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Influence of Environmental Parameters on General health status, Condition Index of Brackish water clam *Meretrix casta* occur in selected estuaries in Sri Lanka

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ABSTRACT

Many economically important bivalve species such as oysters, mussels, scallops, clams and cockles are recorded in the Coastal waters of Sri Lanka. Condition index (CI), is an inexpensive tool for assessing their general health, growth differences and for monitoring environmental pollution. It is a quantitative measure of the relative physical condition of the molluscs. The objective of the present study was to evaluate the CI of *Meretrix casta*, a brackish water clam commonly found in estuarine habitats of Sri Lanka. For this study we selected four sites Namely Mundal Lake, Marawala in Chilaw lagoon, Pamunugama and Pitipana in Negombo lagoon. The effect (If any) of general water quality parameters on the CI of these clams was also assessed. The clams were sampled from each site (n=30) monthly during the period of September 2007 to May 2008 (nine months) and site specific temperature, pH, conductivity, salinity and dissolved oxygen levels in water were measured in each sampling period. The condition index was determined as a percentage of soft tissue weight to whole weight of the animal. The results showed that CI of the clams collected from Mundal Lake and Marawala was significantly lower (ANOVA, Fisher's test, $P<0.05$) than that of clams collected from the sampling sites of Negombo lagoon despite significant temporal variations in CI of clams in each site during the study period. The CI of clams collected from Mundal Lake was significantly correlated with the conductivity ($r = 0.759$, $P < 0.05$) and salinity ($r = 0.759$, $P < 0.05$) of water. However no significant correlations were found between the CI of the clams collected from each site and other water quality parameters. Shell erosion especially in the umbo region was prominent in the clams collected from Mundal Lake (> 90%) and Marawala (> 96%) Chillaw lagoon in all sampling periods compared to the clams collected from Pitipana and Pamunugama in Negombo lagoon. The

shell erosion may have affected the general condition of the clams residing in Mundal Lake and Marawala sites.

Keywords: Coastal Waters, Condition Index ,General Health Status, *Meretrix Casta*, water quality parameters.

INTRODUCTION

The Phylum Mollusca includes animals belonging to Class Bivalvia which contains over 7000 contemporary species (Pechenik, 2000). Bivalves include clams, scallops, mussels and oysters. Most of the species are found in marine and estuarine environments and some are also found in fresh water bodies. Bivalve resources are reported to be abundant in the coastal waters of Sri Lanka. Sri Lanka has a coastline of approximately 1620 km including the shoreline of bays and outlets excluding lagoons (Atlas of Sri Lanka, 1997). The country is very rich in economically important bivalves such as oysters (*Crassostrea madrasensis*, *Saccostrea cucullata*) mussels (*Perna viridis*, *P perna*), clams (*Marcia opima*, *M. hiantina, Meretrix casta*) cockles (*Gafrarium tumidum*, *Anadara granosa*) and pearl oysters (*Pinctada vulgaris*, *P. margaritifera*) around the coastal areas (Sadacharan, 1982). Fishermen harvest bivalves from natural wild habitats in Negombo, Chilaw, Kalpitiya, Mannar, Jaffna, Trincomalee and Southern coastal belt of the country (Fernando, 1977 ;). Nevertheless, bivalve culture is still at the experimental stage in the country (Wanninayake *et al.*, 1990). The condition index (CI) is a rapid measure of ecophysicological state in commercially exploited bivalve and other mollusc species (Lucas and Beninger, 1985; Yildiz and Lök, 2005). CI has been used to evaluate how these bivalve organisms are affected by their environment (Van Dolah *et al.*, Rheault and Rice 1996). The condition index is an inexpensive, representative and responsive tool for monitoring pollution (Scott & Lawrence 1982) and has also been used to estimate growth differences among molluscs living in environment subject to different salinities and temperature regimes. Since the metabolic energy remaining after reproduction and daily maintenance is converted to biomass, a bivalve stressed either by its water quality or by disease has less energy for growth consequently, a comparison of bivalve condition index among the bivalves should be indicative of bivalve health and influence of environmental and contaminant stress (Rebelo *et al.*, 2005). Condition index vary according to the physiological activity of the organisms (e.g. spawning results in a decrease in the condition index) and the degree of environmental stress (e.g. lack of food results in a decrease in the condition index). CI index has been most important in the use of bivalves as indicators of environmental pollution (Palmer et al.1993)

Other conditions that could cause differences in molluscs condition index are the presence of parasites or commensal organisms living in association with molluscs. Parasites and commensals are known to reduce the input of vital resources to a host, thereby reducing its growth and reproductive potential (Mercado-Sliva, 2005). Biological factors as well as environmental factors could affect the health of bivalves. Fouling organisms such as barnacles, limpets, sponges, polycheatworms and bivalve larvae, etc are normal colonists of mollusc shell surfaces and do not normally present a threat to the health of the molluscs. But they can affect health directly by impeding shell opening and closing or indirectly through competition for food resources from that circumstance can weaken the mollusc (Warner, 1977). Water pollution also causes significant impact on health of bivalve molluscs inhabiting the polluted environments. Since molluscs are essentially sessile species, this renders them particularly susceptible to pollutants. Environmental conditions have a significant effect on mollusc health both directly (within the ranges of physiological tolerances) and indirectly enhancing susceptibility to infections. This is especially important for species grown under conditions which bear little resemblance to the wild situation. Water temperature, salinity, turbidity, fouling and plankton blooms are all important natural factors for molluscan health. Extremes and or rapid fluctuations in any one of these can compromise mollusc health (Bondad-Reantaso *et al*., 2001).

