



An Ecological Study on Diversity and Composition of Macrobenthos at the Mouth of the Panadura Estuary in Sri Lanka with Special Reference to Water Quality

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Abstract

The ecological surveys with special reference to the dynamic of macrobenthos in the estuaries have not been comprehensively studied in Sri Lanka. Hence, this study was conducted to determine the correlation between physicochemical parameters and macrobenthos diversity and evaluate the pollution level in the estuary using macrobenthos diversity. Macrobenthos and bottom water samples were collected from randomly selected 19 sampling locations using a Grab and Ruttner sampler respectively from May to July 2015 every month. The results revealed that the macrobenthos consist of 13 families belonging to the phylum Mollusca and Arthropoda. The most abundant families were Planobiidae (12200 individuals/m²), Veneridae (11400 individuals/m²), and Haminoidae (11000 individuals/m²). A total of 21 species belonging to 15 gastropoda; five bivalvia and one crustacea were recorded. The *Venerid* sp and *Muricid* sp were the most abundant (7240 individuals/m²) and the least abundant (520 individuals/m²) species, respectively. The highest species richness and diversity were reported at PE7, exposed to the canal which mainly carried hospital wastes. The lowest species richness and diversity were reported at PE6, consisting of an underlying rock layer, making it difficult for sediment collection. Macrobenthos diversity showed that the sampling locations were separated into several clusters. BOD and nitrate-nitrogen were the main physicochemical parameters affecting macrobenthos diversity based on the correlation analysis. CCA showed that macrobenthos assemblage was affected by the convergence of anthropogenic and natural stressors presented at the lower and upper estuary parts, respectively. Furthermore, H' index and Pollution Tolerance Index in the estuary were between 2 and 3 and ≤ 20 , respectively. Both indices confirmed that the water quality of the estuary was in poor condition. This study is important to determine the baseline information of the Panadura estuary's macrobenthos.

Keywords Macrobenthos · Diversity · Ecological indices · Pollution

Introduction

A biological assessment is an evaluation of the health of a water body using biological indicators species (Engel and Voshell 2002; Parmar et al. 2016). Biological indicators can be aquatic plants and animals that are susceptible to specific types and levels of pollutants (Christiane and

Fernando 2014). Aquatic plants, algae, fishes, and macrobenthos can be used as biological indicators for bioassessment (Barbour et al. 1999; Iliopoulos-Georgoudaki et al. 2003). Certain macrobenthos are considered successful indicators (Duran and Suicmez 2007; Ghosh and Biswas 2015; Selvanayagam and Abril 2016) that are useful in assessing the water quality of specific aquatic ecosystems (Lewis 2016; Selvanayagam and Abril 2016; Kumari and Paul 2020). The macrobenthos are important fauna in estuarine ecosystems and are a major link in the aquatic food chain by providing essential food sources for many organisms. They are especially of great significance for fisheries and act as a food source for bottom-feeding fish (Sharma et al. 2010; Sharma et al. 2013). The species richness of macrobenthos could be altered due to anthropogenic disturbances (Moreyra and Padovese-Fonseca 2015).

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Bolgoda Lake opens into the sea through the Panadura estuary in Sri Lanka. Bolgoda Lake comprises of two vast lakes namely, Bolgoda North Lake and the Bolgoda South Lake (Silva 1996; Dissanayake and De Alwis 2019a). Panadura estuary is exposed to inorganic pollution by chemicals and pesticides from the Rathmalana industrial zone and effluents from apparel industries (Joseph 2003; Dissanayake and De Alwis 2019a). It has resulted in a reduction of fish population and lesions in a few fish species (Dassanayake et al. 1991). More than 50 species of edible fish were found in Bolgoda Lake (Pathiratne et al. 2009), whereas Dissanayake and De Alwis (2019a, b) reported a total of 28 fish species, consisting of 13 inhabiting freshwaters, brackish, and seawater; two exclusively freshwater, two exclusively marine, one exclusively brackish, four marine-brackish and six freshwater-brackish. Also, Mysids in the Panadura estuary were identified as an important food source for fish (Punchihewa et al. 2016). Although Panadura estuary was known to possess ecological and economic values, it has increasingly suffered from anthropogenic activities, thereby contributing significantly to the deterioration of the water quality (Piyadasa and Chandrasekara 2010; Dissanayake and De Alwis 2019a).

Several studies have been conducted globally on benthic communities as bioindicators for assessing the water quality of estuaries. The earliest study carried out in Sri Lanka were on brackish water benthic polychaetes by Pillai (1958) and De Silva (1964). Dahanayaka and Wijeratne (2006) also investigated the diversity of the macrobenthic community in the Negombo estuary, Sri Lanka with special reference to environmental conditions while other researchers have also investigated the physicochemical parameters in the same estuary (Chandrasekera and Hettiarachchi 2011). However, the ecological surveys with special reference to the dynamics of benthic communities in estuaries have not been comprehensively studied in Sri Lanka (Dahanayaka and Wijeratne 2006). The North Lake of the Bolgoda has a high potential for ecotourism, recreation activities and aquaculture practices when compared with other estuaries and lagoons in Sri Lanka (Silva et al. 2013). Therefore, this study offers novel information about macroinvertebrates and pollution in a river estuary. The main objective of this study to identify the diversity of the macrobenthic community in the tropical estuary of Panadura, Sri Lanka. Also, the relationship between macro-benthic invertebrate diversity and physicochemical parameters was evaluated to determine the pollution level of the estuary with respect to the diversity of benthic macroinvertebrates.

Materials and Methods

The Study Site

This survey was carried out at the Panadura estuary in Sri Lanka. Panadura estuary is situated in the Western Province

of Sri Lanka ($6^{\circ}42'58.13\text{ N}$ and $79^{\circ}54'1.99\text{ E}$) from which the Bolgoda Lake opens into an ocean. Panadura estuary receives freshwater mainly from Bolgoda Lake.

A preliminary field investigation was carried out to identify the possible sampling locations within the estuarine environment. Sampling was carried out from May to July, 2015 on a monthly basis. A total of 19 (PE1 to PE19) sampling locations were selected using random sampling techniques with special reference to the middle and edge of the river. The map of the sampling locations in the Panadura estuary is illustrated in Fig. 1. The sampling location, PE 1 was located in the middle of the sea's mouth. Also, the sampling locations PE 2, PE 10, and PE 9 were situated on a linear transects from the Panadura premises (right bank), respectively to the other bank. The PE 10 location was located approximately 135 m from the estuary's mouth. Sampling locations namely PE 2, PE 3, PE 4, PE 6, PE 13, PE 12, and PE 11 were located on the south edge of the bank and were at 136 m, 231 m, 348 m, 456 m, 592 m, 808 m, and 1017 m, respectively from the sea mouth. Furthermore, sampling locations; PE 5, PE 14, PE 15, and PE 16 were situated 226 m, 585 m, 753 m, 753 m, and 1033 m approximately distance from the estuary's mouth through the middle of the river. In addition, sampling locations namely PE 9, PE 8, PE 7, PE 19, PE 18 and PE 17 were located at the west edge of the bank and their approximate distances from the estuary's mouth were 178 m, 310 m, 526 m, 940 m, 1188 m, and 1340 m, respectively.

