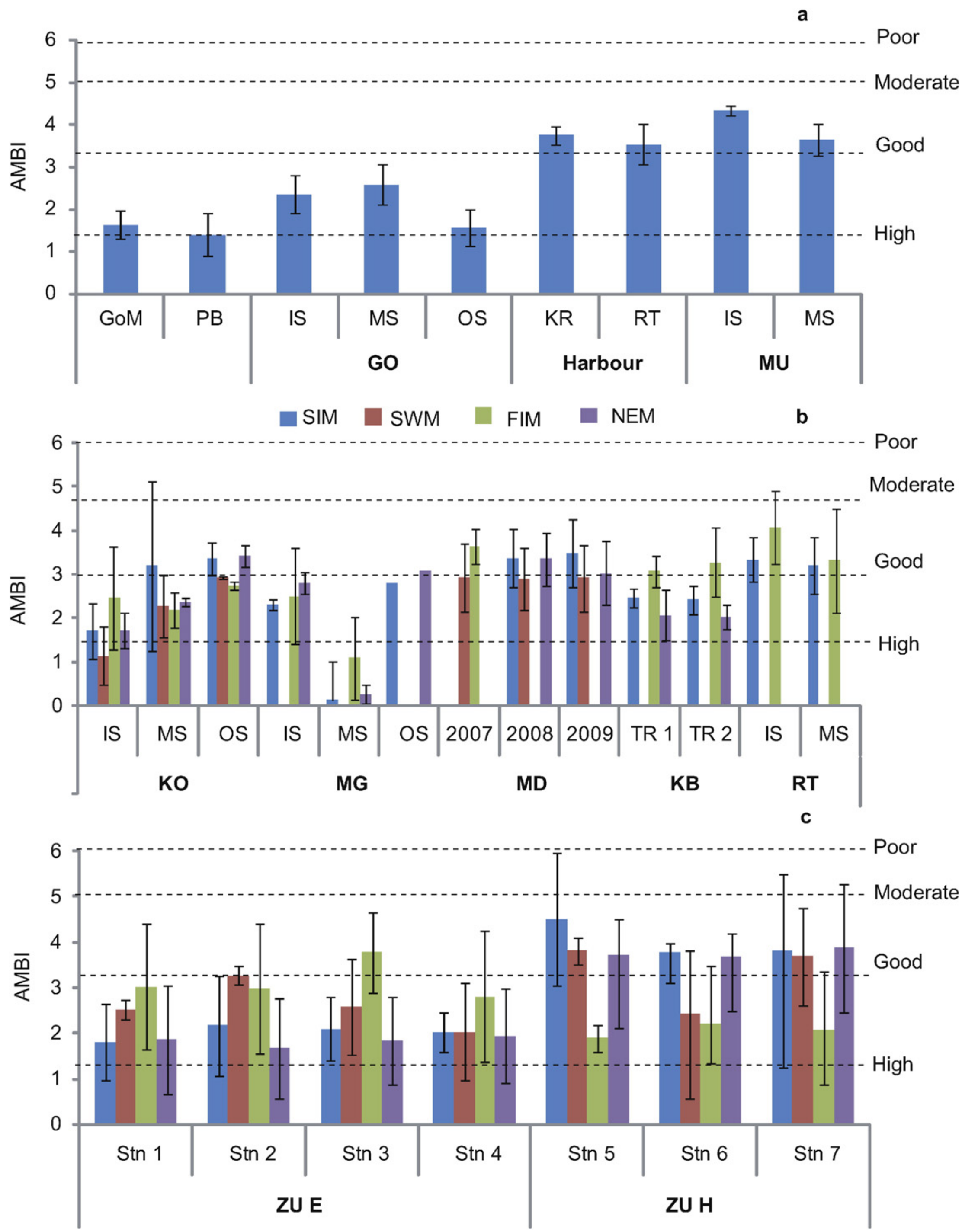

Fig. 3. Seasonal variation in AMBI at (a) Gulf of Mannar (GoM), Palk Bay(PB), GO (Goa), Karwar harbour (KR), Ratnagiri harbour (RT), MU (Mumbai); (b) Kochi (KO), Mangalore (MG), Mandovi (MD), Kalbadevi (KB) and Ratnagiri (RT) and (c) Zuari estuary (ZU E) and harbour (ZU H). (NEM Northeast Monsoon; SIM Spring Intermonsoon; SWM Southwest Monsoon; FIM Fall Intermonsoon; IS inner shelf; MS mid shelf; OS outer shelf; TR Transect).

stations (inner shelf). Within the estuaries including the ZU harbour stations, the multimetric indices showed a 'good' status during the FIM. The values dropped to 'moderate' or 'poor' status during the SIM. In KO, the inner shelf changed from 'good' to 'moderate' or 'poor', while the mid and outer shelf maintained the same status across the season. A similar observation was made in the inner shelf of RT, M-AMBI shifted from 'moderate' to 'poor' (Fig. 5b), IQI (Fig. 6a) showed a shift from 'good' to 'moderate' and DKI classification changed from 'moderate' to 'poor' (Fig. 6c). The NQI and BEQI2 indices maintained the same status between the two sampling seasons (Fig. 7a and c). In MG, although the values fluctuate along the season, they remained within the same ecological status.

