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Original Article

Distribution of intertidal flat macrobenthos in Buntal Bay, Sarawak, Borneo

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Abstract

The distribution of macrobenthos in the intertidal area of Buntal Bay, Sarawak was studied based on systematic sampling conducted in 2014. This study aimed to determine the intertidal macrobenthic horizontal distribution and their relationship with environmental parameters. An analysis of the intertidal flat marobenthos community suggested that polychaetes dominated the community in terms of the number of individuals and species followed by crustaceans and molluscs. Polychaetes of families Nephtyidae, Spionidae, Capitellidae, and Magelonidae contributed to the high densities of macrobenthos. Multivariate analysis performed by the Biotic and Environmental linking analysis indicated that communities in Transect 1 and Transect 2 were best correlated with food availability (sediment chlorophyll *a*), and heterogeneity of sediment type (percentage of fine sand and very fine sand). Heterogeneity of sediment characteristic and food availability were identified as potentially playing a key role in the shaping of the intertidal macrobenthic distribution in Buntal Bay.

Keywords: macrobenthos, intertidal flat, Buntal Bay, horizontal distribution, Sarawak, Borneo

1. Introduction

Intertidal macrobenthos consists of a highly diverse group that is comprised mainly of polychaetes, crustaceans, and molluscs (Lastra *et al.*, 2006; Morais, Comargo, & Lana, 2016; Nakao, Nomura, & Satar, 1989; Netto & Lana, 1997; Peterson & Peterson, 1979; Whitlatch, 1982), as well as three lesser groups, namely echinoderms, nemerteans, and sipunculids (Morais *et al.*, 2016; Whitlatch, 1982). Early studies of intertidal macrobenthos were concerned mainly with macrobenthic zonation, classifying low, mid, and high intertidal zone on the basis of dominance species (Blanchet *et al.*, 2014; Rodil, Lastra, & Sánchez-Mata, 2006). Recently, enormous progress has been made towards comprehension of macrobenthic communities and ecosystem functioning in many parts of the world (Gerwhoing, Drolet, Hamilton, & Barbeau, 2016; Magni, Como, Montani, & Tsutsumi, 2006; Shin, Lam, Wu, Qian, & Cheung, 2008).

Previous studies reported on the environmental factors that influenced macrobenthic communities coupled with variations in tolerance of the macrobenthic organisms (Lu, 2005; Magni *et al.*, 2006; Peterson & Peterson, 1979). Community structure embodies all of the various ways that individual members of communities relate and interact with one another, i.e. spatial and temporal abundance of macrobenthos, and how the community level properties arising from these relations with environment and biological factors (Giller, 1984; Tokeshi, 1993). Alterations in the environmental characteristic of the habitat can strongly affect the composition and abundance of species among sites which influences species diversity (Faraz *et al.*, 2016; Seiderer & Newell, 1999). However, the macrobenthos that inhabit the tropical regions, particular in Sarawak, has been poorly studied. Buntal Bay is one of the few intertidal flats in Sarawak that serves as an important fishery area for the economic species of razor clam *Solen* spp. (Rahim, 2011). To date, macrobenthic communities in this intertidal flat of

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Buntal Bay have never been described. Therefore, the present study aimed to determine the intertidal flat macrobenthic abundance in Buntal Bay with particular emphasis on the context of community structure and the relationship with environmental factors. The findings of this study provide useful baseline data for future ecological and systematic studies of macrobenthos in this area.

2. Materials and Methods

2.1 Study site

Bako-Buntal Bay (N 1°41'52.03'', E 110°22'28. 10'') is a semi-circular bay bordered by Gunung Santubong to the west and Bako National Park to the east (Figure 1A). A mangrove forest stretches between the two promontories. During neap tides, almost a third of the Bay is exposed sand-mud flats. The Bay is globally important as a migration site for waterbirds (Howes, 1986; Mizutani *et al.*, 2006).

2.2 Field sampling

2.2.1 Macrobenthic sampling

Macrobenthic sampling was conducted during low tide in May 2014. The approach taken was by performing the line transects method. Two transects were performed perpendicular to the shoreline starting from the low water mark to the high water mark (Figure 1B). The distance between the transects was 1.5 km. A total of 21 sampling stations were established on these two transects. The distance between each station was 150 m. At each station three quadrates 0.25 m^2 (= three replicates) was placed at 5 meter intervals on the right and left hand side of the transect. Sediment in the quadrates was scooped with a spade approximately 15 cm deep based on preliminary sampling conducted on vertical distribution. In the field, all sediment samples were sieved through a 500 µm mesh sieve and fixed in 5% buffered formalin before further analysis in the laboratory.