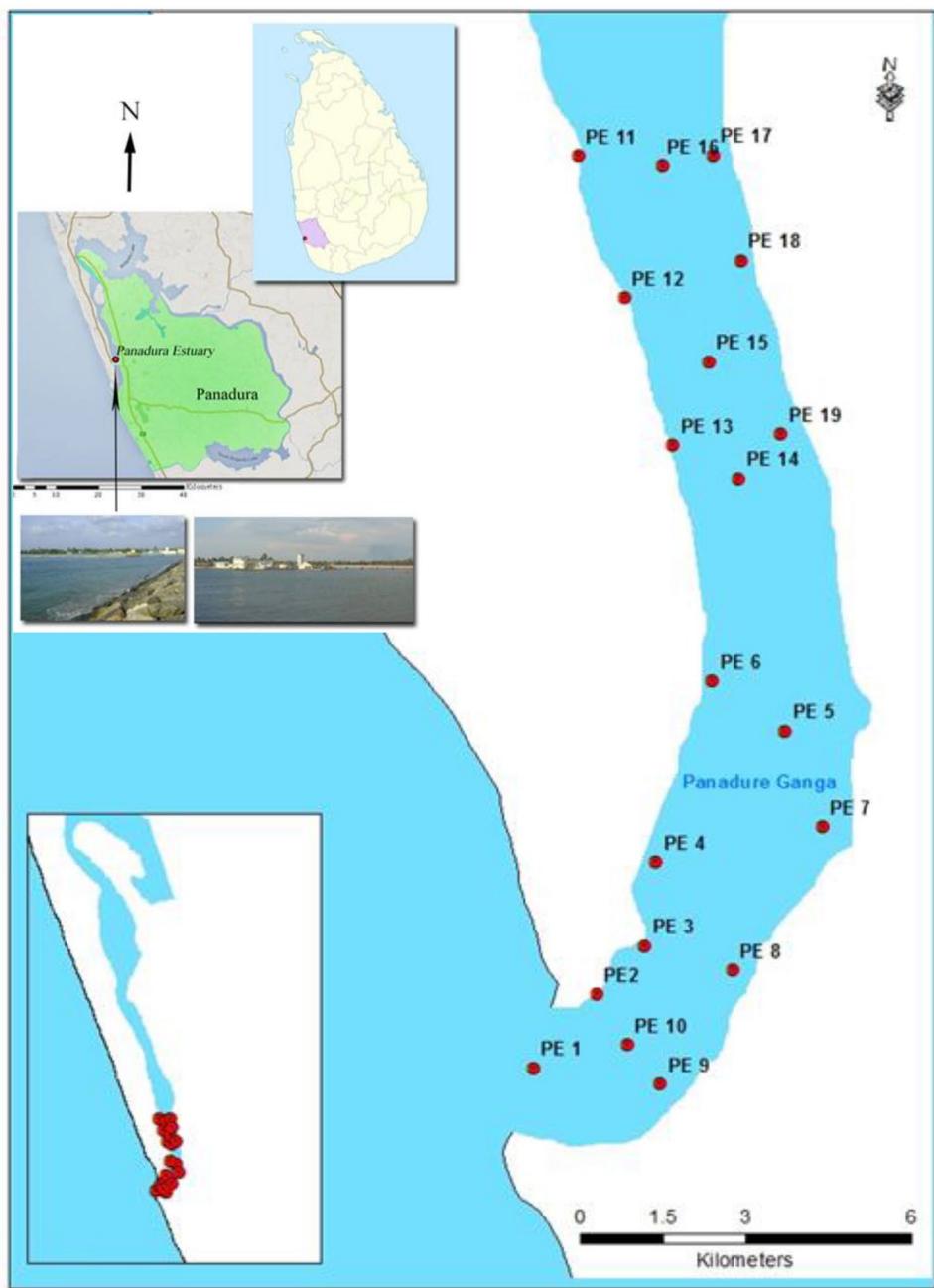
The Global Positioning System (GPS) coordinates of different locations as well as their site descriptions are indicated in Table 1.

Both sediments and water samples were collected from estuary and next moved to PE2, PE10, PE9, PE8, PE4, PE6, PE5, PE7, PE13, PE14, PE19, PE15, PE12, PE18, PE17, PE16, PE11 for easy transportation.

The Analysis of Macrobenthos

Sediment samples were collected from selected locations during May to July, 2015 using a Van Veen Grab sampler. Duplicate sediment samples were taken in each location from different sites and final water sample (composites sample) was generated from these duplicate water collections from 2 sites (APHA 2012). The surface area of each sampling location was about 250 cm^2 . Sediment samples were preserved using Rose Bengal and 70% of alcohol (Zakaria and Mohamed 2018). The alcohol is suitable for preservation of macrobenthos instead of buffered formalin because of faster fixation, optimal preservation, and safer workplace environment (Rahman et al. 2022). Then, samples transferred to the

Fig. 1 Map of sampling locations in the Panadura estuary



laboratory for identification on day of the sample collected. In the laboratory, the sediment samples were wet sieved using a 0.5 mm sieve and macrobenthos were sorted from unwanted materials in the sediment using forceps (APHA 2012). Then, the macrobenthos were stored in 70% ethanol for subsequent examination. The benthic macro-invertebrates were identified to the nearest possible taxonomic category using hand lances and standard identification keys provided by Jessup et al. (2002) and Bouchard (2004).

The Analysis of Water Quality

Water quality parameters namely pH (Orion 260 A), Salinity, Electrical Conductivity (EC), Total Dissolved Solids (TDS) (Hanna HI 8733), Dissolved Oxygen (DO) (Orion 830 A), and turbidity (Hach 2100P) were measured *in-situ* using portable meters. Water samples were collected using a Ruttner sampler and duplicate water samples were taken from each sampling location. All the water samples collected were transported to the laboratory on day of the sample collected,

Table 1 GPS coordinates of sampling locations and their site descriptions

Sampling location	GPS position	Site description
PE1	6°43'0.69 N 79°53'54.44 E	Middle of the estuary mouth
PE2	6°42'58.7 N 79°54'06.8 E	Disposal of sewage, Boat lodging site
PE3	6°43'00.9 N 7°54'09.0 E	Disposal of sewage
PE4	6°43'94.7 N 7°54'09.0 E	Left side after the Bolgoda railway bridge
PE5	6°43'10.7 N 7°54'15.4 E	Middle of the river (in between PE6 and PE7)
PE6	6°43'06.3 N 7°54'17.1 E	Housing area and vegetation patches present (Other side of the river from the UC swimming pool)
PE7	6°43'12.7 N 7°54'17.7 E	Disposal of sewage, near to the UC swimming pool
PE8	6°42'59.8 N 7°54'13.0 E	Disposal of sewage, Boat lodging site
PE9	6°42'54.6 N 7°54'09.7 E	Disposal of sewage, Boat lodging site
PE10	6°42'56.4 N 7°54'08.2 E	Approximately 135 m from the estuary's mouth (in between PE2 and PE9), before the Bolgoda railway bridge
PE11	6°43'16.4 N 7°54'12.5 E	Disposal of sewage, Boat lodging site
PE12	6°43'30.4 N 7°54'08.1 E	Mangrove presence,
PE13	6°43'23.7 N 7°54'10.3 E	Mangrove presence, Boat lodging site
PE14	6°43'22.2 N, 7°54'13.3 E	Mangrove presence
PE15	6°43'27.5 N 7°54'11.9 E	Mangrove presence
PE16	6°43'36.4 N 7°54'09.8 E	Middle of the river (in between PE 11 and PE17)
PE17	6°43'36.9 N 7°54'12.1 E	Near to the Panadura Bridge and private hotel
PE18	6°43'32.1 N 7°54'13.4 E	Disposal of sewage
PE19	6°43'24.2 N 7°54'15.2 E	Mangrove presence

and stored at 4 °C (APHA 2012). The laboratory analysis was carried out as soon as possible after take the water samples to the laboratory to prevent any changes (APHA 2012). In the laboratory, Orthophosphate, Ammoniacal - N, Nitrate - N, Nitrite - N, were measured using the colotrimetric method whereas Total Suspended Solids (TSS) and Biochemical Oxygen Demand (BOD) were measured using gravimetric and titrimetric methods respectively according to standard procedures of Examination of Water and Waste Water, 20th edition (APHA 2012).

Data Analysis

Benthic macroinvertebrate diversity was calculated using the Shannon-Wiener diversity index (H') (Magurran 1991) (Eq. 1)

$$H' = -\sum \pi_i \ln \pi_i \quad (1)$$

$$i = 1$$

where,

H' Species diversity index,

S Number of species,

π_i Proportion of individuals of each species belonging to the “ i ”th species of the total number of individual

while the richness of macrobenthos was calculated using Margalef's index (Eq. 1).

$$\text{Margalef's richness index} = (S - 1) / \ln(n) \quad (2)$$

where,

S the number of taxa,

n the number of individuals

The Pollution Tolerance Index (PTI) was calculated to determine the health status of the locations using sum of the score value of the taxa (> 40 is Good; 20–40 is Fair; < 20 is Poor) (Mitchell and Stapp 1997).

All statistical analysis was performed using Minitab 17 and Primer 7 statistical soft-ware. The normality test was carried out using Andersan darling test. The relationship of physicochemical parameters of water with Shannon-Wiener diversity index (H'), Margalef's richness index and Pollution Tolerance Index (PTI) were determined by using Pearson correlation. The One-way ANOVA test was carried out to analyze the variation of physicochemical parameters between sampling locations. Similarities of benthic communities between selected sampling locations were analyzed using the Bray-Curtis similarity clustering method (Clarke and Warwick 2001). Moreover, the physicochemical parameters of water between 19 sampling

locations were analyzed separately using Principal Component Analysis (PCA) to find out the key physicochemical parameters of each location. Furthermore, the Canonical Correspondence Analysis (CCA) was performed using the CANOCO 4.5 statistical software to determine the water quality influence on the macrobenthos and the sampling sites. Before analyzing data, it is essential to apply the log-transform via $\text{Log}_{10}(x+1)$ to correct the departures from the normal distribution. Therefore, the abundance data of the macrobenthos were log-transformed during the CANOCO program. The triplot ordination diagram was produced using CanoDraw.