To evaluate the ecological statuses classified by different indices were the same, we used the intraclass correlation analysis (ICC). The ICC was analysed for BOPA, AMBI, M-AMBI, IQI, DKI, NQI and BEQI2 (Table 3). The ICC analysis indicates an excellent level of agreement

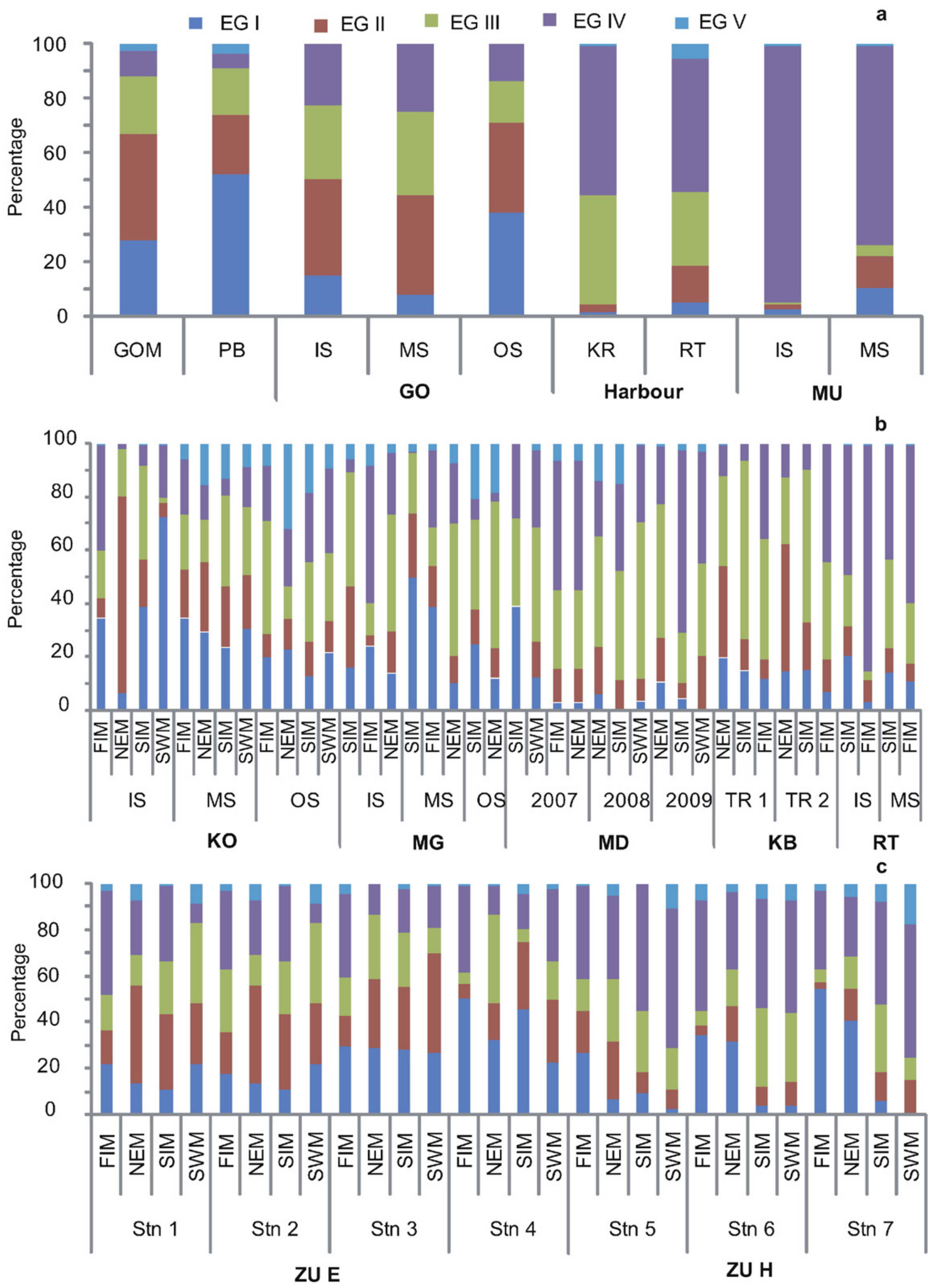

Fig. 4. Percentage composition of ecological groups (EG) (a) Gulf of Mannar (GoM), Palk Bay(PB), Goa (GO), Karwar harbour (KR), Ratnagiri harbour (RT), MU (Mumbai); Kochi (KO), Mangalore (MG), Mandovi (MD), Kalbadevi (KB) and Ratnagiri (RT) and (c) Zuari estuary (ZU E) and harbour (ZU H). (NEM Northeast Monsoon; SIM Spring Intermonsoon; SWM Southwest Monsoon; FIM Fall Intermonsoon; IS inner shelf; MS mid shelf; OS outer shelf; TR Transect).

between the indices ($r = 0.85$; $p < 0.001$). The highest correlations were observed among IQI, DKI, NQI and BEQI2.