Shell surface observations and inner shell observations give valuable information of the condition of the bivalves. The presence of fouling organisms (barnacles, sponges, polycheate worms etc.) on the inner shell surface is a clear indication of a weak and sick mollusc. The inner surfaces are usually kept clean through mantle and gill action. Perforation of the inner surface can be sealed off by deposition of additional conchiolin and nacre. When perforation or other irritants exceed repair, the health of the mollusc is jeopardized and it becomes susceptible to opportunistic infections (Bondad-Reantaso *et al*., 2001).

Meretrix casta is an abundant clam species which is small and living in brackish water environment growing to a maximum size of about 5cm. Meat of *Meretrix casta* is considered more delicious than that of the "great clam" (*Meretrix meretrix*). The species has a high degree of tolerance to salinity variation and prefers muddy sandy bottom. Wherever the species occurs it forms extensive beds. All clams burrow in to the sandy or muddy substratum and the fishers feel their presence in the submerged areas by probing the bottom with their fingers of toes (Jones, 1970).

Most of the studies carried out so far on bivalves in Sri Lanka mainly have focused on their culturing methods (Wanninayake *et al*., 1990, Indrasena and Wanninayake. 1994, Wanninayake and Kumara. 1990) on their population dynamics (Kithsiri *et al*., 2004), and spatial variation (Dahanayaka and Wijayaratne. 2006), Up to date no studies have been done on the health condition of the bivalves inhabiting the natural beds in Sri Lanka. The aim of the present study was to assess the influence of environmental factors on the general health status of the brackish water clam *Meretrix casta* inhabiting selected estuarine environments in Sri

Lanka. using gross clinical observations and Condition Index. *Meretrix casta* can be considered as a culturable bivalve species for human use. It is important to understand how environmental parameters and biological factors such as fouling agents and parasites affect the health status of these clams.

METHODOLOGY

Four sites (A, B, C and D) located in different estuarine environments in Sri Lanka were selected in the present study (Figure 1). These sampling sites contained wild populations of the clam, *Meretrix casta*. Sampling site A is located at Mundal Lake in Mundalama (between longitude 79^0 49' 29.85"E and latitude 7^0 47' 19.51"N approximately). Sampling site B is located in Marawala in Chillaw estuary (between longitude 79^0 48'14.56"E and latitude 7^0 33' 11.49"N). These two sites are located in the North Western Province. The other two sampling sites (site C and site D) are located in Negombo estuary in the Western Province. Sampling site C is located in Pitipana area (between longitude 79^0 49'37.77"E and latitude 7^0 11' 45.71"N) and it is in the northern part of the Negombo estuary. Fourth sampling site (site D) is located in the southern part of the Negombo estuary (between 79^0 50' 50.56"E and 7^0 06'36.01"N), in the Pamunugama area. Samples of clams (*Meretrix casta*) were obtained monthly from the study sites for nine months period starting from September 2007 to May 2008.