Results

Benthic Macroinvertebrates

The identified macrobenthos during the survey are summarised in Table 2. A total of 21 species of macrobenthos belonging to two phyla (Mollusca and Arthropoda), three classes (Gastropoda, Bivalvia, and Crustacea), and 13 families were recorded from the Panadura estuary. Gastropoda were the dominant group in many sampling locations,

representing 64% followed by the bivalvia (35%). Crustacea were the least group representing 1% of the total macrobenthos and were limited to certain locations. However, their distribution was not uniform in the selected sampling locations. Nine families were recorded from Class Gastropoda and three families were recorded from Class Bivalvia. Only one family was recorded from Class Crustacea (Table 2).

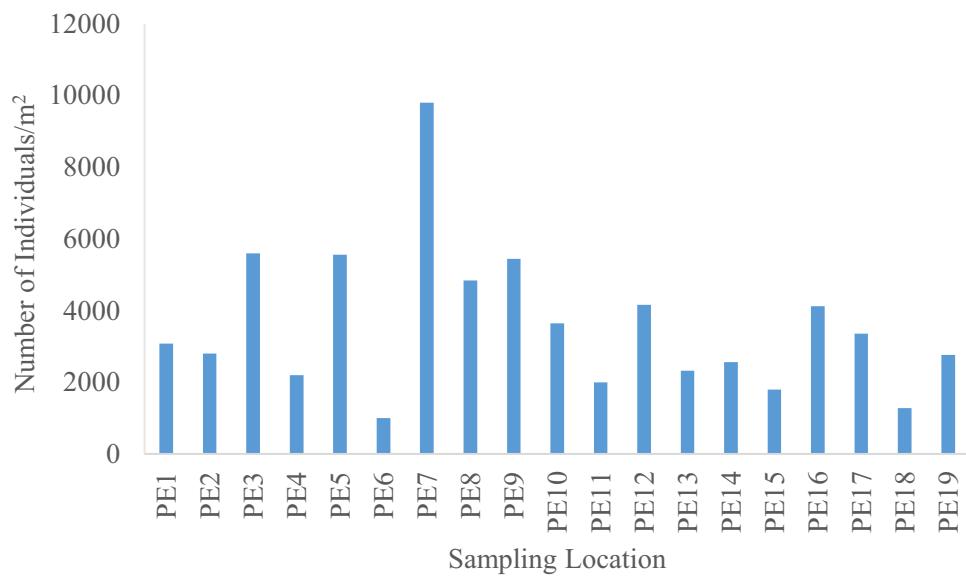
The number of individual macrobenthos in each sampling location at Panadura estuary during the study period is illustrated in Fig. 2. Comparatively, the highest and lowest number of individuals of gastropoda were reported at PE 7 location and PE 6 location, respectively.

Comparatively, the highest and lowest relative abundance of gastropoda were reported at locations PE6 and PE12, respectively during the study period (Fig. 3). Also, the highest composition of gastropoda was recorded at PE5 and PE6 whereas the lowest composition of gastropoda was recorded at PE14 and PE15. Planorbidae was the most dominant gastropoda family recorded (12,200 individuals/ m^2) during the study period. The highest relative abundance of Planorbidae was recorded at the sampling location, PE2 and recorded at all sampling locations, except PE4. Haminoeidae was the second dominant bivalvia family and the highest relative abundance of this family was recorded at

Table 2 Macrofauna recorded from Panadura estuary

Phylum	Class	Family	Species	No of individuals per sp./ m^2	No of individuals per family/ m^2	Relative Abundance of Families
Mollusca	Gastropoda	Haminoeidae	<i>Haminoeid</i> sp 1	5560	11000	16.10
			<i>Haminoeid</i> sp 2	5440		
		Muricidae	<i>Thais tissoti</i>	1000	1520	2.22
			<i>Muriciid</i> sp 1	520		
		Littorinidae	<i>Littoraria undulata</i>	1160	2680	3.92
			<i>Nodilittoria trochooides</i>	1520		
		Potamididae	<i>Cerithidae cingulata</i>	2640	2640	3.86
		Cerithidae	<i>Clypeomorus</i> sp	1120	7240	10.59
			<i>Ceruthiid</i> sp 1	2640		
			<i>Ceruthiid</i> sp 2	3480		
		Neritidae	<i>Neritid</i> sp 1	2480	2480	3.62
		Planorbidae	<i>Planorbid</i> sp 1	6440	12200	17.85
			<i>Planorbid</i> sp 2	5760		
		Naasariidae	<i>Naasariid</i> sp 1	1320	1320	1.93
		Thiaridae	<i>Faunus ater</i>	3000	3000	4.39
Bivalvia	Mytilidae		<i>Brachiodontes</i> sp 1	5600	3000	13.75
			<i>Brachiodontes</i> sp 2	3800		
		Veneridae	<i>Meretrix</i> sp	1920	9400	16.68
			<i>Gafrarium tumidum</i>	2240		
			<i>Venerid</i> sp 1	7240		
Arthropoda	Crustacea	Donacidae	<i>Donax cuneatus</i>	2680	2680	3.92
		Coenobitidae	<i>Eupagurus</i> sp	760	760	1.12
Total				59920		100

Fig. 2 Benthic macroinvertebrate abundance at Panadura estuary



the sampling location, PE6. Though, they were recorded at all sampling locations. Nassariidae was the least dominant gastropoda family recorded and they were dominant at the sampling location, PE8.

Similarly, the variation of relative abundance of and composition of bivalvia during the study period at Panadura estuary is presented in Fig. 4. Comparatively, the highest and lowest relative abundance of Bivalvia were reported at PE12 and PE2, respectively (Fig. 4). Except for sampling locations, PE2, PE3, PE6, PE11, PE15 and PE18, other sampling locations were recorded with the same composition of Bivalvia. Among the bivalvia, Family Veneridae was the most dominant and Family Donacidae was the least dominant. Compared to other sampling locations, Veneridae and Muriciidae were reported as the highest and the lowest

relatively abundant Gastropoda family at PE11 and PE15 respectively. Veneridae family was recorded only at the sampling location, PE6.

The seasonal changes of the macrobenthos based on the family level is summarized in Table 3.

Compared to the all the sampling months, higher abundance was in May and lower abundance was in April 2015. A high abundance of Planorbidae was noted from May 2015 and low abundance was recorded from April 2015. Moreover, the highest abundance of Veneridae was recorded in May 2015 (rainy season), and the lowest relative abundance of Veneridae was recorded in April 2015 (dry season). Similar observation was also recorded in Haminoeidae in April and May 2015. In the April, most dominant group was Planorbidae followed by Veneridae,

Fig. 3 Relative abundance of Gastropoda recorded at each sampling location

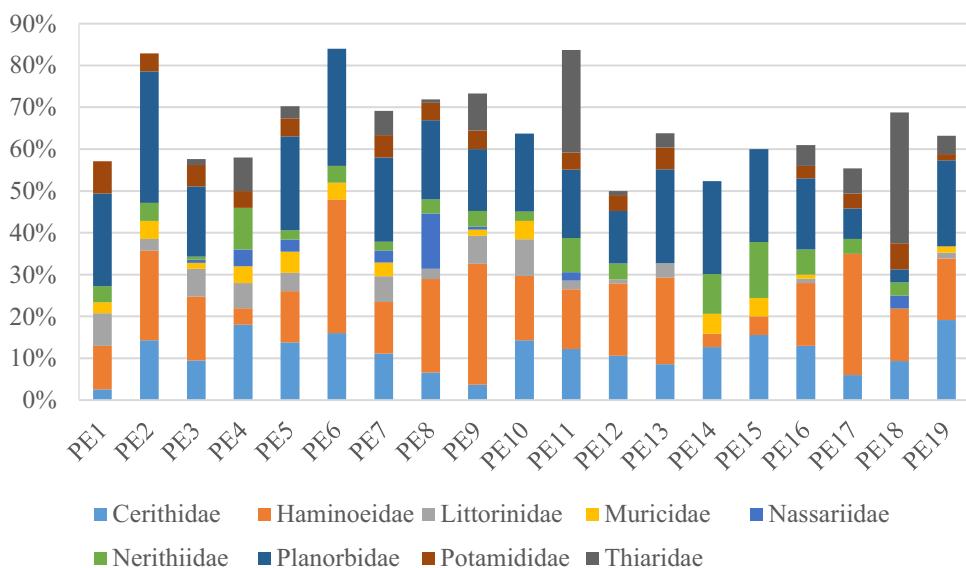
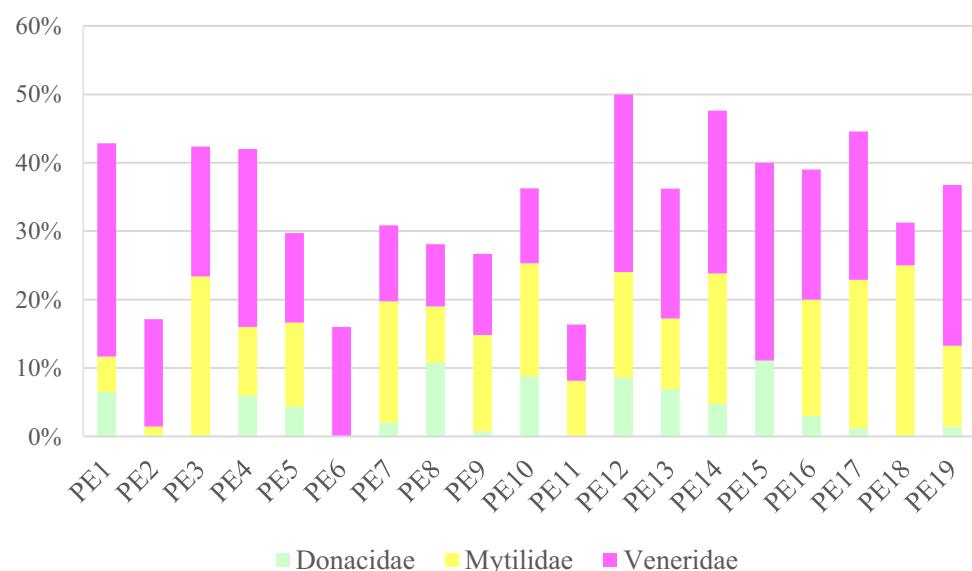