3.6. Pearson's correlation analyses

Pearson's correlation between environmental parameters and benthic indices on the overall data did not show any significant relation. However, when the analysis was carried out at each sampling site, a significant correlation was noticed between the biotic indices and environmental parameters. In Kochi, species number, richness, ES (50), H′, M-AMBI, IQI and NQI showed a significant negative relation with bottom water chlorophyll and phaeopigments ($p < 0.001$; Table suppl. 2). The Infaunal Quality Index (IQI) and NQI, also showed a positive correlation with nitrate and negative with nitrite and temperature ($p < 0.001$). In

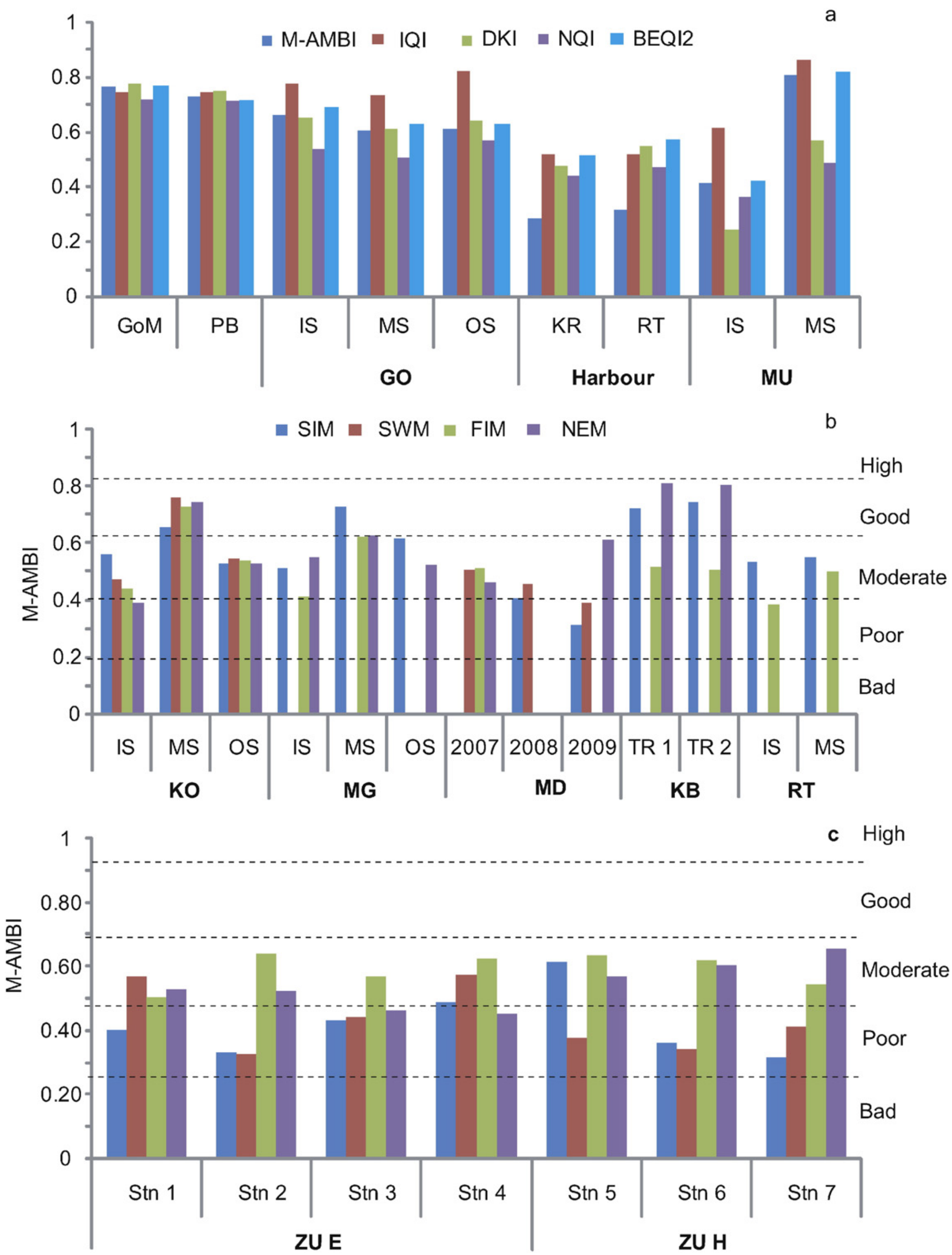

Fig. 5. Seasonal variation in multimetric indices (a) Gulf of Mannar (GoM), Palk Bay(PB), Goa (Goa), Karwar harbour (KR) Ratnagiri harbour (RT), MU (Mumbai); (b) seasonal variation in M-AMBI (a) Kochi (KO), Mangalore (MG), Mandovi (MD), Kalbadevi (KB) and Ratnagiri (RT) and (c) Zuari estuary (ZU E) and harbour (ZU H). (NEM Northeast Monsoon; SIM Spring Intermonsoon; SWM Southwest Monsoon; FIM Fall Intermonsoon; IS inner shelf; MS mid shelf; OS outer shelf; TR Transect).