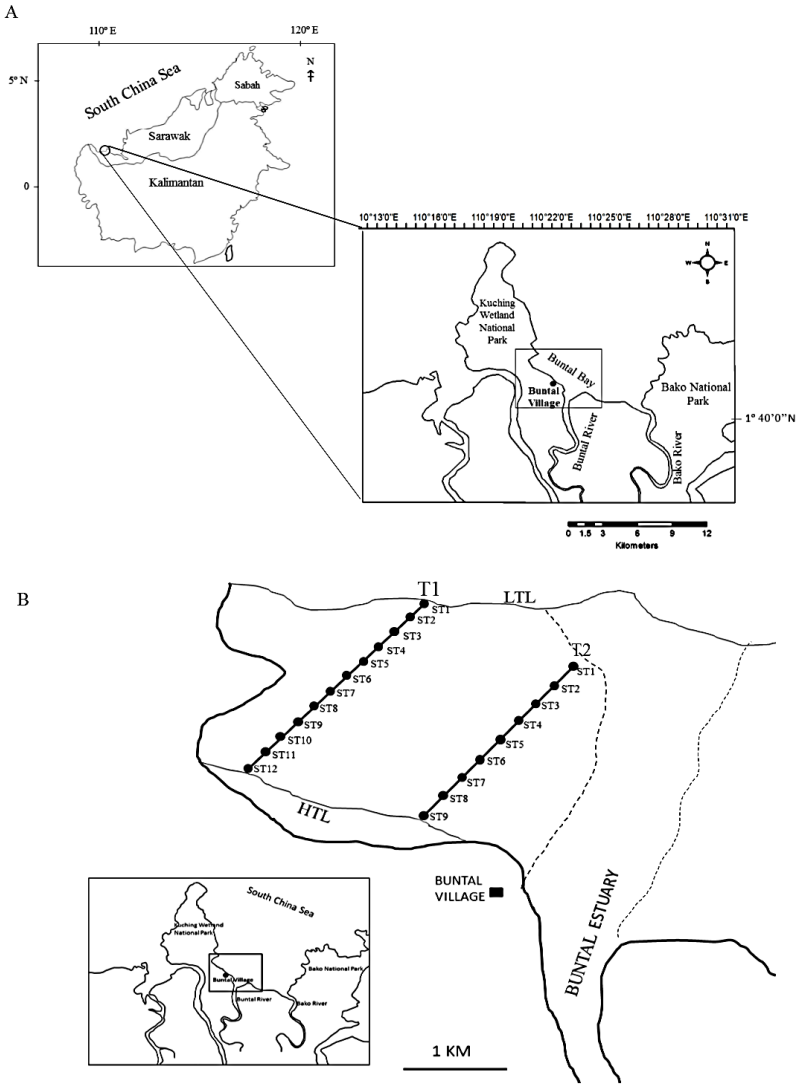

Figure 1. A) Map of Sarawak showing the location of Buntal Bay. B) Illustrations of line transects performed for horizontal distribution study in the intertidal flat of Buntal Beach. T1=Transect 1, T2=Transect 2, ST=Station, HTL=High tide level, LTL=Low tide level.

2.2.2 Environmental parameters

Water parameters for the interstitial water, i.e. salinity, temperature, dissolved oxygen, and pH, were measured *in situ*. Interstitial water was obtained using a modified device that followed Giere, Eleftheriou, and Munson (1988). Three replicates of sediment samples (15 cm) were collected using a perspex corer at each station within the macrobenthic sampling quadrates for the determination of grain size distribution and total organic matter (TOM). Two replicates of 1 cm of surface sediment was also taken within the quadrates using a perspex corer and placed in a plastic bag for determination of chlorophyll *a* (Chl *a*).

2.3 Laboratory analysis

2.3.1 Macrobenthic study

The first step of macrobenthic extraction was carried out after formalin was removed and the macrobenthos were transferred to 70% ethanol before the sorting process. Fine sorting was carried out in order to separate organisms belonging to different high taxa under the stereomicroscope. For a detailed taxonomic identification of macrobenthic specimens, the use of a compound microscope was needed. Species were identified to the lowest practical taxon by referring to the identification keys such as Day (1967) for Polychaeta, Brinkhurst (1982) for Oligochaeta, Valentich-Scott (2003) for Mollusca, Gibson and Knight-Jones (1994) for Nemertinea, Cornelius, Manuel, and Ryland (1994) for Cnidaria, Abele, and Kim (1986) for Decapoda, Barnard and Karaman (1991) for Amphipoda.

2.3.2 Sediment analysis

The method used to determine the grain size was based on the standard method by Bale and Kenny (2005). The sediment grain size analysis was determined using the dry and wet sieving technique in order to determine the fraction mixture of gravel, sand, silt, and clay. A simple estimate of the organic contents can be derived from the mass of loss of ignition. This method involved drying the samples at low temperature (40 ºC) for 24 h, then combusting the organic content at high temperature (450 ºC) for 4 h (Greiser & Faubel, 1988). The loss of weight indicated the amount of TOM in the samples.

The amount of Chl *a* in the sediments was determined using the method by Lorenzen (1967). The method started by grinding the sediment inside a mortar with 90% acetone. An aliquot of 10 mL was then transferred into a centrifuge tube and left overnight before centrifugation for 30 min at 4000 rpm. The supernatants were then transferred into a cuvette and measured in a spectrophotometer (HACH, DR2800) before and after acidification. One drop of 0.2 M hydrochloric acid was added to 1.5 mL of extract volume and absorbance was measured at 665 nm. A turbidity blank was measured at 750 nm.

2.4 Data analysis

Determination of quantitative macrobenthic composition and density was based on the number of macrobenthic individuals per 0.25 m². One-way analysis of variance (ANOVA) was used to test the difference between the environmental variables between the stations. The statistical significance of differences among sites was assessed using analysis of similarities (ANOSIM) and a non-metric method based on randomization of rank-similarities among all samples and multiple pair-wise comparisons (Clarke, 1993). A significance level of $P<0.05$ was used in all tests. The number of species in each sample was used as a direct measure of the species richness index. The Shannon-Wiener diversity index is widely used as an absolute measure of diversity. Species equitability was determined by Pielou's evenness index. A cluster analysis was carried out to delineate the macrobenthic communities of the sampling stations into different groups using a Bray-Curtis similarity measure based on the presence/absence transformed data and group-average linkage. The relevance of the station groups obtained was evaluated by the similarity profile routine (SIMPROF) tests (Clarke & Gorley, 2006). Differences in the composition of the macrobenthic assemblages among stations were verified through non-metric multidimensional scaling (NMDS). Subsequently, the contribution of species in each group similarity was assessed using the SIMPER (similarity percentages) procedure (Clarke & Gorley, 2006). Macrobenthic assemblages were characterized using univariate and multivariate measures using PRIMER v6 for determination of community structure (Clarke & Gorley, 2006). Biotic and Environmental linking (BIO-ENV) and Spearman's rank coefficient analysis were performed to test which environmental variables were correlated with the macrobenthic community.