Fig. 4 Relative abundance of Bivalvia recorded at each sampling location



and Haminoidae. However, in dry season, most dominant group was Cerithidae, followed by Mytilidae, and Potamididae (Table 3).

The ANOVA results showed that Veneridae showed the significant difference of benthic macroinvertebrate family's abundance among the sampling months ($p < 0.05$). However, all the other families indicated the insignificant variation of benthic macroinvertebrate family's abundance among the sampling months ($p > 0.05$) (Table 4).

Comparatively, the highest and lowest value species richness was recorded at sampling locations PE3, and PE18, respectively (Table 5). The Shannon-Wiener diversity index (H') of all the selected sampling locations at Panadura estuary ranged from 2 to 3. The maximum and minimum values

of H' and Margalef's index were reported at PE 7 and PE 6, respectively. These values confirmed the results of invertebrate composition in selected locations. Moreover, the PTI index was 20 or less than 20 in all the sampling locations (Table 5).

The seasonal variation of species richness and diversity of the macrobenthos with special reference to the sampling locations are included in Table 6. In April, the highest species richness was recorded at sampling location, PE7, whereas the lowest species richness was recorded at sampling location, PE6. The current study yielded the highest species richness at sampling location, PE7 in May and the lowest species richness at sampling locations, PE6 and PE13 in May. Moreover, the highest species richness was recorded at

Table 3 Seasonal variation of macrobenthos taxa abundance during the study period at Panadura estuary

Family	No of individuals per family/m ²		
	April	May	June
Cerithidae	2520	2360	2360
Coenobitidae	200	360	200
Donacidae	840	840	1000
Haminoidae	3360	3760	3880
Littorinidae	960	760	960
Muricidae	520	400	600
Mytilidae	1380	1000	620
Naasariidae	550	240	520
Neritidae	600	1000	880
Planorbidae	3785	4145	4270
Potamididae	1240	1000	400
Thiaridae	1000	800	1200
Veneridae	1680	3960	3760

Table 4 Results of ANOVA for benthic macroinvertebrate families among the season (sampling months)

Family	Degree of Freedom	P value
Cerithidae	.052	.950
Coenobitidae	1.071	.350
Donacidae	.125	.883
Haminoidae	.138	.871
Littorinidae	.183	.833
Muricidae	1.764	.181
Mytilidae	.265	.768
Naasariidae	.530	.592
Neritidae	.834	.440
Planorbidae	.80	.923
Potamididae	2.698	.76
Thiaridae	.375	.689
Veneridae	6.690	.003

Table 5 Species richness, Shannon- Wiener diversity index (H'), Margalef's Index, and PTI Index of macrobenthos recorded at selected sampling locations of Panadura Estuary

Sampling Location	Species richness	H'	Margalef's Index	PTI Index
PE 1	15	2.48	2.08	17
PE 2	15	2.45	1.88	13
PE 3	20	2.48	2.23	15
PE 4	18	2.78	2.99	16
PE 5	22	2.86	2.63	20
PE 6	10	2.04	1.86	9
PE 7	22	2.87	2.96	20
PE 8	19	2.63	2.50	16
PE 9	20	2.64	2.24	12
PE 10	16	2.53	1.99	17
PE 11	16	2.46	2.57	14
PE 12	17	2.65	1.72	15
PE 13	15	2.46	2.23	13
PE 14	15	2.50	1.92	13
PE 15	12	2.23	2.61	14
PE 16	17	2.62	2.16	16
PE 17	16	2.45	2.03	14
PE 18	11	2.07	2.01	13
PE 19	17	2.52	0.58	13

sampling location, PE7, in June whereas the lowest species richness was recorded at sampling location, PE18.

Figure 5 shows the cluster analysis based on the Bray-Curtis similarity matrix. Benthic macroinvertebrate communities of locations PE 11 and PE 18 were clustered together at a 72% similarity level in the first cluster. These results signified that both PE 11 and PE 18 were inhabited by a macroinvertebrate community in which the composition was more or less similar to each other. The benthic communities in PE 8, PE 3, PE 9, PE 5, and PE 7 clustered together at 78% similarity level. Benthic communities in PE 1, PE 2 and PE 10 clustered together at a 78% similarity level, while benthic communities in PE 12, PE 13, PE 14, PE 15, PE 16, PE 17, and PE 19 aggregated together at a similarity level of 75%.

Water Quality

The minimum and maximum concentration of DO were 4.29 mg/L and 8.41 mg/L respectively. The BOD₅ concentration ranged from 1.00 mg/L-1 to 23.5 mg/L across all sites. Also, total suspended solids concentration ranged from 6.00 mg/L to 112.90 mg/L. Moreover, minimum and maximum concentration of ammoniacal-nitrogen were 0.22 mg/L to 1.52 mg/L (Table 7). The One-way ANOVA analysis indicated that physiochemical parameters; pH, turbidity, TSS, DO, EC, BOD, nitrite-nitrogen, orthophosphate and ammoniacal-nitrogen concentration varied significantly among the sampling

Table 6 Seasonal variation of species richness and diversity of the benthic macroinvertebrates