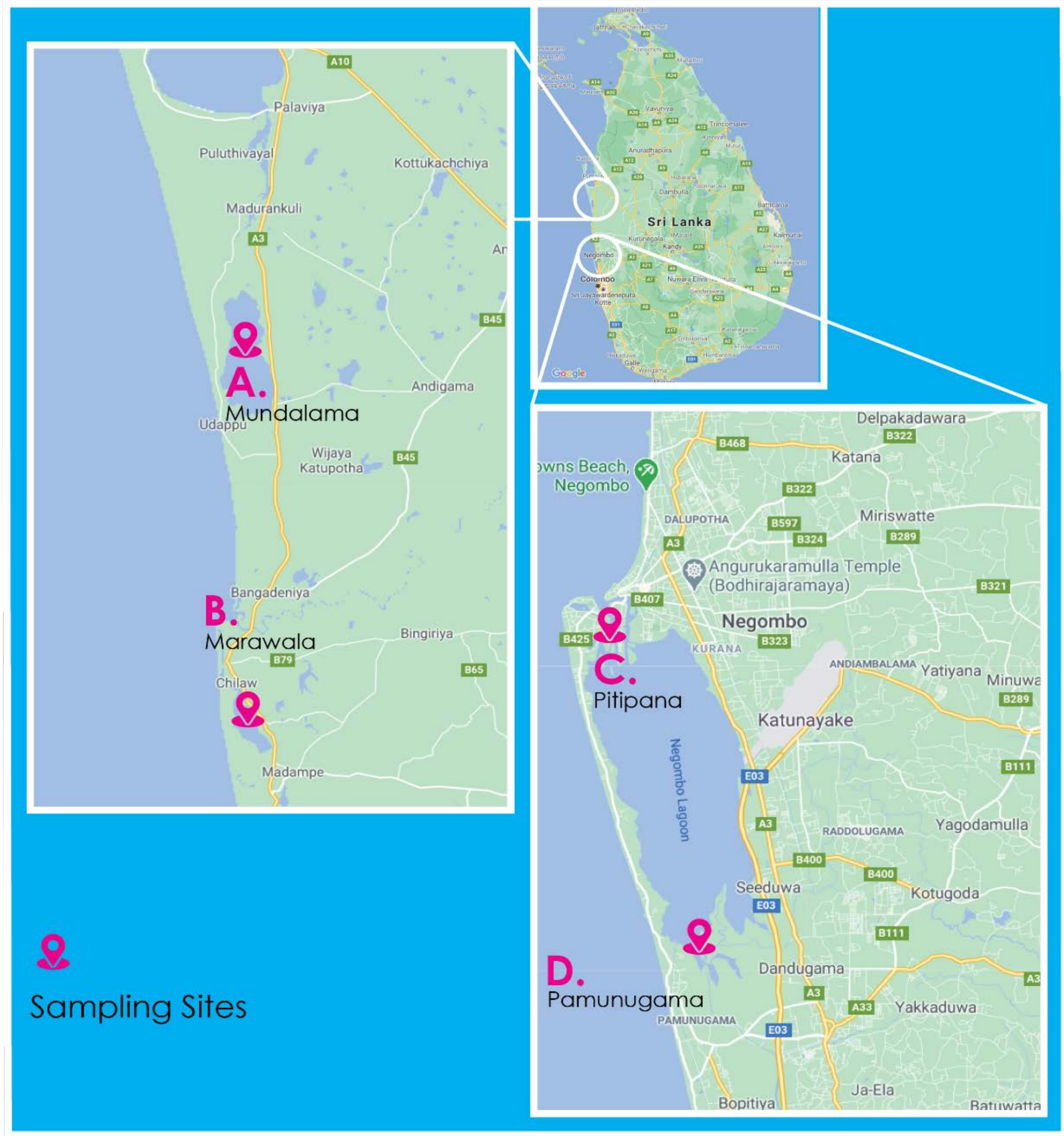


Figure 1- Location of the sampling sites A. Mundalama (Mundal Lake) B. Marawala (Chilaw estuary) C. Pitipana and D. Pamunugama (Negombo estuary)

Collection of samples

The four sampling sites were visited monthly for nine months from September 2007 to May 2008 and on each sampling day, physico-chemical parameters in the water were measured in situ in the sampling sites. Water pH, salinity, dissolved oxygen level, conductivity and temperature were recorded using water quality monitors (pH measured by pH meter-model PH315i, salinity measured by –Refractometer model-ATAGO S-28, dissolved oxygen measured by DO meter model-OXI315i, conductivity measured by conductivity meter-model-COND340i and temperature measured by mercury thermometer).

Daily and monthly rainfall data for all four sites during the nine months period were collected from the Meteorological department, Colombo, Sri Lanka.

The brackish water clams (*Meretrix casta*) were collected from the four selected sampling sites on each visit. Thirty individuals of clams from each site were collected and transported to the laboratory in oxygenated polyethylene bags for further investigations. In the laboratory, clams were kept separately in plastic basins with the water collected from the same sampling sites, (not more than 24 hours) until further studies. The water in the basins was aerated continuously.

All the clams collected from each of the four sampling sites on each visit were observed for the presence of external fouling organisms on the shells, shell erosion and shell boring as described by Bondad-Reantaso., *et al* (2004). Then the shell length, shell width and whole weight of each clam were measured and their shells were opened and gross appearance of soft tissues and abnormalities (if any) in the internal side of the shell were recorded. Standard length and the width of the shell was measured using Vernier caliper before tissue removal. Then the weight of the soft tissue of each individual was recorded. Whole weight and soft tissue weight of clams were measured using an electrical balance.

Data Analysis

Condition index was determined for each clam collected from all four sites during nine months period using following formula (Davenport and Chen, 1987).

Condition index = soft tissue weight / whole weight * 100

Temporal variations and/or spatial variations in the environmental parameters, condition index of the clams were analyzed by Analysis of Variance (ANOVA) followed by Fisher's test. Correlations between condition index of the clams and environmental parameters were tested by Pearson correlation test. The data were transformed to log(x+1) to reduce the variance and normalize their distribution prior to analyze them with ANOVA. and correlation test.

Results

Physico-chemical characteristics in water and rainfall pattern

Temporal variations in general water quality characteristics in the study sites and rainfall pattern in the areas are presented in Figure 2. Physico-chemical characteristics in water collected from four sites and rainfall pattern in the areas during the study period (pooled data) are presented in Table 1 as ranges and mean ± SD values.

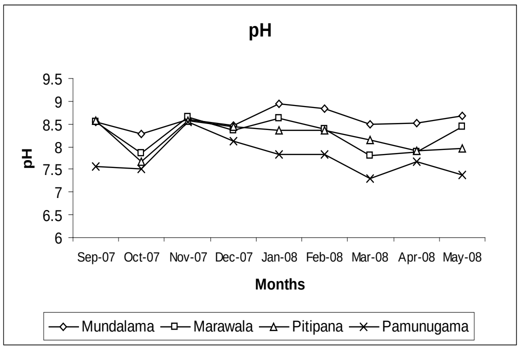
Temperature and pH levels in water samples collected from the four sites during the nine months period ranged from 29 to 35^0C and 7.3-8.93 respectively. In Mundalama site, water pH values during nine months period were comparatively high: (pH 8.29- pH 8.93). Lowest pH values in water were found in the Pamunugama site (Figure 2). Overall pH of water during nine months period was significantly higher in the sites located at Mundalama, Marawala and Pitipana compared to the site at Pamunugama (Table 1). No significant differences were found among the four sites with respect to the overall mean of the pooled data on water temperature during the study period.