3. Results

3.1 Environmental parameters

ANOVA analysis showed that the physico-chemical parameters of the water in both transects were significantly different ($P=0.0001$). Generally, the water temperatures in Transect 1 (T1) and Transect 2 (T2) ranged from 33.30 ºC to 36.10 ºC and 29.03 ºC to 33.57 ºC, respectively. Salinity tended to be much higher in T1 than in T2 (30.3 to 33.3 psu vs. 20.33 to 24.73 psu). The dissolved oxygen concentration in T1 ranged from 0.14 to 2.76 mg/L and between 0.17 to 0.47 mg/L recorded in T2. The recorded pH values in T1 ranged from 7.47 to 7.93 and in T2 from 6.6 to 7.73.

A summary of sediment characteristics is presented in Table 1. The sediment grain size in both transects consisted of medium sand and moderately sorted sands. ANOVA analysis showed that the TOM was significantly different among the stations for both T1 ($P=0.0001$) and T2 ($P=0.0001$). Total Chl *a* concentrations ranged from 2.98 to 53.49 mg/m^3 in T1 and 1.73 to 83.05 mg/m^3 in T2 (Table 1). ANOVA analysis showed that the Chl *a* concentrations were significantly different among the stations in T1 ($P<0.05$) and T2 ($P<0.05$).

3.2 Species composition and density

A total of 97 macrobenthic species were identified in the intertidal zone of Buntal Bay which were composed mainly of Polychaeta, Crustacea, Mollusca, and Nemertinea. The majority of macrobenthic species was polychaete worms

Table 1. Summary of sediment granulometry and sediment biological parameters at Transect 1 and Transect 2.

Station		Cs	Ms	Fs	Vfs	SC	Mean	Sorting	Skewness	TOM	Chl *a*
Transect 1	ST1	28.5	43.6	21.7	5.9	0.4	1.7	0.8	0.2	0.4	3.0
	ST2	23.0	44.0	30.3	1.6	0.1	1.8	0.8	-0.2	1.6	29.6
	ST3	25.0	51.3	18.3	4.6	0.7	1.7	0.7	0.2	2.0	25.6
	ST4	32.0	48.0	16.2	2.7	0.3	1.6	0.7	0.2	2.0	21.5
	ST5	25.8	47.0	21.9	4.7	0.5	1.7	0.8	0.1	1.5	3.6
	ST6	29.6	53.6	14.7	2.0	0.1	1.6	0.7	0.2	0.2	19.0
	ST7	21.9	56.3	18.3	3.1	0.2	1.6	0.7	0.5	3.1	5.0
	ST8	28.4	42.9	24.2	2.4	0.3	1.7	0.8	-0.1	2.8	15.0
	ST9	30.4	43.4	22.8	1.9	0.3	1.6	0.8	0.0	1.6	10.2
	ST10	29.3	42.8	23.7	2.3	0.1	1.7	0.8	0.0	2.7	5.1
	ST11	29.6	43.4	22.0	1.8	0.1	1.6	0.8	-0.1	1.1	53.5
	ST12	28.7	44.3	22.5	2.8	0.2	1.7	0.8	0.0	1.5	19.0
Transect 2	ST1	29.2	45.2	19.4	4.2	1.0	1.7	0.8	0.2	2.6	45.5
	ST2	32.7	52.1	12.5	0.8	0.1	1.5	0.7	0.1	2.6	2.1
	ST3	21.4	40.0	31.3	6.3	0.8	1.8	0.8	0.2	0.5	8.8
	ST4	15.4	35.8	39.7	7.9	0.9	2.1	0.8	-0.4	0.8	18.1
	ST5	23.1	32.2	28.1	14.3	2.2	1.9	0.9	-0.1	1.7	52.1
	ST6	19.3	19.4	28.1	27.4	2.9	2.1	1.0	-0.5	0.5	83.1
	ST7	32.7	36.3	20.2	7.7	1.0	1.7	0.9	0.2	2.7	10.0
	ST8	27.5	40.5	23.0	6.7	2.0	1.7	0.8	0.2	1.7	1.7
	ST9	23.3	31.1	30.6	13.0	1.9	1.9	0.9	-0.1	3.2	2.1

Notes: Cs, Coarse sand (%) = 1 mm; Ms, Medium sand (%) = 250 µm; Fs, Fine sand (%) =150 µm; Vfs, Very fine sand = 63 µm; SC, Silt and clay (%) = <63 µm; TOM, Total organic matter (g/g sed); Chl *a*, Chlorophyll *a* (mg/m^3).

(Annelida). Other groups with fewer numbers of species were Oligochaeta and Echinodermata.

In total, the mean density of macrobenthos collected in both transects was 3461.67 ind.m^{-2}. The mean density of all macrobenthos varied from 63.33 to 397.67 ind.m^{-2} in T1 and 26.67 to 450.67 ind.m^{-2} in T2. In T1, the highest density was recorded at ST8 (397.67 ind.m^{-2}) followed by ST6 (296.67 ind.m^{-2}). The lowest density was recorded at ST11 (63.33 ind.m^{-2}). The high densities in ST8 and ST6 corresponded to the occurrence of the high densities of the polychaetes *Prionospio* sp. 1 and *Nephtys sphaerocirrata*. Four species of polychaetes recorded a greater density value, namely *Nephtys sphaerocirrata* (217.3 ind.m^{-2}), *Prionospio* sp. 1 (186.7 ind.m^{-2}), *Nephtys* sp. 1 (109.3 ind.m^{-2}), and *Spiophanes* sp. 1 (80.0 ind.m^{-2}). However, two species of molluscs, *Umbonium elegans* (150.7 ind.m^{-2}) and *Tellina* sp. 1 (126.7 ind.m^{-2}), also contributed to the high density of macrobenthos.

In T2, the highest macrobenthic density was recorded at ST7 (450.67 ind.m-^{2}) followed by ST8 (302.67 ind.m^{-2}). The lowest density was recorded at ST4 (26.67 ind.m^{-2}). The highest density in ST7 was contributed by the high densities of *Magelona* sp. (61.33 ind.m^{-2}) and *Tellina* sp. 1 (66.67 ind.m^{-2}). Similar to T1, polychaete species were dominant in this transect followed by molluscs. Three species of polychaetes which had the highest density value were *Nephtys sphaerocirrata* (122.7 ind.m^{-2}), *Barantolla* sp. (161.33 ind.m^{-2}), and *Magelona* sp. 1 (146.7 ind.m^{-2}). The occurrence of *Tellina* sp.1 (137.33 ind.m^{-2}) also contributed to the high density of macrobenthos.