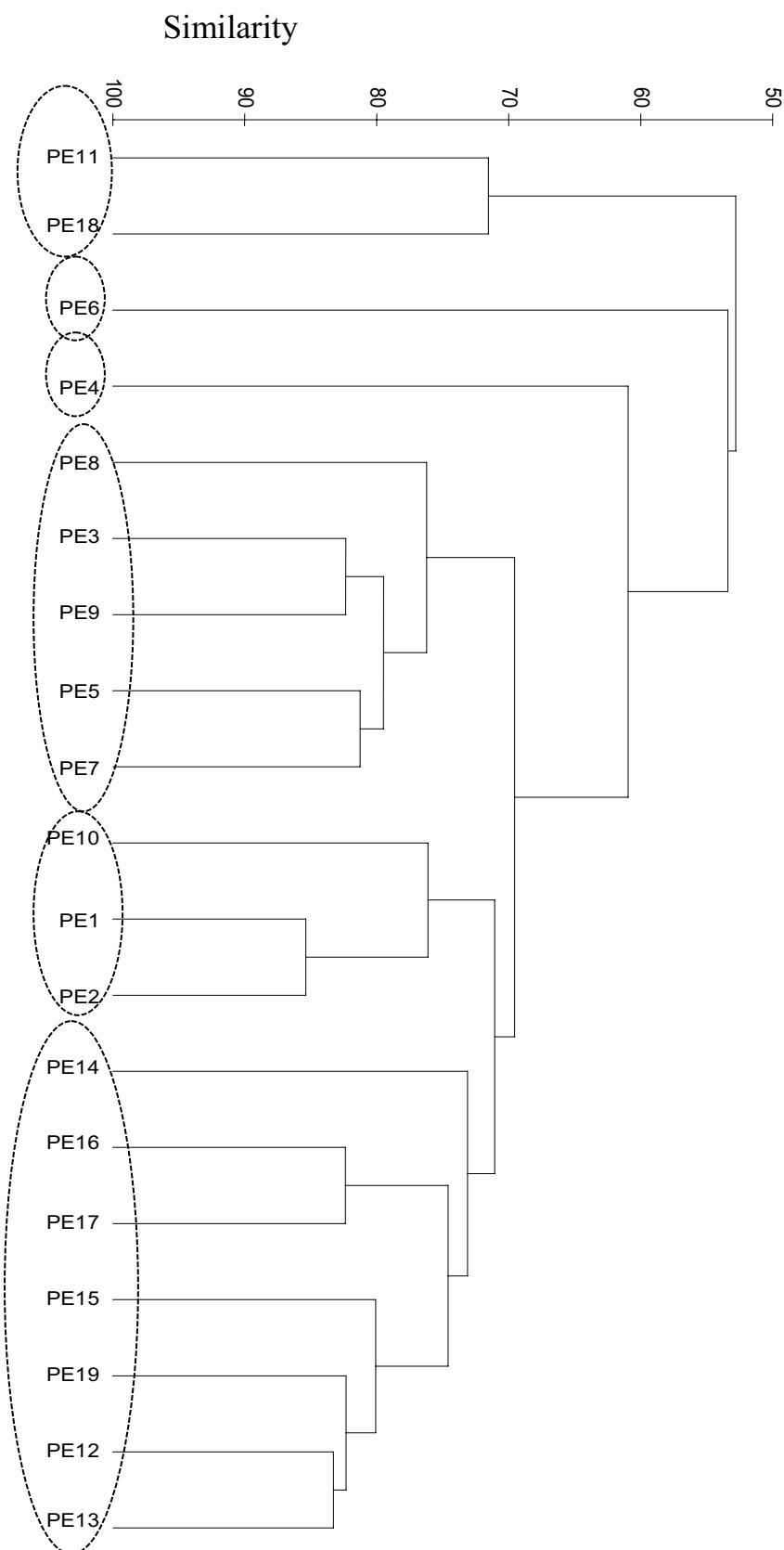
Sampling Location	April		May		June	
	Species richness	H'	Species richness	H'	Species richness	H'
PE 1	11	1.80	14	1.97	11	1.65
PE 2	11	1.56	12	1.67	13	1.94
PE 3	17	2.02	15	1.82	18	2.07
PE 4	14	2.28	12	1.97	12	2.02
PE 5	20	2.08	19	2.26	18	2.24
PE 6	5	1.01	7	1.33	6	1.49
PE 7	22	2.39	21	2.33	19	2.21
PE 8	18	2.05	14	2.11	11	1.79
PE 9	19	2.32	15	1.97	14	1.90
PE 10	15	2.04	8	1.94	11	2.02
PE 11	14	1.98	9	1.86	4	0.89
PE 12	16	2.04	12	1.83	13	1.82
PE 13	12	1.98	7	1.62	8	1.64
PE 14	13	1.34	8	1.62	9	1.49
PE 15	7	1.08	8	1.79	5	1.55
PE 16	14	1.92	14	2.16	13	1.89
PE 17	11	1.60	9	1.47	10	1.71
PE 18	10	1.85	8	1.81	2	0.69
PE 19	15	0.27	12	1.69	6	1.53

locations ($p < 0.05$). However, the nitrate-nitrogen concentration, water temperature, and salinity were not significantly different among the sampling locations ($p > 0.05$) (Table 7).

Tukey pairwise comparison (after one-way ANOVA test) revealed that, sampling location, PE1 has higher average DO in compared to location PE 11 – PE 19. Also, the average BOD in PE 1 was higher than PE 2, PE 3, PE 4, PE 5, PE 7, PE 12, PE 13, PE 15 and PE 18. Similarly, there was a higher average pH value of PE 13 compared to locations PE 1, PE 2, PE 3, PE 4, PE 6, PE 8, and PE 9. The higher significant differences were observed in the average TSS in PE 1 compared to PE 12, PE 13, and PE 19 sampling locations, where mangrove distribution could be seen. The sampling location, PE1 has higher average TSS compared to PE 10, and PE 16, which was lacking any disposal canal. The higher significant difference was observed in average ortho-phosphate in PE11 compared to PE 18. Additionally, the sampling location PE11 has higher average ammoniacal-nitrogen in PE 11 in comparison to locations PE 13, PE 14, and PE 16. Also, the parameter showed higher significant different in PE 12 compared with PE 13, PE 14, PE 16, and PE 17. Sampling locations, PE 13 and PE 14 were located near mangrove areas and PE 16 and PE17 were not exposed to disposal canals.

The correlation matrix of PCA revealed many water quality parameters had 0.3 coefficients and above. The

Fig. 5 The Cluster diagram showed the macrobenthos between the different sampling locations in Panadura estuary



Kaiser-Meyer-Olkin (KMO) test value and the χ^2 of Bartlett's test were 0.599 and 144.279, respectively, at a significance level of $p < 0.05$ (Table 8). The KMO value exceeded the threshold of 0.6, indicating the suitability of the dataset for PCA. Table 8 shows the eigenvalue of the first, the second, third and the fourth components exceeded one, accounting for 11.205%, and 8.228% (jointly 76.125%) of the total variance, effectively assigning the data into four major components. Table 8 shows the loading of the varimax-rotated-component matrix, in which the first component is usually more correlated to the variables than the second component (Liu et al. 2003).

The varimax rotated component matrix was loaded for each water quality parameter (Table 8). In general, component loadings higher than 0.6 are used to evaluate the extracted components in a PCA (Tashtoush 2015). Typically, component loading levels of 0.75, between 0.75 and 0.50, and between 0.50 and 0.30 are considered high, moderate, and weak, respectively (Liu et al. 2003). The first principal component showed strong positive loading on DO, salinity, and EC but moderate positive loading on pH, TDS. Furthermore, water temperature showed strong negative loading on the first component. Meanwhile, the turbidity, ammoniacal-N, and nitrite-nitrogen showed strong positive loadings on the second principal component so on (Table 8).

The results of the Principal Component Analysis (PCA) between physicochemical parameters during the study period are illustrated in Fig. 6. The PCA revealed that the sampling location of PE 19 was characterized by the highest nitrate concentration (0.0202 ± 0.0016 mg/L) during the study period. Also, PE 11 was characterized by high levels of turbidity (19.60 ± 3.14 NTU). Sampling locations, PE14 and PE17 were clustered together and characterized by a high BOD (PE14 = 12.67 ± 9.50 mg/L;

Table 8 Principal components (PC) and varimax rotated component matrix

Eigenvalue explained by PCs				
	5.776	1.594	1.457	1.070
Percentage of total variance explained				
	44.432	12.260	11.205	8.228
Component matrix				
Variables	PC1	PC2	PC3	PC4
Water Temperature	-.857	.249	.174	.119
pH	.707	-.320	-.074	.441
DO	.854	-.286	.271	-.082
Turbidity	-.389	.753	.202	.110
Salinity	.749	-.081	.066	-.008
EC	.837	-.052	-.009	-.284
TDS	.660	-.334	-.187	.055
Ammonia-N	-.056	.835	-.041	.137
Nitrate-N	-.556	.106	-.529	.369
Nitrite-N	-.232	.918	-.172	.017
Ortho- Phosphate	-.254	.440	.055	.734
BOD ₅	-.205	.078	.774	.004
TSS	.373	-.416	.602	.381

PE17 = 22.50 ± 1.00 mg/L). Furthermore, sampling locations of PE10 and PE4 were clustered together and characterized by low nitrate-nitrogen concentration. Recorded nitrate nitrogen concentration was 0.012 ± 0.000 mg/L and 0.015 ± 0.000 mg/L in sampling locations PE4, and PE10, respectively. Additionally, PCA analysis confirmed that location PE1 was characterized by high dissolved oxygen concentration (8.41 mg/L) compared to other sampling locations (Fig. 6).