Zuari, the correlation was observed between the biotic indices and some of the physicochemical parameters (Table Suppl. 1). For example, salinity showed a positive correlation with IQI ($r = 0.43$; $p = 0.01$) and NQI ($r = 0.45$; $p = 0.01$). Dissolved oxygen was positively correlated with Δ ($r = 0.55$; $p < 0.001$) and Δ^* ($r = 0.55$; $p < 0.001$) while AMBI showed positive correlation with sediment Hg ($r = 0.43$; $p = 0.001.001$). A significant positive correlation between organic matter and species number, abundance, ES(50), M-AMBI, IQI, DKI, NQI and BEQI 2 were observed at KB ($p < 0.001$).

4. Discussion

The main objective of the present study was to test the applicability of selected benthic indices along the coastal waters of India and understand their natural variability due to the monsoon associated changes in the benthic community. The present study is the first that tested various benthic indices for assessing the coastal and estuarine waters of India. The indices were tested in different habitats and seasons and from relatively undisturbed to disturbed areas (naturally and anthropogenically).

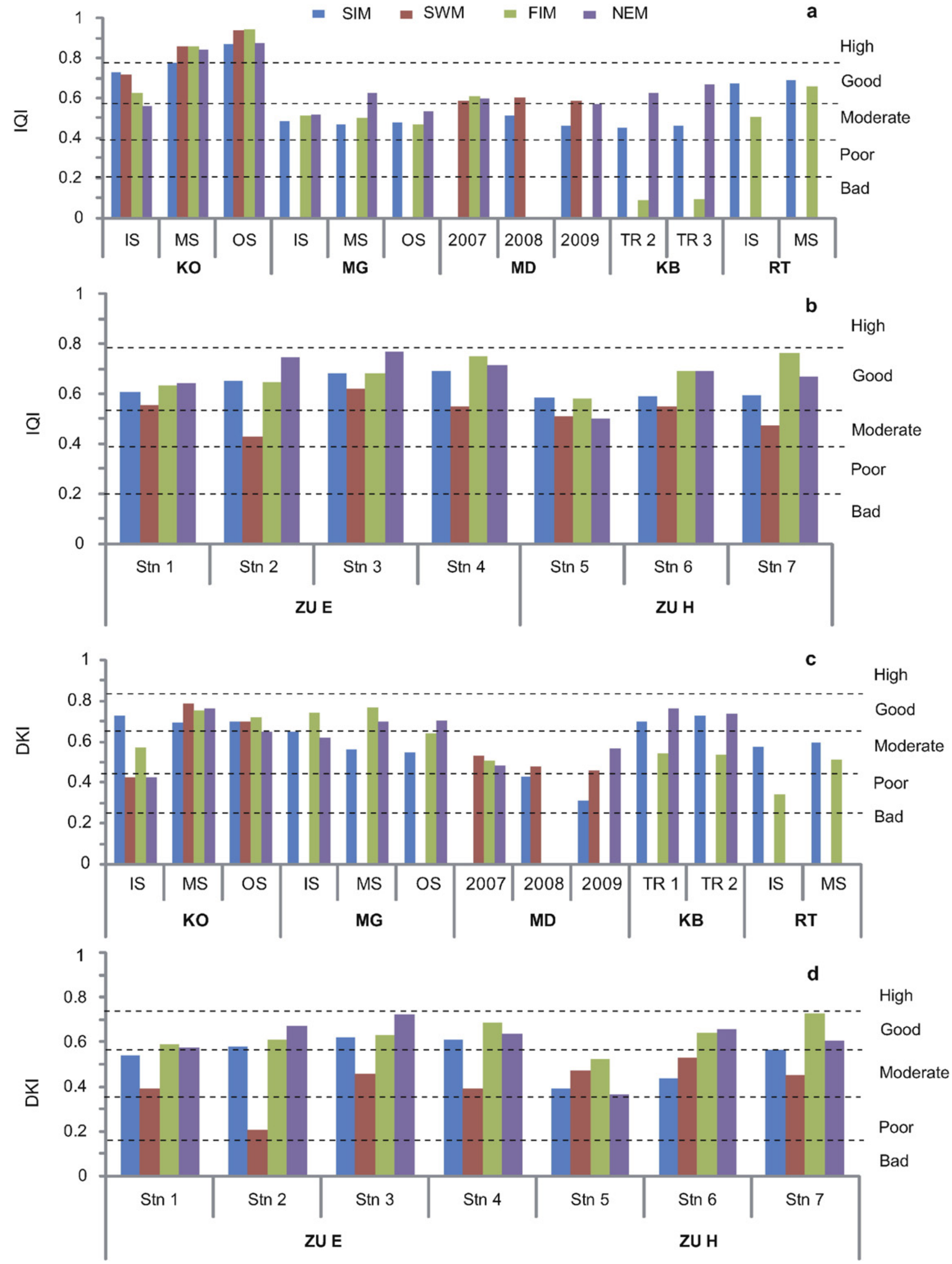

Fig. 6. Seasonal variation in IQI (a–b) and DKI (c–d) at Kochi (KO), Mangalore (MG), Mandovi (MD), Kalbadevi (KB) and Ratnagiri (RT), Zuari estuary (ZU E) and harbour (ZU H). (NEM Northeast Monsoon; SIM Spring Intermonsoon; SWM Southwest Monsoon; FIM Fall Intermonsoon; IS inner shelf; MS mid shelf; OS outer shelf; TR Transect).