3.3 Species number, richness, diversity, and evenness

In general, the total number of species was found higher in T1 than T2 (Figure 2A and Figure 2B). With regard to the ecological indices between tide marks, number of species and species richness index showed similar patterns in T1 (Figure 2C) and T2 (Figure 2D). In both transects the number of species and species richness index was significantly different between the stations ($P<0.05$). In T1, the highest number of species and species richness index were recorded at ST8 (23.67±5.51 and 3.78±0.87, respectively) and the lowest number of species and species richness index were recorded at ST2 (4.33±2.3 and 0.74±0.45, respectively). In T2, the number of species and species richness index value was slightly lower compared to T1. The highest number of species and species richness value in ST7 were 20±6.56 and 3.10±0.86, respectively and the lowest values were in ST9 (3.67±1.15 and 0.51±0.03, respectively).

In both transects, the species diversity index and species evenness index fluctuated in both transects from the low tide level (LTL) to the high-tide level (HTL). In T1, the highest species diversity was observed at the mid-tide level (MTL) in ST8 (2.79±0.21) (Figure 2E and Figure 2F). High species diversity in these stations corresponded to a greater total number of species recorded. The lowest species diversity index value was recorded in ST2 (LTL) with a value of 1.21±0.51. For other stations, the species diversity index value ranged from 1.15 to 2.29. In T2, the highest species diversity was observed at ST7 (MTL: 2.58±0.34). The lowest species diversity index value was recorded in ST9 (LTL: 1.03±0.06).

The high evenness index (on a scale of 0-1) indicated that macrobenthic species were evenly distributed among the stations. The highest evenness index value in T1 was observed at ST1 (0.91±0.1) and the lowest values were at ST11 and ST12 (0.83±0.09 and 0.83±0.032, respectively) (Figure 3). Values for other stations ranged from 0.86 to 0.90. In T2, the highest value recorded was at ST4 (0.94±0.037) and lowest value was at ST1 (0.75±0.17). The evenness index values for the other stations ranged from 0.83 to 0.93 (Figure 2E and Figure 2F).

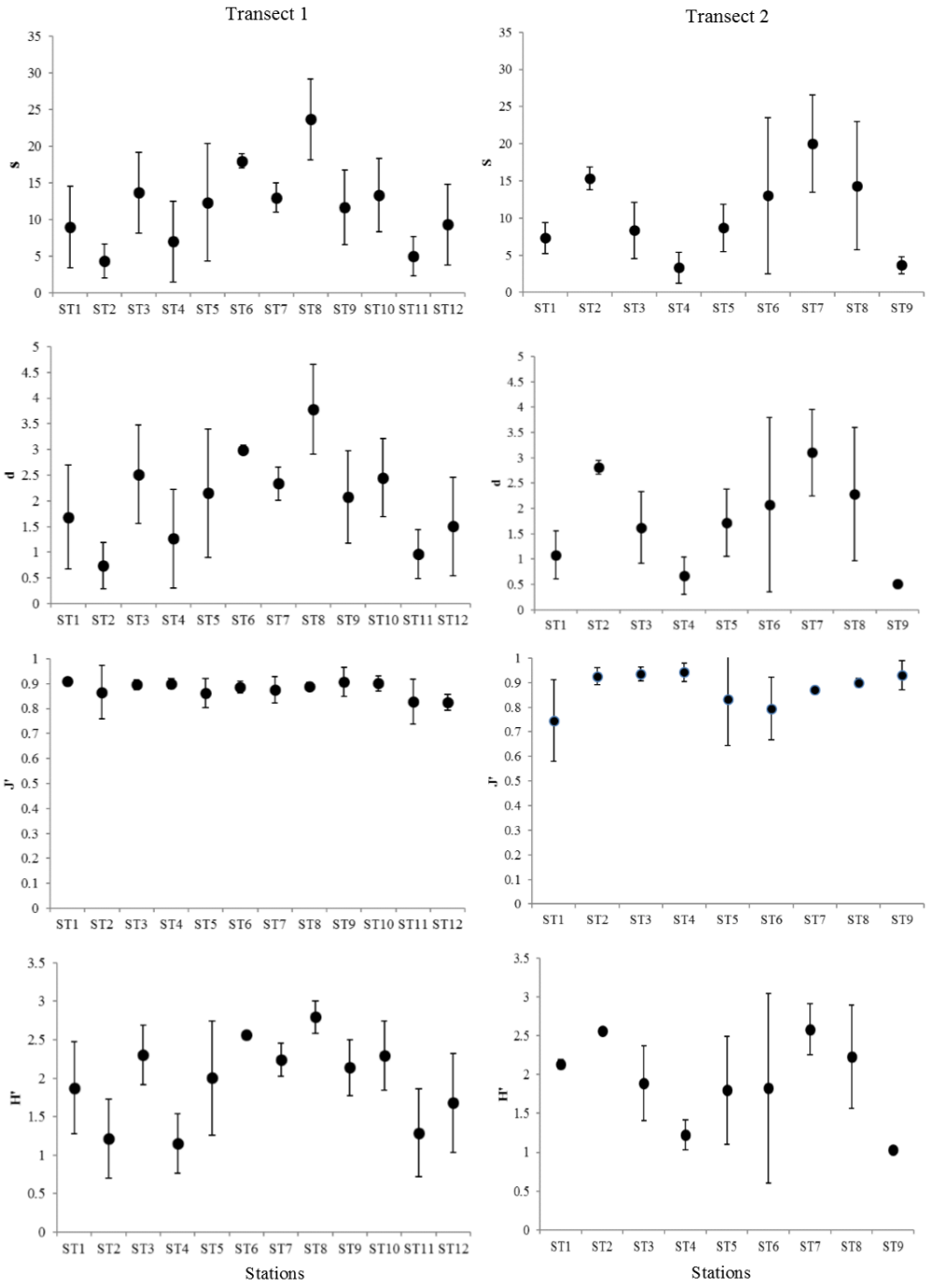

Figure 2. Means and SDs for univariate measures of the macrobenthic community indices: Number of species (S), Species richness index (d), Species evenness index (J') and Shannon's species diversity index (H') in Transect 1 and Transect 2 of Buntal Beach.