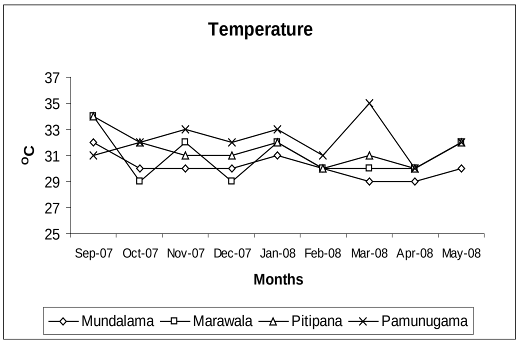
Water salinity was recorded as zero at Marawala and Pamunugama sites in December 2007 and April 2008 (Figure 2). Salinity level in water in Mundalama site varied from 10 to 34 g/l during the nine months period. Water salinity was significantly higher at Mundalama site compared to the values at Marawala and Pamunugama (Table 1).

Water conductivity at the sampling sites varied widely during the study period specially at Marawala, Pitipana and Pamunugama sites (Figure 2). Water conductivity was very low at the Pamunuama site during the period October 2007 to December 2007. Overall mean conductivity values were significantly higher at Mundalama and Pitipana sites compared to those at the other two sites (Table 1).

Highest dissolved oxygen level in water was reported in December 2007 in all four sites, whereas lowest dissolved oxygen level was reported in April 2008 in all sites except in Pitipana (Figure 2). However, no site-specific significant differences in dissolved oxygen levels were found (Table 1).

Pamunugama and Pitipana sites (Negombo estuary) in March 2008 had the highest rainfall; rainfall was lower in all four sites in January and February 2008 (Figure 2). Overall monthly rainfall was higher in Negombo estuary (Pitipana and Pamunugama) than that in the other two sites (Table 1).





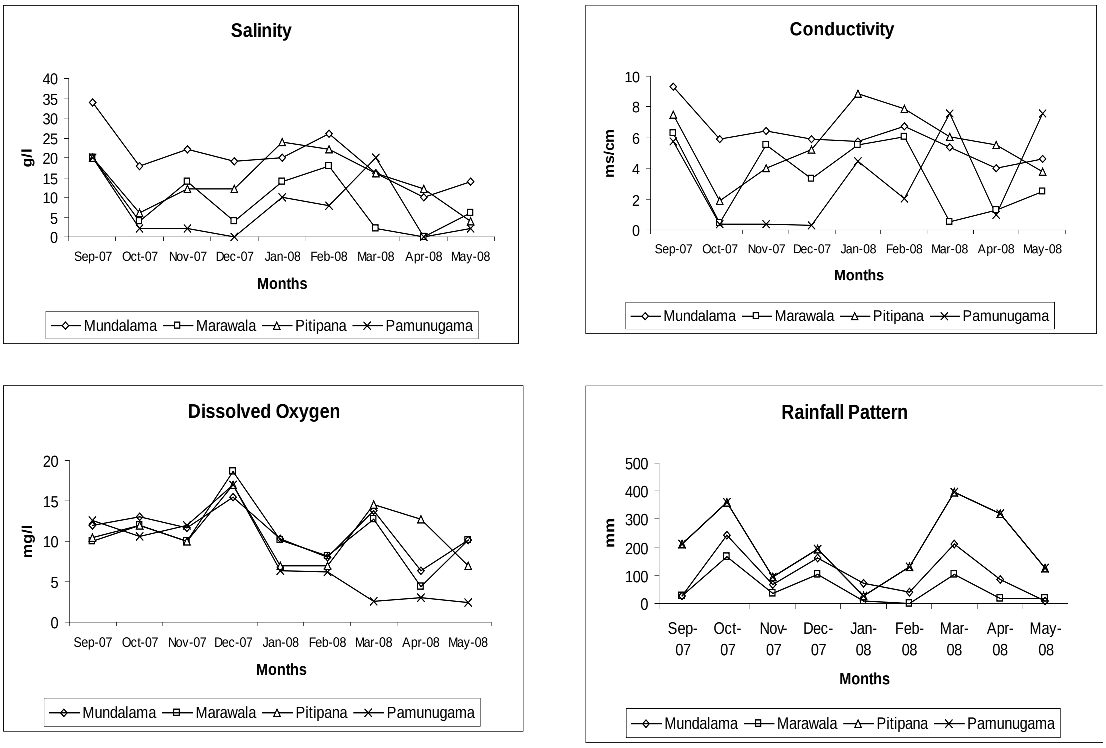


Figure 2- Temporal variations in physico-chemical characteristics in water collected from four study sites and rainfall pattern in the area

Table 1 Physico-chemical parameters in water collected from four study sites and rainfall pattern in the area during the study period *

Parameter	Mundalama	Marawala	Pitipana	Pamunugama
pH	8.6 ±0.19^{b} (8.29-8.93)	8.28 ± 0.34^{b} (7.81-8.64)	8.22 ± 0.32^{b} (7.66-8.58)	7.75 ± 0.39 ^{a} (7.30-8.54)
Temperature(^{0}C)	30.11 ± 0.93^{a} (29-32)	30.89 ±1.69^{a} (29-34)	31.44 ±1.24 ^{a} (30-34)	32.11 ±1.45 ^{a} (30-35)
Salinity (g/l)	19.89±7.01^{a} (10-34)	9.11±7.42^{b} (0-20)	14.22±6.88^{ab} (4-24)	7.11±8.07^{b} (0-20)
Conductivity(ms/cm)	6.02±1.51^{a} (4.01-9.35)	3.51±2.41^{b} (0.47-6.32)	5.65±2.21^{a} (1.93-8.86)	3.27±3.09^{b} (0.31-7.58)
Dissolved Oxygen(mg/l)	11.20±2.83^{a} (8-15.5)	10.71±3.83^{a} (4.4-18.7)	10.85±3.57^{a} (6.9-14.5)	8.07±5.20^{a} (2.4-10.6)
Rainfall(mm)	101.9±83.4^{a} (9.3-241.3)	53.8±56.8^{a} (1.1-165.0)	206.1±128.1^{b} (25.5-397.8)	206.1±128.1^{b} (25.5-397.8)

* Pooled nine months data (September 2007-May 2008) are presented as mean ± SD and ranges. For each parameter the mean values indicated with different superscripts are significantly different from each other (ANOVA, Fisher's test, $P<0.05$). Data were transformed to log(x+1) before analysis.