Table 7 Overall water quality results and One way-ANOVA results of water quality at Panadura estuary

Parameter	Minimum	Maximum	Mean \pm SD	F value	p value
Water Temperature (°C)	29.30	32.30	30.45 ± 0.90	1.00	0.483
pH	7.09	8.61	8.21 ± 0.24	0.88	0.000
Dissolved Oxygen (mg/L)	4.29	8.41	5.70 ± 1.32	3.83	0.000
Turbidity (NTU)	8.45	26.50	1.12 ± 5.80	2.86	0.004
Salinity (ppt)	32.6	39.00	31.10 ± 2.12	0.64	0.823
Total Suspended Solids (mg/L)	6.00	112.90	41.05 ± 22.24	2.20	0.021
Electrical Conductivity (mS/cm)	15.78	51.70	43.23 ± 10.03	1.91	0.049
Total Dissolved Solids (mg/L)	110.20	3456.00	1235.70 ± 987.40	33.78	0.000
Biochemical Oxygen Demand (mg/L)	1.00	23.50	9.42 ± 6.05	6.72	0.000
Ortho- Phosphate (mg/L)	0.03	5.40	0.38 ± 1.09	2.58	0.008
Nitrite Nitrogen (mg/L)	0.001	0.03	0.001 ± 0.09	2.66	0.006
Nitrate Nitrogen (mg/L)	0.001	0.03	0.01 ± 0.01	0.51	0.935
Ammoniacal-Nitrogen (mg/L)	0.22	1.52	0.07 ± 0.30	3.60	0.001

Correlation Between Macrobenthos Indices and Water Quality

Correlation analysis (Table 9) demonstrated that, BOD ($r = -0.370$; $p = 0.046$) and nitrate concentration ($r = -0.308$; $p = 0.027$) had a negative significant relationship with the Shannon - Wiener diversity index. Also, BOD had no significant relationship with Margalef's index ($r = -0.237$; $p = 0.082$). Likewise, the BOD ($r = -0.210$; $p = 0.189$) and water temperature ($r = -0.208$; $p = 0.127$) were not significantly associated with the PTI index (Table 9).

Figure 7 shows the CCA triplot with eigenvalues of 0.050 and 0.035 for the first and the second axes, respectively. These two axes described the higher percentage of the families' variance in the study and their relationships with the environment parameters (TSS and BOD) assigned to the first (x) axis. The second (y) axis included the DO, pH, water temperature, Salinity, EC, turbidity, ammoniacal-nitrogen, nitrite-nitrogen, turbidity, and ortho-phosphate. The first axis explained 19.6% of the variance, while the second was only 13.8%. However, the third and the fourth eigenvalues axes variance were comparatively low, i.e., at 0.030 and 0.017, respectively. Additionally, the percentages for their relation to species and environmental parameters were only 11.6% and 6.8%, correspondingly.

As per Fig. 7, macrobenthos were labeled using Arabic numbers. Therefore, the assigned numbers for macrobenthos

were included after each taxa name for easy identification. Group A was prominent in the CCA triplot, and consisted of several families (Fig. 7). Among them, Littorinidae (Baroudi et al. 2020), Muricidae (Bassey et al. 2020), and Planorbidae (Bouchet and Rocroi 2005) were positively related to a high concentration of DO, pH, salinity, and moderate concentration of EC and TSS presented at sampling locations, PE1, PE4-5, PE9-10 (lower part of the estuary).

Group B was distinguished with the high abundance of Cerithidae (Afshan et al. 2013), Coenobitidae (American Public Health Association (APHA) 2012), Haminoeidae (Barbour et al. 1999), Nerithiidae (Bouchard 2004), Thiaridae (Christiane and Fernando 2014), Donacidae (Clarke and Warwick 2001), Mytilidae (Dahanayaka and Wijeyaratne 2006), and Veneridae (Dassanayake et al. 1991) were affected by a high value of water temperature, turbidity and high concentration of nitrate-nitrogen and moderate concentration of BOD, ammoniacal-nitrogen, nitrite-nitrogen, and ortho-phosphate presented at sampling locations, PE11-14, PE16-19 (upper part of the estuary).

Discussion

Macrobenthos are considered the best indicators for bio-monitoring (Sharma et al. 2010). Diversity, abundance, and composition of benthic macro-faunal communities can be

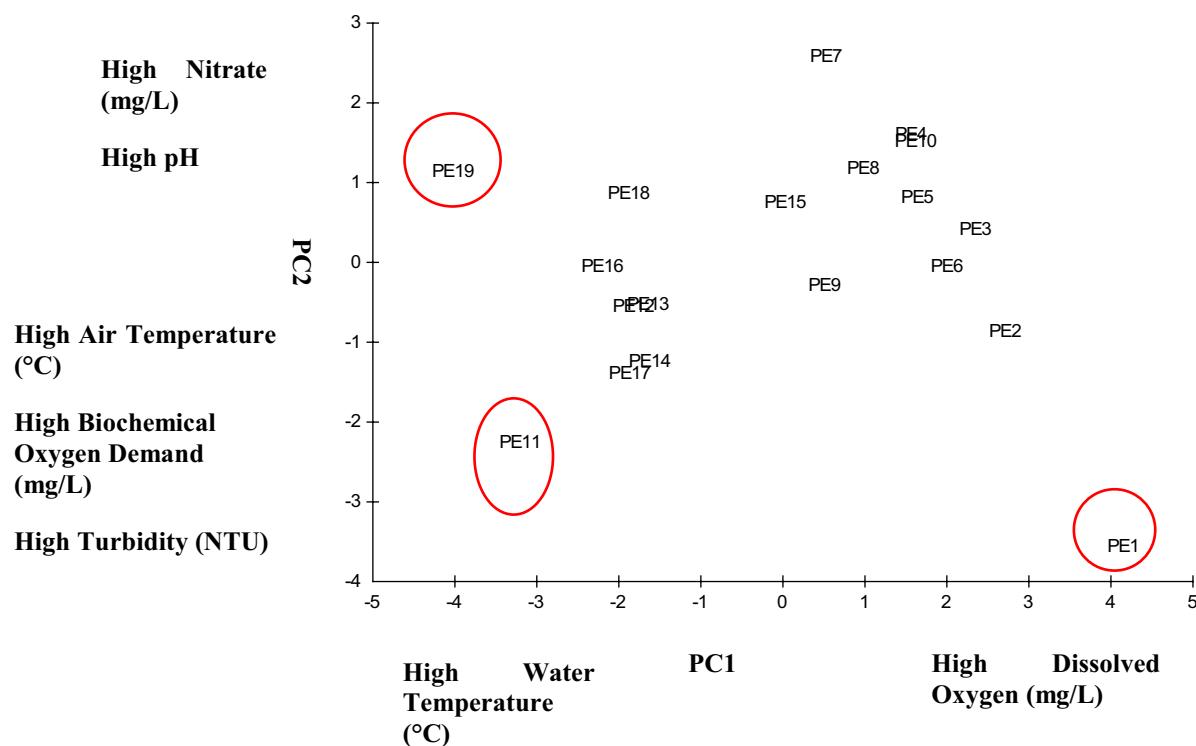


Fig. 6 Ordination of the 19 sampling locations based on PC1 and PC2 scores of Principal Component Analysis between physicochemical parameters during the study period

Table 9 Relationship between Shannon - Wiener diversity index (H'), Margalef's index, PTI, and physicochemical parameters

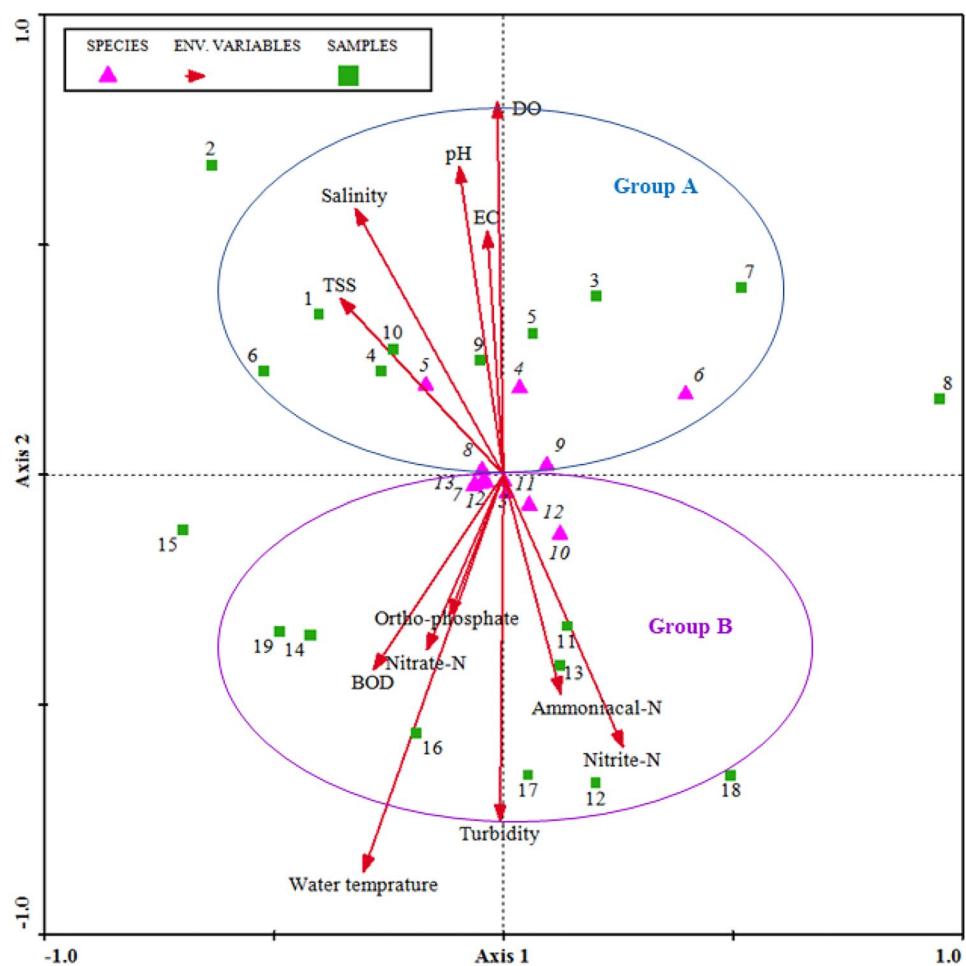
Water quality parameter	H'		Margalef's index		PTI	
	correlation coefficient	P value	correlation coefficient	P value	correlation coefficient	P value
Water temperature	0.021	0.0881	-0.112	0.417	-0.208	0.127
pH	0.044	0.748	0.074	0.594	-0.079	0.567
Turbidity	-0.210	0.276	0.055	0.689	-0.071	0.609
EC	-0.021	0.881	-0.114	0.408	0.064	0.645
DO	0.090	0.541	-0.032	0.814	0.095	0.489
Salinity	0.079	0.568	-0.020	0.888	0.032	0.816
TDS	0.134	0.321	0.032	0.814	0.246	0.065
TSS	-0.199	0.192	-0.008	0.955	-0.043	0.753
BOD	-0.370	0.046	-0.237	0.082	-0.210	0.189
Nitrite - N	0.096	0.421	-0.054	0.695	-0.061	0.657
Nitrate - N	-0.308	0.027	0.099	0.472	-0.028	0.837
Ortho - Phosphate	-0.079	0.566	0.025	0.879	-0.150	0.275
Ammoniacal - N	-0.189	0.346	-0.020	0.888	-0.012	0.933