4.1. Ecological status of the Indian coast

In general, the univariate indices show variation among the studied sites. Species richness, H′ and ES (50) were higher in the southern stations (KO mid shelf), and remarkably higher abundance was recorded in the northwest sites (RT and MU). The high abundance at the northwest stations was the result of the organic enrichment of the sediment during the FIM period (Ingole et al., 2014). Despite the fact that the EG I and II dominated the macrobenthic community at GoM and PB, the H′ values indicate disturbed conditions. One reason for this is that some species may occur naturally in relatively high densities and, hence, affect the indices values. And high macrofaunal abundance is associated with low evenness and diversity values (Pearson and Rosenberg, 1978). Also, since H′ does not consider the type of species (sensitive/tolerant), it makes this index difficult to compare areas with different biodiversity levels (Zettler et al., 2007). The phylogenetic diversity was also

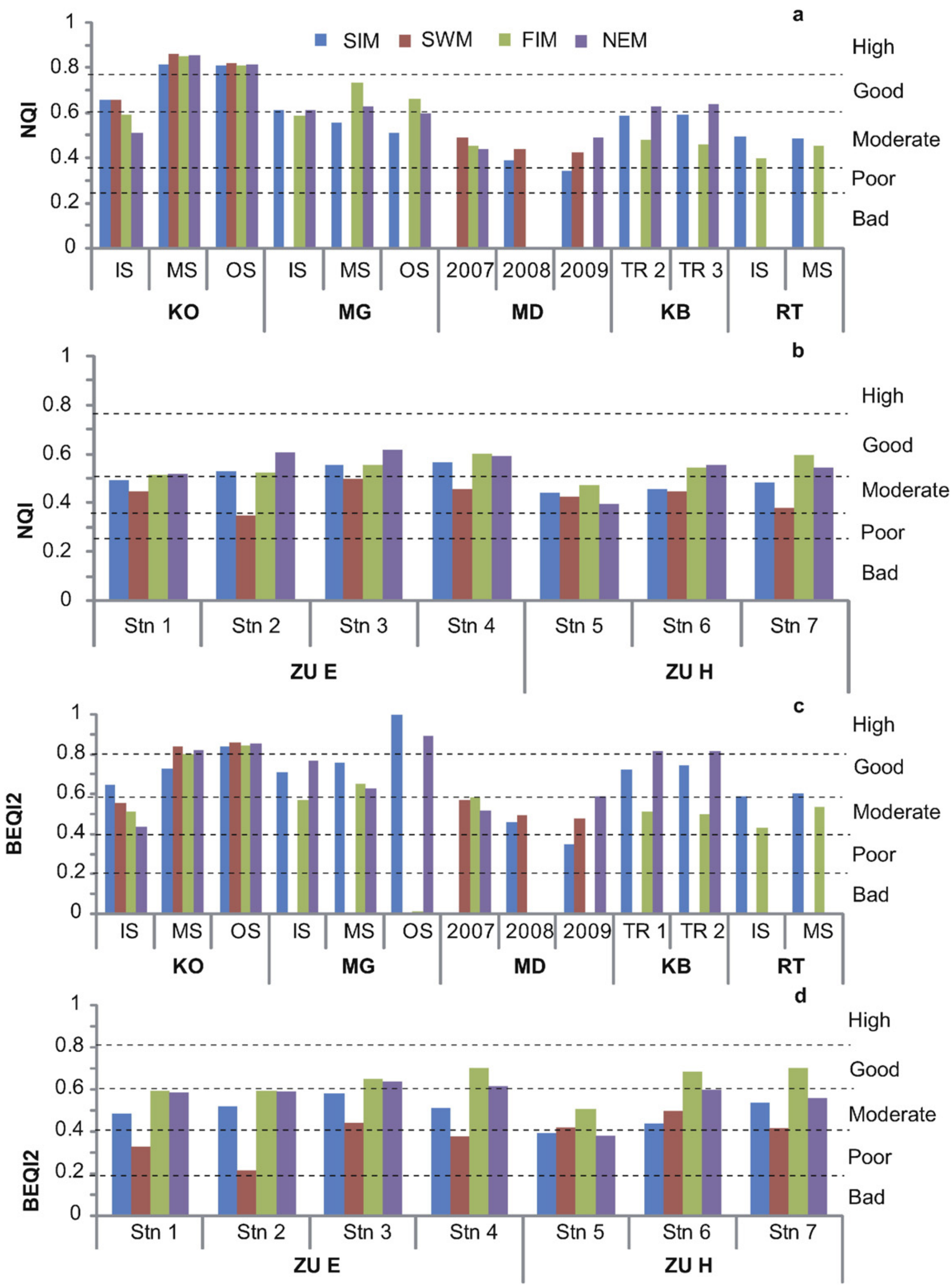

Fig. 7. Seasonal variation in NQI (a–b) and BEQI2 (c–d) at Kochi (KO), Mangalore (MG), Mandovi (MD), Kalbadevi (KB) and Ratnagiri (RT), Zuari estuary (ZU E) and harbour (ZU H). (NEM Northeast Monsoon; SIM Spring Intermonsoon; SWM Southwest Monsoon; FIM Fall Intermonsoon; IS inner shelf; MS mid shelf; OS outer shelf; TR Transect).

not very effective in detecting the ecological status in the region as the $\Delta+$ and $\Lambda+$ were within the 95% confidence funnel. According to Warwick and Clarke (1993), the univariate indices such as diversity, evenness and other related indices, reduce a great amount of information into a single summary index and cannot discriminate between naturally and anthropogenically affected communities.

The PA ratio was >1 in most locations, including the undisturbed locations of KB, GoM and PB. While BOPA was less severe since it classified KB, GoM and PB in a good ecological conditions, whilst harbours were in 'poor' to 'bad'. However, BOPA failed in assessing the naturally enriched regions of RT and MU. Further, this index classified the harbours in 'poor' to 'bad' ecological state when other indices classified them in a 'moderate' status. One of the strengths of the PA and BOPA is that the need for the taxonomic expert is reduced and, hence, saves time. However, it is widely criticised because the sensitivity to pollution for the same taxonomic group differs from species to species (Afli et al., 2008; Andrade and Renaud, 2011). Further, the amphipods had naturally low abundance or were absent in many locations, indicating a disturbed

Table 3
Inter-item correlation matrix for the selected biotic indices.

	BOPA	AMBI	MAMBI	IQI	DKI	NQI	BEQI2
BOPA	1						
AMBI	0.42	1.00					
MAMBI	0.28	0.14	1.00				
IQI	0.43	0.27	0.35	1.00			
DKI	0.51	0.40	0.47	0.62	1.00		
NQI	0.59	0.43	0.39	0.67	0.72	1.00	
BEQI2	0.47	0.32	0.48	0.63	0.76	0.71	1.00