Shell size and appearance of the shell and soft tissues of the clams (*Meretrix casta*) collected from four study sites

Shell sizes of the clams collected from the four sites during the study period are given in Table 2. Shell length of the clams collected from four sites followed the order: Pitipana ≈ Pamunugama > Marawala > Mundalama and the width of the clams collected from four sites followed the order Pamunugama > Pitipana ≈ Marawala > Mundalama. Lowest shell size was recorded in the clams collected from the site at Mundalama during the sampling period. The shells of the clams collected from Pitipana and Pamunugama sites were relatively larger than shells of the clams in the other two sites.

Algae were observed on the external surface of the shells of the clams in all four sites during the sampling period (Figure 3A). Algae attachments on the external side of the shell of the clams were higher at Mundalama, Marawala and Pamunugama sites than at the Pitipana site (except during the period October 2007 – January 2008). Barnacles were present only on the shell surface of the clams collected from the Pamunugama area during the sampling period (Figure 4). Those barnacles had attached to the siphoning area of the clam interfearing its siphoning movement (Figure 3B). Fouling organisms were not present in the inner side of the shells of clams collected from all four sites during the nine months period.

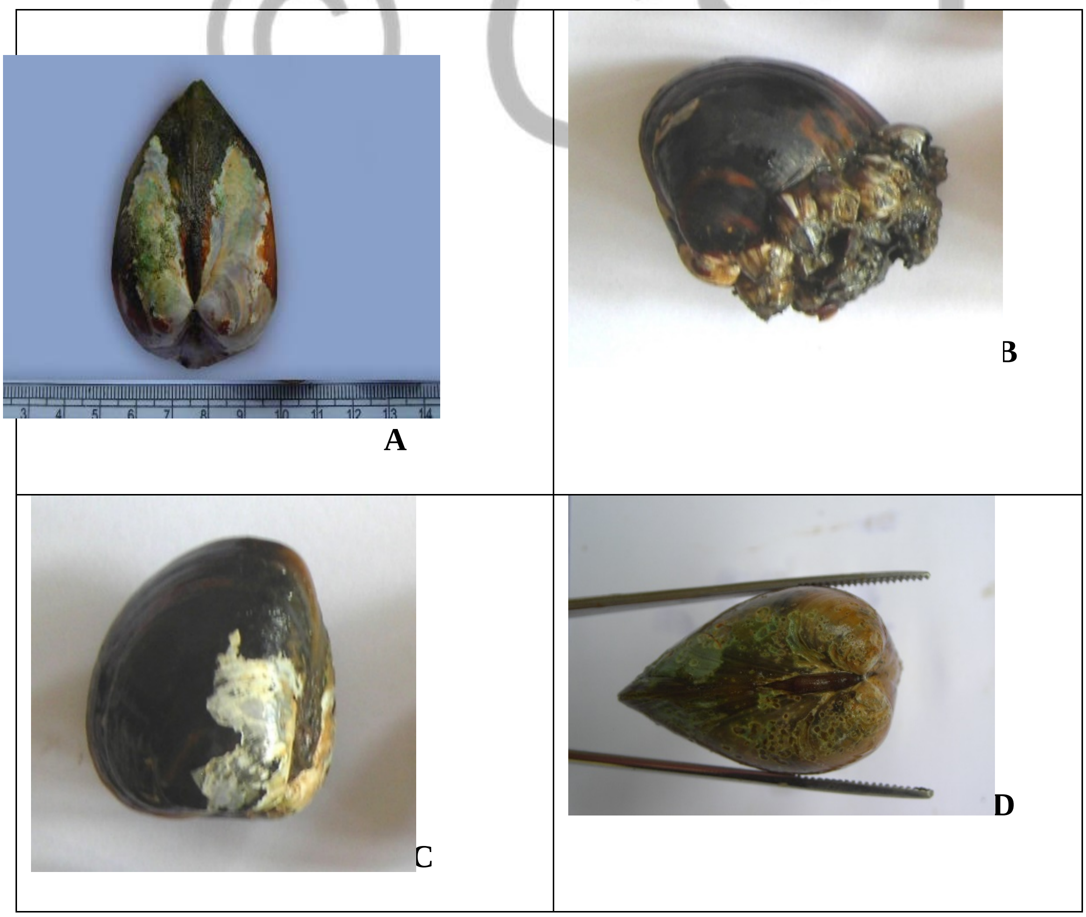
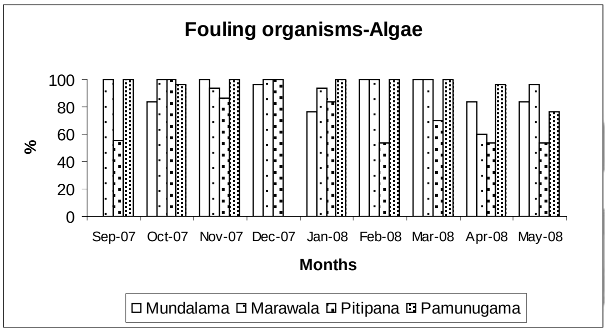