used to evaluate the water quality of the aquatic ecosystem (Ghosh and Biswas 2015; Selvanayagam and Abril 2016; Lewis 2016; Kumari and Paul 2020). Results of Panadura estuary mouth also convinces this fact as most of the

organisms found in the estuary mouth was pollution tolerant organisms (Table 1). Idroos and Manage (2012) conducted a study at Bolgoda canal and Waga stream with respect to physicochemical parameters and bio indicators. *Planorbid* sp

Fig. 7 Canonical Correspondence Analysis graph indicating the abundance distribution of macrobenthos by water quality (environmental parameters).

Note: The numbers denote various taxa, where 1 = Cerithidae, 2 = Coenobitidae, 3 = Haminoeidae, 4 = Littorinidae, 5 = Muricidae, 6 = Nassariidae, 7 = Neritiidae, 8 = Planorbidae, 9 = Potamididae, 10 = Thiaridae, 11 = Donacidae, 12 = Mytilidae, 13 = Veneridae. Longer arrows denote more decisive influence on the macrobenthos and the sampling sites and vice versa. Samples; Sampling site



(Planorbidae) and *Cerithiidae* sp (Cerithidae) were recorded in the Bolgoda Canal (Idroos and Manage 2012), which is consistent with the present study as both species were also identified in the Panadura estuary.

Mollusks bivalvia are used as a bioindicator of aquatic environmental contamination. Based on the previous studies in the world, bivalvia recorded in this study namely Mytilidae (Oliveira et al. 2016), Veneridae (Liu et al. 2011; Hamza 2019), and Donacidae (Tlili 2018) have identified as bioindicators. Also, gastropoda have a significant role in any aquatic ecosystem because they directly involved in the function aquatic food chain and they can be used as a bioindicator of water quality because it is sensitive to water pollution. Donacidae (Mahmoud and Abu Taleb 2013), Muricidae (Mahmoud and Abu Taleb 2013), Littorinidae (Syahrial and Ezraneti 2021) would be the potential bio indicators recorded this study whereas recorded least abundance of crustaceans (Sanchez-Moyano et al. 2010) in present study proved that they were the most sensitive macrobenthos group to pollution according to the previous literature. However, these references have been belonged to some specific genus or species. Identifying taxa up to species level is difficult due to the lack of taxonomic expertise; hence, the eDNA barcoding method with the power of the next-generation sequencing technique can be applied to species level confirmation to determine the bioindicators.

Based on the results, high and low abundance and diversity were recorded at the sampling site, PE7 and PE6, respectively. The sampling location, PE 7 location is exposed to the canal that is mainly responsible for the discharge of hospital wastes. This location would be an ideal place for the survival of pollution tolerant macrobenthos because of a high amount of organic matter and nutrients (average ammoniacal-nitrogen: 1.392 ± 0.0278 mg/L, average orthophosphate: 0.0278 ± 0.0057 mg/L, average nitrate-nitrogen: 0.0149 ± 0.0007 mg/L) arising from the canal. These water quality parameters insignificantly related to the pollution index ($p < 0.000$). The sampling location, PE 6 location consisted of an underlying rock layer, which made it difficult for sediment collection. In addition, the sampling location PE 6 was devoid of any material in its surrounding (Table 1). This might have affected the diversity of macrobenthos in sampling locations PE 7 and PE 6.

Also results of species richness showed that the highest richness was recorded at sampling location, PE3 whereas the lowest richness was reported at sampling location, PE18. The disposal of sewage was observed in both locations. However, water quality results showed that sampling location, PE3 indicated comparatively the second highest average DO concentration (7.65 ± 0.00 mg/L) among the selected all sampling locations and low average BOD (4.25 ± 0.35 mg/L) and ortho-phosphate concentration (0.0168 ± 0.0233 mg/L). Although, sampling location,

PE18 showed comparatively low average DO concentration (4.66 ± 0.21 mg/L) and high average ammonical-nitrogen concentration (0.1927 ± 0.0915 mg/L). The changes of water quality results in different way in these two sampling locations would be the reason for variation of species richness. According to the study carried out in tropical estuary (Buenaventura Bay), Colombian Pacific, Duque et al. (2019) and Duque et al. (2020) found that a low species richness of fish was reported in low-quality waters due to the high phosphate and nitrate concentrations and the lowest species richness of macrobenthos due to the highest nitrite, nitrate, and DO concentrations and low water temperatures respectively. Similarly, based on the study conducted in Mahanadi estuary, east coast of India. Nayak et al. (2022) confirmed that lower abundance, richness, diversity of species shows the its' environmental stress.

According to the Bray-Curtis similarity matrix of the first cluster, sampling locations PE 11 and PE 18 were exposed to waste drainage canals, these canals carry similar kinds of wastes containing more diverse materials, including nutrients and chemicals. The family of Thiaridae was the highest composition of mollusks-invertebrates recorded in both sampling locations. This family is a common inhabitant of fresh and brackish water sources (Bouchet and Rocroi 2005). Many previous studies proved that Thiaridae is high tolerance against extremes of physicochemical component of water (Tinoco-Perez et al. 2019; Parra et al. 2021). In the second cluster, except PE5, other locations were exposed to disposal canals. More or less similar chemicals, materials, and nutrients could have entered through these canals. More importantly, these discharges might be preferred by the more or less similar macrobenthic community. The sampling location PE5 was not exposed to the disposal canal and this similarity might have been created by other natural or anthropogenic factors, including river flow, sand movement, and currents. The Haminoeidae is common in locations where clustered in second group. Similarly, Dahanayaka and Wijeyaratne found this family in many selected locations in Negombo Lagoon, Sri Lanka.

Regarding the third cluster, PE 2 was exposed to a disposal canal; these accumulated wastes could be flushed away by water flow and turbulence of water as it is located near to the mouth of the estuary like PE 1 and PE 10. As these locations were close to the mouth of the estuary, this might have contributed to the high survival rate of freshwater, seawater, and brackish water organisms could with 78% similarity of macroinvertebrate community structure. Planorbidae is dominant in these locations. Baroudi et al. (2020) found that Planorbidae can be used as environmental pollution bio-monitors. Also, Sindern et al. (2022) found that this species in estuaries, Chennai Metropolitan Area, India. For the last cluster, patchy distribution of mangroves could be identified at sampling location PE 12, PE 13, PE 14, PE 15, and PE

19. Most of the macroinvertebrates are inhabited in mudflats and intertidal settling of mangroves for feeding, shelter, and reproduction purposes. Mollusks and crustacea namely were the recorded species from these locations. As they were deposit feeders, mangroves may provide ideal places for their survival. Gastropoda species, which are primarily grazers and deposit-feeders, they were recorded compared to crab species at a tropical mangrove (Stiepani et al. 2021).