3.4 Community structure

Results of one-way ANOSIM test indicated significant distribution of the macrobenthic community between the stations in T1 ($r=0.558$, $P=0.1$). Cluster analysis and SIMPROF test revealed that the macrobenthic assemblages in T1 were significantly different between the stations ($P<0.005$) with Bray-Curtis similarities of 17% ($P=0.001$) and 33% ($P=0.02$). Based on 33% similarities, NMDS ordination (stress: 0.11) and cluster analysis suggested that T1 consisted of two groups (Figure 3). Group 1 was comprised of ST2 and ST11. Group 2 had the highest number of stations and species, that represented the MTL and LTL (excluding ST1), and it was relatively homogenous with respect to species composition. In T1, species homogeneity was the greatest in ST2 with average similarities of 68.28% followed by ST1

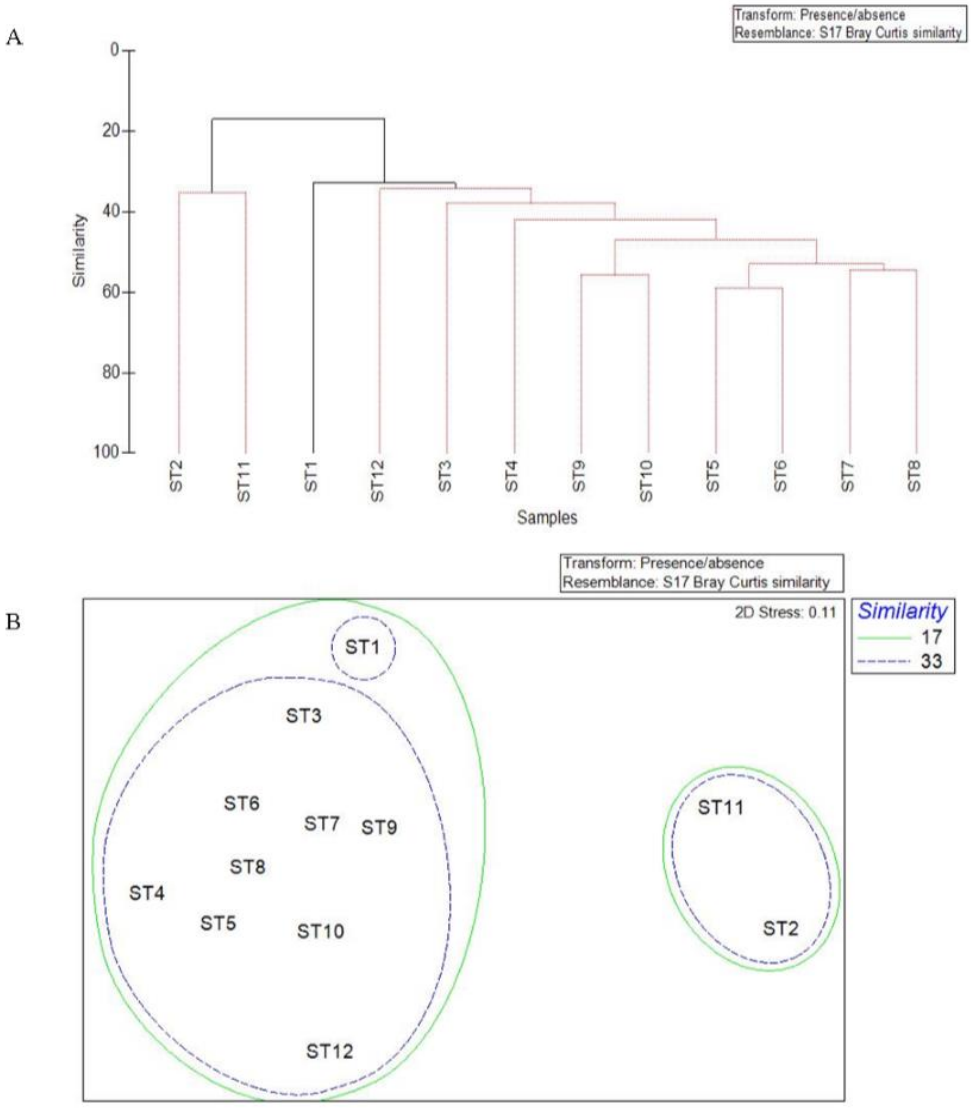

Figure 3. A) Clustering analysis of the macrobenthic community in Transect 1 based on Bray-Curtis similarity. The cluster grouping can be identified by dark black lines after performing the SIMPROF test. B) Macrobenthic assemblages based on cluster analysis and SIMPROF separated in the non-metric multidimensional scaling ordination plot.

(61.92%) and ST6 (53.27%), and the least was in ST5 (15.53%) (Table 2). According to the SIMPER analysis, this shift was mainly caused by a difference in densities of *Tellina* sp. 1, *Barantolla* sp., *Prionospio* sp. 1, and *Nephtys* sp. 1.