Figure 3: A -Algae on Clam ,B-*Meretrix casta* with attached barnacles on the shell surface, C-*Meretrix casta* with eroded shells, D- *Meretrix casta* with shell pores, E-*Meretrix casta* with abnormal internal shell colouration, F-*Meretrix casta* with normal internal shell colouration but with watery soft tissues



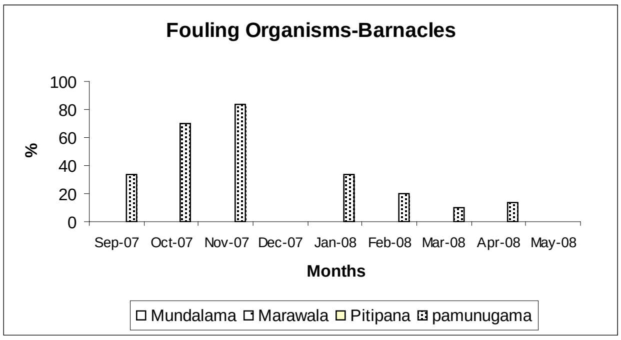


Figure 4: Percentage of clams with attached algae and barnacles on the shells

(In September 2007 at Mundalama site and in December 2007 at Pamunugama site, the clams were not sampled)

Shell erosion (Figure 5) was prominent in the clams collected from Mundalama site in Mundal Lake (>90%), Marawala site (>96%) in Chillaw estuary and Pamunugama (>90%) site in the Negombo estuary. In all sampling periods, lowest shell erosion was observed in the clams collected from Pitipana site in Negombo estuary (<60%) in almost all months. Erosion of shell occurred mainly in the umbo area of the clams (Figure 3C) collected from most sites. In some clams, both valves of the clam shell were eroded, and in some clams the erosion of shell occurred in only one valve of the two valves. Shell erosion was severe in some of the clams collected especially from Pamunugama site. In these clams the erosion area was at least 1/3 of the shell area in each valve.

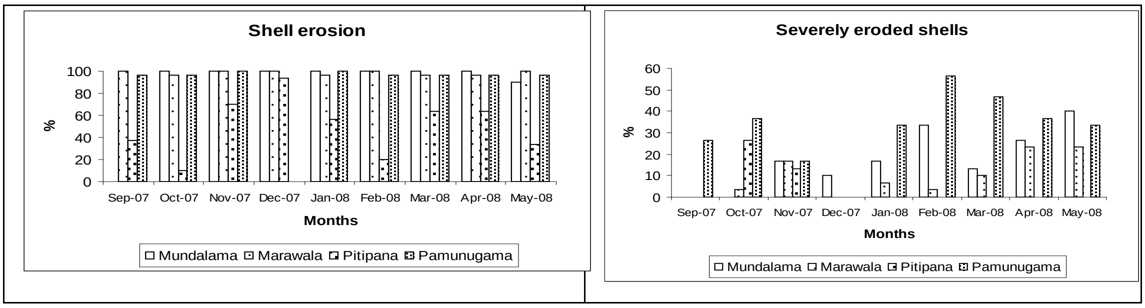


Figure 5: Percentage of clams with eroded shells collected from four sites, during nine months period. In September 2007 at Mundalama and in December 2007 at Pamunugama, clams were not sampled.

Pores in the shells of clams were also present in the shells of the clams collected from the four sites (Figure 3D). At the site of Marawala during the study period pores in the shell of clams were reported in each month of the study period (Figure 6). In the site of Pitipana, shells with pores were found only in few clams compared to the clams in the other three sites. In some months (September to November 2007 and February 2008, April and May 2008) pores in the shell of clams were not found in the Pitipana site (Figure 6).

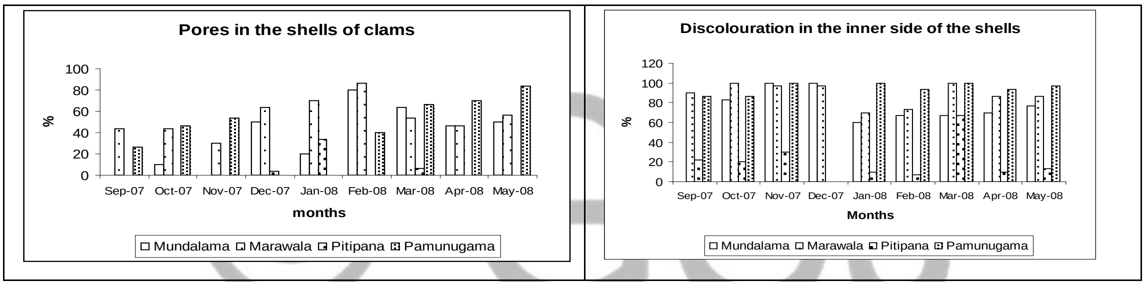


Figure 6: Percentage of clams with pores in the shell and percentage of clams with abnormal colour pattern in the inner side of the shell. In September 2007 at Mundalama and in December 2007 at Pamunugama clams were not sampled.

Abnormal colour pattern in the inner side of the shell (Figure 3E), were observed in all four sites during the sampling period. In some shells the dark coloration was present as rings and in some shells it was present as a big spot. Shells with abnormal colour pattern were higher in the clams at Pamunugama and Marawala sites in comparison to the clams collected from the site at Pitipana (Figure 6).