condition, which was not the case (KB, GoM and PB). Therefore, the PA and BOPA is not a good indicator for detecting the ecological status of the Indian coast. This conclusion is in agreement with the use of BOPA within the Canary Islands and along the Tunisian Coast (Afli et al., 2008; Riera and de-la-Ossa-Carretero, 2014). According to Riera and de-la-Ossa-Carretero (2014), BOPA was reliable only in strongly impacted areas such as harbours and did not show a clear response to other anthropogenic disturbance. The ITI values in most of the study area were >60 and, therefore, our study confirms that the index is inefficient in detecting disturbance (Salas et al., 2006; Afli et al., 2008).

The ABC curve and W-Statistic suggest that *r*-selected species dominate the macrofaunal community along the coastal water of India. Similarly, the geometric abundance curve gave a rather confusing result as the curve in all the study area was irregular and extended from class 7 to 15. In polluted conditions, the community is usually dominated by a few opportunistic species and, hence, represented by an irregular curve with higher geometric class. Further, all the three indices showed a temporal variation.

The AMBI shows an increase from the southeast towards the northwest coast, indicating that the coastal waters have a 'good' to 'moderate' condition from south to north. However, the interpretation of the results must take into account that the Arabian Sea (west coast of India) is one of the five largest upwelling regions of the world (Naqvi et al., 2009). The seasonal upwelling during the monsoon period results in phytoplankton bloom followed by its collapse and enrichment of benthic system during FIM (Ingole et al., 2014). The dominance of EG IV (>50%) in the northwestern stations (RT and MU) reflected such an enrichment in the benthic indices with a higher value of AMBI. The organic enrichment from the seasonal upwelling favours smaller, faster-growing species and, as such, these conditions become unfavourable for more complex species to develop along the west coast of India. Such dominance of *r*-selected species may also occur under an undisturbed condition that may give an incorrect ecological status (Salas et al., 2006).

However, the reason for the low quality in the northwest coast of India could also be due to the muddy sediment in this region compared to the sandy sediment in the southwestern coast. Muddy sediment is known to be rich in organic matter. According to Muniz et al. (2005), muddy sediment along with organic matter can increase the AMBI index value. The effect of sediment texture was also seen in KB and KO (mid shelf), which are dominated by sand. Although the area becomes enriched with organic flux, the organic matter was low due to the low retention capacity of sand (Sivadas et al., 2013). Therefore, although the west coast of India is influenced by the upwelling, the organic matter retention was higher in the northwest due to the muddy substratum. Therefore, this natural pattern should be considered while evaluating the impact of anthropogenic disturbance along the Indian coast.