The majority of aquatic organisms prefer a pH range of 6–9.5, whereas most macrobenthos prefer a pH range of 7.5–9 (Tagliapietra and Singovini 2010). The average pH value of the selected sampling locations in the estuary was within the preferred pH range for live benthic macroinvertebrates. The highest average DO concentration was recorded in sampling location PE 1 (8.41 mg/L) because of the mixing of saline and fresh water in this location. This may be attributed to the turbulence of surface water by the effect of wind, thereby causing the water to saturate. In addition, the lowest average DO concentration and highest average BOD were recorded in sampling location PE 17, which is exposed to disposals via waste canals. The highest average ortho-phosphate was recorded at the sampling location of PE 11, which might be attributed to the exposure to disposal canal from nearby fish market. The highest average ammoniacal nitrogen was 0.93 mg/L which was recorded from sampling location, PE 12. Likewise, this location is exposed to a disposal canal which may carry out sanitary waste and livestock runoff.

Salinity is a key stress factor for many species diversity including macrobenthos in coastal water. The degree of natural instability induced by marine water intrusions should be considered in indices used to evaluate the ecological potential of water bodies, such as diversity indices (Mrozinska et al. 2021). Based on the salinity types, species of macrobenthos would be vary. These salinity types are Tidal Freshwater, Oligohaline, (0.5–3 ppt), Mesohaline (3–16 ppt), Polyhaline (16–30 ppt) and euhaline (more than 30 ppt) (Gillett and Schaffner 2009). Mulik et al. (2020) found that the salinity is the most important contributing factor to the variation of macrobenthos structure in three estuarine systems, Maharashtra district, India. In this study, Salinity varies from 32.6 to 39.00 ppt at selected sampling locations, Panadura estuary. The last sampling location is around 2 Km from the sea mouth to upward direction. Also, this study was conducted only for the three-month period. Species of macrobenthos are difficult to classified into different groups based on the salinity range in present study. Although, their response could be species-specific and depending on salinity range (Taxon salinity tolerance). Similarly, variation of abundance of macrobenthos and salinity was observed by Hangzhou Bay, China (Jia et al. 2022).

CCA analysis proved that high concentrations of DO, pH, salinity, and moderate concentrations of EC and TSS were

indicated at sampling locations which were located in the lower part of the estuary. Among these parameters, high EC, Salinity, and DO at the lower part of the estuary is due to the mixing of saline water via the sea mouth. Similarly, Mosley et al. (2010) and Nascimento et al. (2021) found high salinity and pH were near the estuary mouth due to the presence of oceanic water entering through the mouth of estuaries. Similar pattern result in present study was also observed by Fatema et al. (2016) in the Merbok Estuary, Malaysia and Rozirwan et al. (2021) in the Musi Estuary, South Sumatra, Indonesia. Also, there was a significant positive correlation between physical-chemical water quality parameters (salinity, pH, and DO) and the diversity of gastropoda in Coastal Waters of Ambon Island, Indonesia (Rumahlatu and Leiwakabessy 2017). Families Littorinidae, Muricidae, and Planorbidae were mostly influenced by these water quality parameters. Many studies indicated Planorbidae as a freshwater fauna (Afshan et al. 2013 Jayachandran et al. 2022). However, this family also found in coastal areas due to the environmental disturbance (Palasio et al. 2018). As mentioned in previously, Planorbidae was dominant family in present study. In addition, many authors found Littorinidae in most coastal areas (Rosenfeld et al. 2022).

Although, high values of water temperature, turbidity, and high concentration of nitrate-nitrogen and moderate concentration of BOD, ammoniacal-nitrogen, nitrite-nitrogen, and ortho-phosphate were reported at sampling locations which were situated upper part of the estuary. This would be due to the discharge of wastewater from many anthropogenic activities, variation of physical transport process and variation of river flow. Similarly, Goncalves et al. (2015) stated that the nutrients are generally noticed in higher concentrations in the inner channels of the estuary. Cerithidae, Coenobitidae, Haminoeidae, Nerithiidae, Thiaridae, Donacidae, Mytilidae, and Veneridae were influenced by these water quality parameters and many of them are bioindicators of the low water quality. Similar finding recorded by many authors in tropical countries as well as all over the word (Liu et al. 2011; Hamza 2019; Oliveira et al. 2016; Tlili 2018).

When Shannon- Wiener diversity index (H') is in the range of 2–3, the location is considered as moderately polluted (Trivedi 1981). Based on the present results, the surveyed Panadura estuary can be considered moderately polluted. Similar finding has been recorded by many sampling locations in the Negombo Lagoon, Sri Lanka by Dahanayaka and Wijeyaratne in 2006. On the other hand, the Pollution Tolerance Index (PTI) is based on the concept of indicator organisms and their tolerance levels (Mitchell and Stapp 1997). PTI Index was 20 or less in all the sampling locations (Table 2), which indicated that the water quality was poor and paved the way to decrease the health quality of the estuary. Similar observation has been recorded based on the study conducted in Bolgoda Canal by (Idroos and Manage 2012). Also, Olomukoro and Dirusi (2014) has been used PTI of

macrobenthos to classify water quality in Edion and Omodo Rivers in Derived Savannah Wetlands in Southern Nigeria and they found fairly and poor water quality status. Furthermore, the PTI confirmed the results of Shannon-Wiener diversity index. Both indices indicated that the biological condition and health status of the Panadura estuary are poor.

A negative moderate relationship between Shannon - Wiener diversity index and BOD as well as Nitrate-N was found during the study period. This finding corroborates the result in the Negombo lagoon, where salinity was positively correlated with species diversity (Chandrasekera and Hettiarachchi 2011). However, there was a negligible relationship with Shannon-Wiener diversity index because no significant variation of salinity was recorded in the estuary during the study period. Based on the study conducted in Lagos Lagoon, Nigeria related to the diversity macrobenthos and some water quality by Bassey et al. (2020) found that there was no relationship between diversity of macrobenthos and pH, TDS, conductivity, DO, alkalinity, salinity, water temperature, nitrate, sulphate, and turbidity. Also, Lu et al. (2021) found a moderate relationship between diversity of macrobenthos and pH in Jiaozhou Bay in China. Hence, the relationship between macrobenthos diversity and water quality parameters depend on the condition of the lagoon or estuary (sampling site).

Some of the most marked temporal fluctuations in species abundances are linked to seasons (Shimadzu et al. 2013). Mortality in sedentary bivalvia may occur during the wet season in places where rain water collects, as well as during dry periods when evaporation may cause porewater to become hypersaline (Beasley et al. 2005). The present study results revealed that except one family, all the other macrobenthos families showed no significant difference between sampling months. This would be due to the limited time period conducted in present study. Also, high molluscs abundance recorded in April (dry period). The ability of molluscs to reproduce during dry period may be a reason for higher diversity of molluscs in that season. The recruitment and density of macrobenthos may be impacted by the seasonal reproductive cycles of some species Beasley et al. (2005). Therefore, it is needed to deep understanding about the species and their reproductive cycle. Similarly, most of the researchers reported less abundance of benthos during wet season was due to monsoon rain and huge water flow from northern state of India. The seasonality of the environmental conditions described the major variations of the macrobenthos in subtropical estuary, Bangladesh (Matin et al. 2018).

Conclusion

The BOD and nitrate were identified as the major factor influencing the diversity of benthic fauna in the studied estuary. Families Littorinidae, Muricidae, and Planorbidae

were positively related to a high concentration of DO, pH, salinity, and moderate concentration of EC and TSS presented at lower part of the estuary. Also, families Cerithidae, Coenobitidae, Haminoeidae, Nerithiidae, Thiaridae, Donacidae, Mytilidae, and Veneridae were affected by a high value of water temperature, turbidity and high concentration of nitrate-nitrogen and moderate concentration of BOD, ammoniacal-nitrogen, nitrite-nitrogen, and orthophosphate presented at upper part of the estuary. Hence, marobenthos assemblage was affected by the convergence of anthropogenic and natural stressors.

The PTI of 19 selected locations indicated the presence of pollutant tolerance and moderately pollution-tolerant species in the Panadura estuary. There were significant differences in species richness, species diversity, and physio-chemical parameters with sampling locations in the Panadura estuary. Shannon Weiner's diversity index in each location in the Panadura estuary showed that the water is moderately polluted. There was a significant difference in physio-chemical parameters; pH, DO, BOD, TSS, orthophosphate, and ammoniacal-N between the sampling locations. Except family Veneridae, all the other macrobenthos families showed in significant difference between sampling months. The further studies should be included the determine the impact of seasonal variability and diverse macrofauna including polychaetes with considering the throughout the year.

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Declarations

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