Abscess lesions, water blisters and gill deformities were not observed in the clams collected from the four sites during the sampling period. However, most clams had watery soft tissues (Figure 3F). Percentage occurrence of clams with watery tissues is given in the Figure 7. Percentage of clams with watery tissues was comparatively lower at Pamunugama site during the sampling period, except in the months of March 2008 and May 2008.

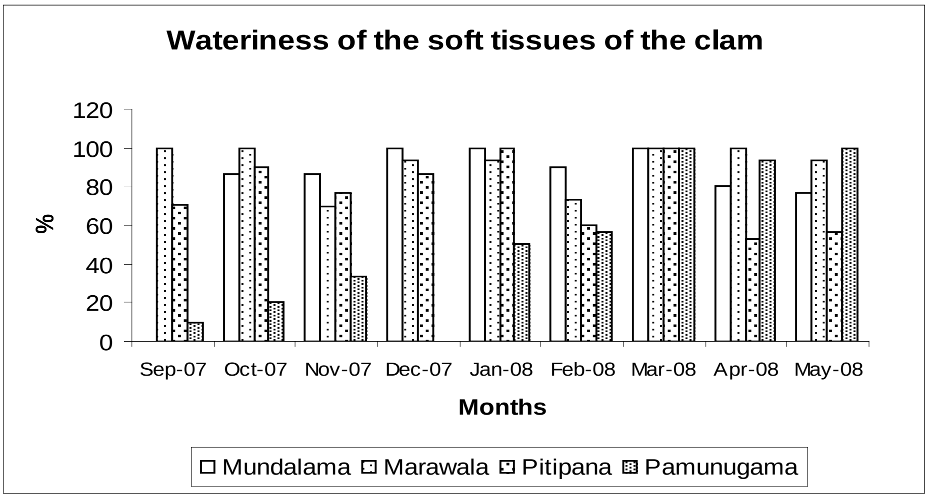


Figure 7: Wateriness of the clam tissues during the study period

In September 2007 at Mundalama and in December 2007 at Pamunugama clams were not sampled.

Table 2: Shell size of *Meretrix casta* collected from four sites during the study period

Sampling period	Mundalama		Marawala		Pitipana		Pamunugama	
	Length(cm)	width(cm)	Length (cm)	width(cm)	Length(cm)	width(cm)	Length(cm)	width(cm)
Sep 2007	----		3.17±0.15^{a}	1.99±0.15^{a}	4±0.97^{b}	2.3 ±0.57^{b}	3.5±0.39^{c}	2.46±0.19^{b}
Oct 2007	1.99 ± 0.16^{a}	1.17±0.40^{a}	2.94±0.24^{b}	1.71±0.19^{b}	3.86±0.51^{c}	2.32±0.42^{c}	3.57±0.31^{d}	2.48 ±0.19^{c}
Nov 2007	2.03±0.13^{a}	1.15±0.11^{a}	3.17±0.17^{b}	1.87±0.18^{b}	3.36±0.74^{c}	1.95±0.44^{b}	3.78±0.28^{d}	2.71±0.27^{c}
Dec 2007	2.1±0.14^{a}	1.2±0.11^{a}	3.38±0.18^{b}	2.1±0.14^{b}	3.53±0.64$^{b\ c}$	1.99±0.40^{b}	----	
Jan 2008	2.1±0.15^{a}	1.24±0.10^{a}	3.41±0.22^{b}	2.06±0.13^{b}	3.77±0.79^{c}	2.14±0.46^{b}	3.68±0.23$^{b\ c}$	2.51±0.15^{c}
Feb 2008	2.36±0.34^{a}	1.5±0.23^{a}	3.78±0.29^{b}	2.4±0.22^{b}	3.26±0.49^{c}	1.84±0.34^{c}	3.61±0.39^{b}	2.5±0.25^{b}
Mar 2008	2.09±0.10^{a}	1.24±0.08^{a}	3.33±0.24^{b}	2.07±0.15^{b}	3.56±0.72^{c}	2.07±0.53^{b}	3.54±0.31^{c}	2.45±0.23^{c}
Apr 2008	2.01±0.13^{a}	1.15±0.10^{a}	3.52±0.37^{b}	2.22±0.29^{b}	3.68±0.62^{b}	2.11±0.34^{b}	3.66±0.21^{b}	2.52±0.20^{c}
May 2008	2.12±0.12^{a}	1.25±0.08^{a}	3.45±0.18^{b}	2.08±0.17^{b}	3.92±0.50^{c}	2.37±0.32^{c}	3.59±0.23^{d}	2.48±0.19^{c}
Overall	2.1 ± 0.12^{a}	1.24±0.11^{a}	3.35 ± 0.24^{b}	2.06±0.20^{b}	3.66 ± 0.25^{c}	2.12±0.18^{b}	3.62 ± 0.09^{c}	2.51±0.08^{c}

In September 2007 at Mundalama and in December 2007 at Pamunugama, clams were not sampled. The data are presented as mean ± SD, n=30.

In each row, with respect to one parameter (length or width) data represented by different superscripts are significantly different from each other (ANOVA, Fisher's test, $P<0.05$).

Condition index of the clams collected from the four study sites: effects of environmental factors

The condition index of *Meretrix casta* collected from the four sites are presented in Table 3 for the nine months period. Temporal changes in the Condition Index of the clam collected from the four sites were significantly different. The condition index of the clams collected in April 2008 from site A (Mundalama) was significantly lower than that of the clams collected from the same site during other periods. The CI of the clams collected in April 2008 from the site B (Marawala) was also low but not significantly different from the values recorded in November 2007, January – March 2008 and May 2008. At the site C (Pitipana), the lowest CI values were recorded in September 2007, March-April 2008. The lowest CI was recorded in the clams collected from site D (Pamunugama) in March 2008.