One-way ANOSIM test in T2 indicated that significant distributions of the macrobenthic community had occurred (r=0.859, P=0.1). Based on cluster analysis and SIMPROF, the macrobenthic distribution in T2 was significantly different between the stations (P<0.005) with Bray-Curtis similarities of 11% (P=0.001) and 24% (P=0.003). Based on 24% similarities, NMDS (stress: 0.05) and cluster analysis suggested that the macrobenthic communities in T2 were distinguished by three groups (Figure 4). The first group was separated from the others represented by the macrobenthic communities from ST1. The second group was made up of macrobenthic communities from ST4 and ST9. The third group consisted of a combination of ST2, ST3, ST5, ST6, ST7, and ST8 communities. According to the SIMPER analysis, species similarities were the highest in ST4 (79.48%), ST9 (74.57%), and ST1 (71.02%) and the least was in ST6 (34.42%). The differences in the densities of *Tellina* sp. 1, *Magelona* sp., *Glycera* sp. 1, and *Barantolla* sp. accounted for the distribution differences in this transect (Table 3).

3.5 Macrobenthos relationship with environmental parameters

To determine whether the environmental parameters might influence the variability in the macrobenthic community distributions in both transects, a multivariate analysis using the BEST BIO-ENV routine, and Spearman's rank correlations were carried out. The BIO-ENV analysis showed that the correlations between environmental variables and macrobenthic density were modest in T1 and T2 (Table 4).

Table 2. Species responsible for similarities of macrobenthos in each station at Transect 1 as indicated by the SIMPER procedure which is based on presence/absence data and percent contribution (percentage contribution of total similarity).

Species	Percent Contribution	Percent Cumulative
Group ST1 Average similarity: 61.92%		
Nephtys sp. 1	20.67	20.67
Prionospio sp. 1	20.14	40.8
Nephtys sphaerocirrata	17.51	58.31
Group ST2 Average similarity: 68.28%		
Barantolla sp.	34.2	34.2
Carinoma sp. 4	24.54	58.74
Group ST3 Average similarity: 30.76%		
Cumella sp.	30.12	30.12
Group ST4 Average similarity: 17.23%		
Nephtys sp. 1	12.21	12.21
Group ST5 Average similarity: 15.53%		
Nephtys sp. 1	20.85	20.85
Group ST6 Average similarity: 53.27%		
Prionospio sp. 1	19.07	19.07
Umbonium elegans	17.71	36.78
Group ST7 Average similarity: 41.66%		
Nephtys sphaerocirrata	30.13	30.13
Group ST8 Average similarity: 39.39%		
Prionospio sp. 1	32.4	32.4
Group ST9 Average similarity: 21.75%		
Tellina sp. 2	37.92	37.92
Group ST10 Average similarity: 26.25%		
*Tellin*a sp. 1	41.9	41.9
Group ST11 Average similarity: 45.40%		
Barantolla sp.	48.81	48.81
Group ST12 Average similarity: 49.03%		
Tellina sp. 1	61.42	61.42

Table 3. Species responsible for similarities of macrobenthos at each station in Transect 2 as indicated by the SIMPER procedure which is based on the presence/absence data and percent contribution (percentage contribution of total similarity).

Species	Percent Contribution	Percent Cumulative
Group ST1 Average similarity: 71.02%		
Nephtys sp.2	23.56	23.56
Glycera sp.3	17.63	41.19
Nephtys sp. 1	17.63	58.82
Glycera sp.1	9.99	68.81
Group ST2 Average similarity: 40.79%		
Prionospio sp. 1	21.15	21.15
Group ST3 Average similarity: 51.07%		
Ophiura sp.1	31.06	31.06
Precephalothrix sp.	24.56	55.62
Group ST4 Average similarity: 79.48%		
Barantolla sp.	86.1	86.1
Group ST5 Average similarity: 43.28%		
Magelona sp.	45.1	45.1
Group ST6 Average similarity: 34.42%		
Barantolla sp.	54.06	54.06
Group ST7 Average similarity: 56.69%		
Tellina sp. 1	23.98	23.98
Spiophanes sp.	14.87	38.85
Magelona sp.	11.51	50.36
Group ST8 Average similarity: 43.97%		
Magelona sp.	26.85	26.85
Group ST9 Average similarity: 74.57%		
Barantolla sp.	38.22	38.22
Notomastus lineatus	28	66.22

Table 4. Summary of BIO-ENV analysis based on the Bray-Curtis similarities with fourth root transformed data performed for macrobenthic density at Transect 1 (T1) and Transect 2 (T2).

Station	No. of variables	Factors	Correlation
T1	3	1, 3, 15	0.450
	3	1, 4, 15	0.450
	3	1, 6, 15	0.450
T2	2	2, 14	0.487
	2	6, 14	0.487
	2	14, 15	0.487
	4	2, 3, 6, 14	0.457

Notes: The environment factors associated to correlation selection. 1: Dissolved oxygen; 2: pH; 3: Salinity; 4: Temp; 6: Chl-*a*; 14: Fine sand; 15: Very Fine sand.

The results of BIO-ENV revealed that the microbenthic densities were all significant at T1 (rho=0.45, P=0.26) and T2 (rho=0.487, P=0.37). When all environmental parameters were included in the analysis, the macrobenthic communities in T1 correlated the best with dissolved oxygen, pH, salinity, sediment Chl *a*, and heterogeneity of sediment type (percentages of fine sand and very fine sand). To gain an insight on how the environmental parameters affect the structure of the intertidal macrobenthic communities, Spearman's rank correlation was performed (r) between community indices (species density, species diversity and species evenness) and the environmental parameters (Table 5). In T1, the strongest Spearman's rank correlation showed a statistically significant correlation ($P<0.05$) between species evenness and dissolved oxygen. Based on the BIO-ENV analysis, the macrobenthic communities in T2 were correlated the best with pH, Chl *a*, salinity, and heterogeneity of the sediment type (percentage of fine sand and very fine sand). In T2, the strongest Spearman's rank correlation revealed a statistically significant correlation ($P<0.05$) between species evenness and Chl *a* (Table 6). Species diversity showed a significant correlation between sediment type (fine sand) and salinity. Species density showed a strong correlation between salinity and temperature; however, no significant level was detected.