The AMBI index and all the multimetric indices worked satisfactorily in the harbours, revealing a 'moderate' quality condition. Moreover, the higher values were observed within the harbours with higher activity (ZU > KR > RT). The Zuari harbour is one of the major ports along the west coast of India and, hence, the activities are more compared to the harbours of Karwar and Ratnagiri. Station 5 in the ZU harbour was the most disturbed, as dredging is carried out every year immediately after the monsoon in order to deepen the channel from 8 to 13 m. This station had the lowest macrofaunal abundance and diversity throughout the study period, except during FIM due to recruitment, and the indices reflected this condition.

In Zuari, in addition to the harbour activities, fishing jetties, wharf, municipal waste and small ship repairing workshops are all located along the river. Despite this, most indices indicate a 'moderate' condition. One reason is that the Zuari estuary has a very good physical "self-cleaning" capacity. As a result, the disturbance of the environment was seen only around the harbour (Sivadas et al., 2011). Similarly, although there are several activities (tourism, sewage disposal and fishing jetties) along the Mandovi estuary, the overall water quality remains considerably good (VishnuRadhan et al., 2015). Further, the high freshwater runoff in the Mandovi estuary during the monsoon season dilutes and flushes the contaminants out of the estuary (VishnuRadhan et al., 2015). The impact of dilution of contaminants due to water circulation and its subsequent influence on the benthic community and, in turn, on the benthic indices have been reported from other areas (Borja et al., 2003; Muniz et al., 2005; Muxika et al., 2005).

The GoM and PB were classified as having 'good' ecological status by all the indices, except for the PA ratio. The Kalbadevi Bay is another relatively undisturbed region also confirmed by AMBI and all multimetric indices. The area is a potential placer mineral (ilmenite) mining site along the west coast of India. Since mining in the region has not yet commenced, the values can be considered baseline for the region.

In general, after the ICC analysis, the response of AMBI and multimetric indices is consistent. In the present study, sampling method, year, season and habitat differed. Despite the differences, high agreement between different indices was seen in assessing the ecological status.

The Pearson correlations between overall data of benthic indices and environmental parameters were not significant. When the correlation was carried out for each location, a significantly positive correlation between organic matter and species number, abundance, ES (50) and multimetric indices was observed at KB. According to Dauvin et al. (2007), it is the type of organic matter along with the sediment texture that will influence the benthic community. In this study, AMBI showed a positive correlation with carbohydrate.

4.2. Effect of seasonality on the benthic indices

In tropical regions, the annual rainfall brings about drastic changes in the environmental condition which, in turn, affects the benthic composition. The changes occur due to (1) increased organic matter flux stimulated by the increased river runoff (estuaries) and upwelling in the coastal zone, (2) colonisation of species from the adjacent regions, (3) initial decrease in abundance and diversity followed by sudden increase due to recruitment and, (4) initial colonisation by opportunistic species.

Most biological indices respond to any disturbance, whether caused by natural processes or by anthropogenic ones (Wilson and Jeffrey, 1994). The present study indicated that the effect of seasonality was higher in the estuaries and some coastal stations (inner shelf). In the ZU (estuary and harbour), AMBI shows 'bad' to 'poor' condition during the SWM while 'good' ecological status was observed during FIM-NEM. The change in ecological status was because of the natural defaunation occurred during the SWM followed by recruitment and migration during FIM-NEM (Alongi, 1990; Sivadas et al., 2011; Gaonkar et al., 2013). As a result, the condition in the area shifted from 'bad/poor' to 'good'. The 'good' condition observed in the ZU harbour (FIM-NEM) was because of the larval recruitment from the adjacent area. An example is the recruitment of the bivalve, *Timoclea scabra* (EG II) located at the estuarine mouth into the harbour region. The juveniles dominated the *T. scabra* population, and the highest value was recorded during the FIM-NEM (Sivadas et al., 2011). The values dropped to 'moderate' or 'poor' status during the SIM when the community becomes dominated by EG IV (*Paraprionospio* sp.). In general, the EG I and II group dominated the harbour community during FIM-NEM, while EG III and IV (*Mediomastus* sp. and *Paraprionospio* sp.) dominated during SIM and SWM. Moreover, reduction in harbour activities during the monsoon due to the rough weather conditions, allowed for the survival of sensitive species (EG I and II). With the end of monsoon, the activities are resumed with dredging of the channel to increase the depth, which increases the turbidity in the region. The result is the sudden decline of *T. scabra* and other species (Sivadas et al., 2011). On the other hand, the area outside the harbour (ZU estuary) showed a disturbed condition only during the SWM, which was due to the monsoonal defaunation and initial colonisation by EG III and IV. This indicates that the estuarine communities are structured by natural factors while anthropogenic activities would be more important in determining the benthic communities in the harbour.