4. Discussion

In this study, 97 species of macrobenthos were collected. The macrobenthic species identified in Buntal Beach were mostly contributed by Polychaeta, Mollusca, Crustacea, and Nemertinea. Among the macrobenthos, the most taxonomically diverse group was the polychaete which accounted for 56% to 62% of total macrobenthos collected in TI and T2, respectively, which was previously reported as an important taxa inhabiting the intertidal habitat in terms of the

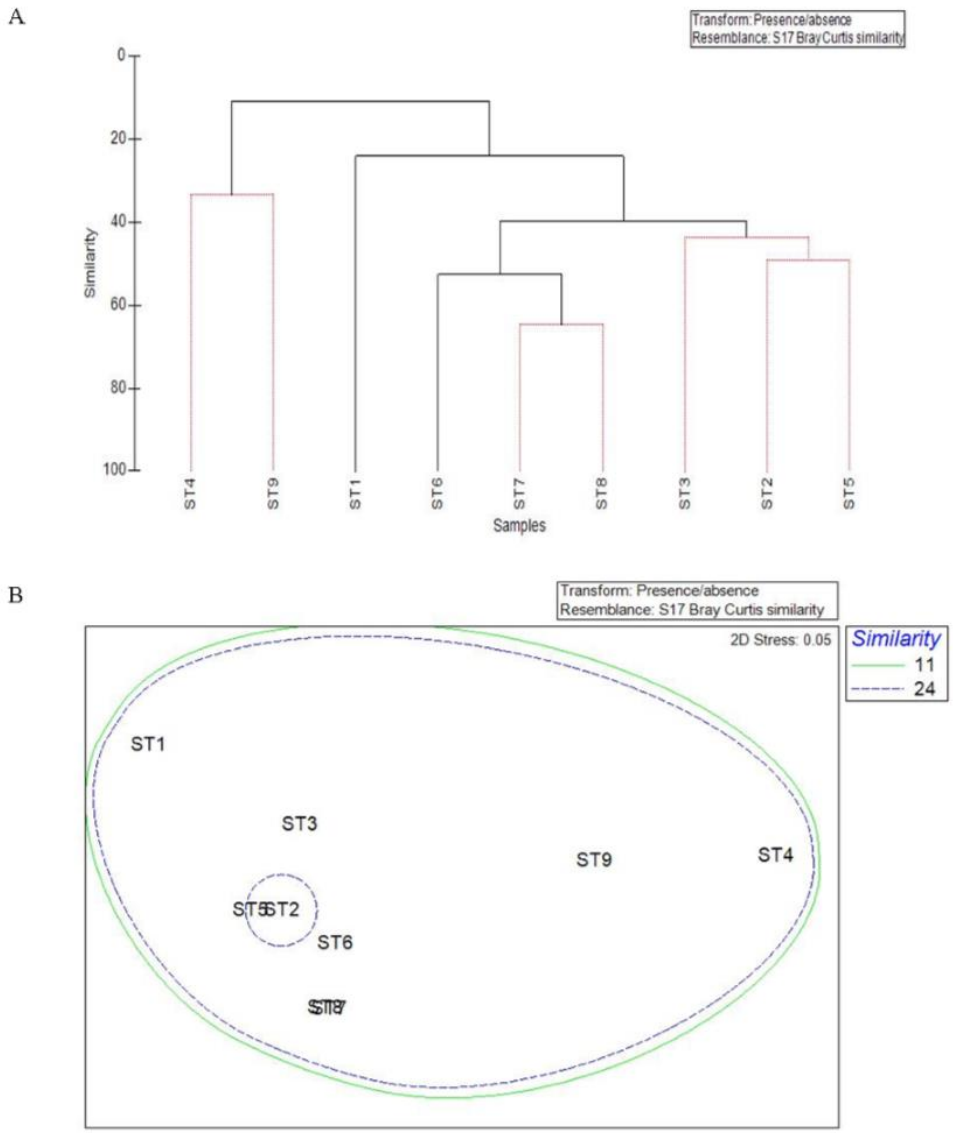

Figure 4. A) Clustering analysis of macrobenthic community in Transect 2 based on Bray-Curtis similarity. The cluster grouping can be identified by dark black lines after performing the SIMPROF test. B) Macrobenthic assemblages based on cluster analysis and SIMPROF separated in the non-metric multidimensional scaling ordination plot.

Table 5. Spearman's rank correlations (*r*) between species density, species diversity index (H') and species evenness index (J') with environment parameters at Transect 1.

		Species density	H'	J'
Dissolved oxygen	*r*	-0.235	0.186	**0.648**
	P-level	0.463	0.564	**0.023***
Salinity	*R*	0.380	0.162	-0.303
	P-level	0.223	0.615	0.339
Temperature	*R*	-0.160	-0.178	-0.046
	P-level	0.618	0.579	0.886
Chlorophyll *a*	*R*	-0.420	-0.329	-0.406
	P-level	0.175	0.297	0.191
Fine sand	*R*	0.196	0.140	0.161
	P-level	0.542	0.665	0.618

Note: (*) indicates a significant difference at P<0.05.

Table 6. Spearman's rank correlations (*r*) between species density, species diversity index (H') and species evenness index (J') with environment parameters at Transect 2.

		Species density	(H')	(J')
pH	*r*	0.261	-0.252	0.609
	p-level	0.498	0.513	0.082
Salinity	*r*	**0.571**	**0.804**	-0.281
	p-level	**0.109**	**0.009***	0.464
Temperature	*r*	**-0.527**	0.083	0.274
	p-level	**0.145**	0.832	0.475
Chlorophyll *a*	*r*	-0.035	-0.087	**-0.794**
	p-level	0.929	0.825	**0.011***
Fine sand	*r*	-0.381	**-0.848**	0.299
	p-level	0.311	**0.004***	0.434

Note: (*) indicates a significant difference at P<0.05.

number of species and species density (Ditmann, 2002; Lastra *et al.*, 2006; Morais *et al.*, 2016).

The present study showed that the high number of species collected in this area was greater than other regions with the same habitats such as in Guaratuba Bay, Brazil with 75 species (Morais *et al.*, 2016), Paranagua Bay, Brazil with 52 species (Netto & Lana, 1997), Gulf of Mexico with 21 species (Coblentz, Henkel, Sigel, & Taylor, 2015), North of Portugal with 22 species (Veiga, Rubal, Cacabelos, Maldonado, & Sousa-Pinto, 2014), North of Spain with 31 species (Lastra *et al.*, 2006), Wenzhou Bay, China with 38 species (Bao-Ming, Yi-Xin, & Hong-Yi, 2011), and Karnafuly, India with 33 species (Islam *et al.*, 2013). On a regional scale, macrobenthic community studies in the intertidal area of Buntal Bay have not been done in detail. The number of macrobenthic species collected in Buntal Bay were relatively higher compared to other studies of similar habitats such as in Teluk Aling, Pulau Pinang, Malaysia with 46 species (Ahmad, Fang, & Yahya, 2011); Kuala Selangor, Malaysia with 44 species (Nakao *et al.*, 1989), and Barangay Tagpangahoy, Philippines with 39 species (Medrano, 2015). The macrobenthic species obtained at the intertidal area of Buntal Bay resembled the communities in other intertidal habitats reported in Malaysia coastal waters (Broom, 1982; Nakao *et al.*, 1989; Ahmad *et al.*, 2011).

In this study, the macrobenthic community pattern fluctuated in both transects and did not followed or vary with tidal gradients. Occurrence of some dominant species was apparent at both transects. This can be explained by the high dominance of *Barantolla* sp., *Glycera* sp., *Nephtys sphaerocirrata*, and *Carinoma* sp. The dominance of one or a few taxa in intertidal habitats was observed in similar studies along with the variability of species composition (Coblentz *et al.*, 2015; Mclachlan & Jaramillo, 1995;). Giller (1984) suggested that communities can differ in species diversity due to the variability of available food resources, niche width of species composition, and lastly the degree of niche overlap.

The environmental data and macrobenthic data make it possible for us to test the general hypothesis that environmental factors influence the macrobenthic community structure in an intertidal area of Buntal Bay. Surprisingly, the dissolved oxygen in the interstitial water was positively correlated to species evenness in T1. The lowest values of species evenness and species density were recorded at ST11 and ST12 (T1) with low dissolved oxygen concentration (0.1 to 1.05 mg/L). Bottom sediments are the final sink for many anthropogenic contaminants and they can accumulate great amounts of organic matter that affect the oxygen content of the bottom water (Whitltach, 1982). Hypoxia/anoxia causes reduced macrofauna abundance and in turn reduces the amount of bioturbation activities (Rosenberg, Hellman, & Johansson, 1991; Gray, Wu, & Or, 2002). This study observed that interstitial water salinity showed a significant correlation with species diversity in T2. Lower salinity indicates the influence by freshwater input. Freshwater input not only lowers the salinity but also the nutrient enrichment which results in a macrobenthic community that is different from high salinity conditions (Nishijima *et al.*, 2013). Thus, freshwater input from an adjacent river or rainfall influenced the salinity and affected the macrobenthic communities in T2.

Sediment characteristics were among the major factors that best explained the pattern of macrobenthos in T2. Results from the correlation analyses in T2 showed that sediment properties (e.g., sediment mean grain size of coarse sand and fine sand) were significantly correlated with species diversity. The most frequently reported feature associated with a macrobenthic community of particular species or assemblages is sediment type (Coblentz *et al.*, 2015, Peterson & Peterson, 1979;). In T2, sediment distribution exhibited variability among the stations. Based on statistical values (mean, sorting, and skewness), most of the sediment distribution in T1 and T2 consisted of coarse, medium, fine, and very fine sand with more prevalence of medium sand. Gooday *et al.* (2010) stated that, sediment habitat heterogeneity can be generated by hydrodynamic features such as bottom currents and biological effects such as bioturbation activity, which in turn creates variability of species distribution, species composition, and species diversity. Linear regression showed that the species diversity value increased with a decrease in the sediment mean value towards coarser sand. This was in agreement with Lastra *et al.* (2006) who reported that grain size significantly affected the community characteristic in which coarse sand sediment resulted in a low number of species and species density than finer sand. This suggested that species diversity of intertidal macrobenthos was affected by sediment properties in this transect.

The variability of benthic Chl *a* in intertidal sediments was investigated by several authors at various spatial and temporal scales (Magni & Montani, 2006; Sin, Ryu, & Song, 2009). Sediment microalgae (Chl *a*) play an

important role both as food for benthic organisms and source of nutrients for the overlying water column after decomposition (Fabiano & Danovaro, 1994; Josefson, Forbes, & Rosenberg, 2002; Magni, Abe, & Montani, 2000). The biomass of sediment Chl *a* ranged from 1.73 to 83.05 $mg.m^3$ in this transect and was relatively high compared with other intertidal systems such as Kwangyang Bay, Korea (4.4 to 81.2 $mg.m^3$) and Seto Inland Sea, Japan (4 to 25 $\mu mol.g^1$). The comparison of the Chl *a* concentrations in the surface sediment in this study provides further general information to evaluate the extent of environmental variability at ebb tide of a tropical intertidal flat of Malaysia in particular. Cook, Butler & Eyre (2004) reported that low primary productions were observed more at coarse sand sediments (high water energy) than fine grain sand. In contrast, Sin *et al.* (2009) reported that grain size was not a major factor controlling the biomass of benthic microalgae (Chl *a*). In this study, the sand was composed of medium sand, fine sand, and very fine sand but the concentration of Chl *a* varied. However, further studies including mesocosm experiments are required for a better understanding of the direct effects of sediment Chl *a* on the macrobenthic community in Buntal Bay.

5. Conclusions

The present study showed that sediment granulometry and interstitial water salinity was a significant explanatory factor in the structure of the macrobenthic community in T2. However, in T1 the best observable correlation that influenced the community structure was dissolved oxygen. The findings of the present study suggest that the environmental variables related to macrobenthos examined in a small spatial scale resulted in different community structures and environmental factors that regulate the pattern of the community.

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