Similarly, AMBI (RT coast) showed seasonality with low values during FIM in the inner shelf region. In shallow coastal areas (inner shelf), disturbance and resuspension of sediment is higher and it is a natural disturbance to benthic fauna. In fact, the changes during upwelling, i.e., high primary production due to nutrient enrichment and, hence, increased organic matter flux to the benthic system was also higher in the inner shelf compared to mid and outer shelf region. This was reflected in the indices as the community in the inner shelf region was dominated by EG III-IV (SWM-FIM). The community during this period was dominated by *Magelona* sp. (EG III) along the southwest coast (KO and MG) and *Paraprionopsio* sp. (EG IV) in the northwest coast (MU, RT and GO). On the other hand, multimetric indices, except for M-AMBI were less influenced by the seasonal variation in the macrobenthic communities. The effect of seasonality on those indices was observed only in estuaries. Since multimetric indices incorporate diversity, abundance and ratio of opportunistic/sensitive species measures, they are better in detecting the natural changes in the benthic communities (Kröncke and Reiss, 2010; Borja et al., 2011; van Loon et al., 2015). Overall, the present study confirms the usefulness of benthic indices developed in temperate waters, for assessing the ecological status of tropical waters (Muniz et al., 2005; Bigot et al., 2008).

An important step in the effective use of the biotic indices for management is to determine the sampling period, as the strength of various disturbances changes with a season (Karakassis et al., 2013). In the tropical region, the SWM period is associated with drastic changes in the physicochemical parameters that, in turn, affect the macrobenthic community structure (Gaonkar et al., 2013). Also, the monsoonal disturbance is always followed by the recolonization by opportunistic species (EG IV) such as *Paraprionospio* sp. and *Minuspio cirrifera* (Sivadas et al., 2011, 2013; Gaonkar et al., 2013). Further, the abundance of crustaceans is also low during this period. We suggest that for efficient use of benthic biotic indices in the tropical region, the sample should be collected during the dry period (January–May). Moreover, during the SWM, the weather conditions are bad along the Indian coast which affects the collection of samples. Since this period is also the primary recruitment of the benthic community, species richness and abundance show great fluctuation that will be reflected in the indices. In India, for the comprehensive environmental impact assessment of coastal waters, three season sampling is mandatory in a year (http://www.moef.nic.in). Therefore, identifying a suitable period for the effective assessment of the ecological status of an ecosystem will result in reducing the sampling efforts and hence reduce the cost of EIA studies.

4.3. Identifying the reference condition and species ecological status

Borja et al. (2012) suggested that there is a need to set a correct reference condition for all the indices. However, defining a reference condition is a difficult task as each area has its own environmental condition. Although, KB, GoM and PB were relatively undisturbed, the sediment in the region was dominated by sand while in rest of the area it was dominated by silty-clay. The Indian Coast has heterogeneous sediment and with the lack of historical data setting the reference condition was difficult. An example is that of ZU estuary, where the harbour was characterised by mud, while silty-sand was dominant outside the harbour. Borja et al. (2012) suggested the use of 15% increase in structural parameters as a 'precautionary' approach for the reliable use of the M-AMBI index. The result of the two methods, as mentioned in the methods section, showed no significant variation. For calculating IQI, DKI, NQI and BEQI 2 values, the highest values were used as a reference. The ICC analysis gave an excellent agreement. As mentioned by Borja et al. (2007) the high level of agreement between the indices in the classifications of the ecological status suggests that use of highest values as a reference can give reliable results.

Teixeira et al. (2010) recommended some modification for efficient use of AMBI index — (1) Use of expertise in assigning an ecological classification. In the present study, changes were made to the EG group based on expert advice and from our own experience. *Magelona* sp. was considered as EG III as the species showed a sudden increase after the upwelling season. The abundance of *Magelona* sp. increased to >10,000 ind m^{-2} in some stations after the upwelling and declined thereafter. Similarly, *Protodorvillea* sp. and *Staurocephalus* sp. was considered as EG III based on our study; (2) calibrate the categorization scaling using expert judgement — the west coast is characterised by high organic matter, and the muddy substratum are naturally dominated by opportunistic species (EG III and IV). Therefore, based on more detailed sampling and expert advice the scaling for AMBI index may be modified for the region; (3) use of different benthic indices; and (4) transformation of abundance data.

5. Conclusion

Deciding the set of ecological indices is a critical step in effectively assessing the health of the ecosystem. In the absence of a method to evaluate the ecological status of India, indices developed in the temperate waters were used. The indices were tested in different habitats and over temporal scales. The AMBI and multimetric indices performed satisfactorily to assess the ecological status of the coastal waters of India. Based on the results, we suggest that the non-monsoon period (January–May) as a suitable time of the year to use the indices for effective assessment of tropical coastal waters. A major problem was related to identifying a reference area because India has a vast coastline with complex oceanographic features and heterogeneous substratum. The temperate benthic indices were efficient in assessing the ecological status of Indian coastal waters, however, complementary use of different indices is recommended in order to accurately determine the environmental quality.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.marpolbul.2016.03.026.

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