Overall means of the pooled CI of the clams collected from the four sites are given in Figure 8. During the study period, overall CI of the clams (based on pooled data) was the lowest in the samples collected from Mundal Lake, whereas highest CI was reported in clams collected from Negombo estuary: Pamunugama and Pitipana.

Correlations between condition index of the clams and the environmental parameters are given in Table 4. The condition index of the clams residing in Mundal Lake was positively correlated with conductivity and salinity of water. CI of the clams residing in Pamunugama estuary was positively correlated with Dissolved Oxygen and pH of water. However, no other significant correlations were found between the CI of the clams collected from each site and the water quality parameters and rainfall.

Table 3: Temporal variations in Condition index (CI) of *Meretrix casta* collected from four study sites

Sampling Period	A-Mundalama	B-Marawala	C-Pitipana	D-Pamunugama
SEP 2007	-----	13.70 ± 11.77^{a}	11.45 ± 6.00^{a}	13.40 ± 1.02^{cd}
OCT 2007	7.82 ± 1.76^{b}	12.35 ± 2.14^{a}	14.02 ± 2.15b^{c}	13.47 ± 1.94^{c}
NOV 2007	9.06 ± 1.63^{cd}	8.57 ± 1.19^{b}	15.42 ± 2.02^{c}	14.53 ± 2.62^{cd}
DEC 2007	8.42 ± 1.66^{bd}	10.95 ± 1.39^{ac}	13.97 ± 2.27^{b}	-----
JAN 2008	7.46 ± 1.28^{b}	10.36 ± 1.49^{bc}	13.69 ± 2.37^{b}	13.93 ± 0.59^{c}

FEB 2008	9.62 ± 2.12[c]	9.88 ± 1.52[bc]	15.85 ± 7.96[c]	13.12 ± 1.67[c]
MAR 2008	8.89 ± 1.31[c]	10.47 ± 1.27[bc]	11.39 ± 1.31[a]	**10.43 ± 1.38[a]**
APR 2008	**6.40 ± 1.60[a]**	**7.98 ± 1.23[b]**	**11.03 ± 1.51[a]**	11.15 ± 1.66[b]
MAY 2008	8.18 ±1.19[bd]	9.67 ± 1.45[b c]	14.29 ± 2.59[bc]	11.84 ± 1.95[b]

Results are presented as Mean ± SD values. Data indicate with different superscript letters in each column are significantly different from each other. (ANOVA, Fisher's test , P<0.05). In September 2007 at Mundalama and in December 2007 at Pamunugama clams were not sampled.

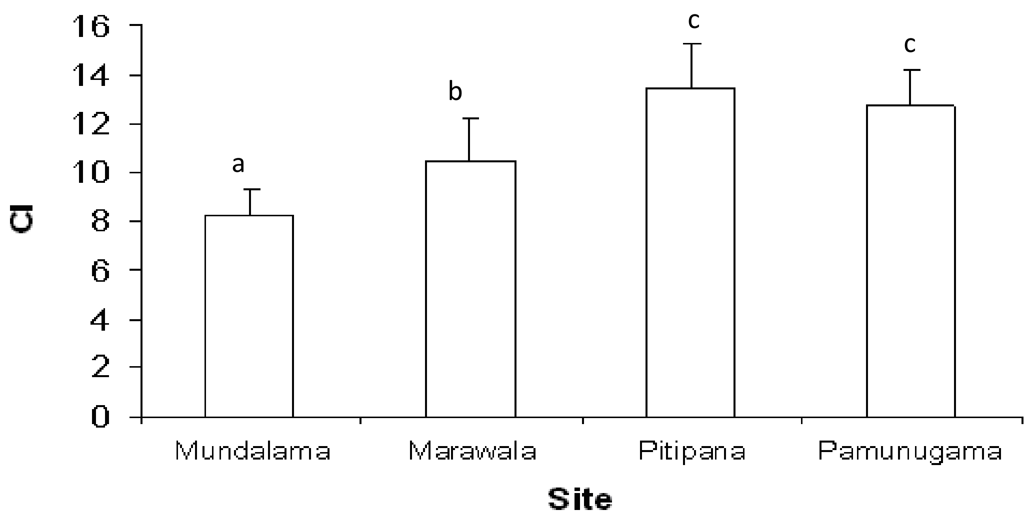


Figure 8: Overall condition index (CI) of *M.casta* collected from four sites during study period. Results are presented as mean ± SD. Bars indicated with different letters are significantly different from each other. (ANOVA, Fisher's test, P<0.05).

Table 4: Correlations (r) between Condition Index of the clams collected from four sites and environmental characteristics in the area during the study period.

parameter	Mundalama Site A	Marawala Site B	Pitipana Site C	Pamunugama Site D
conductivity	**0.759***	0.118	-0.167	-0.479
salinity	**0.762***	0.316	-0.079	-0.176
DO	0.335	0.425	-0.453	**0.813***
Temperature	0.088	0.217	-0.250	-0.116
pH	0.187	0.044	0.179	**0.701***
Rainfall	-0.050	0.414	-0.614	-0.642

Pb in water	-0.121	0.162	-0.142	0.161
Pb in sediment	-0.237	-0.255	0.143	0.128
Cu in water	0.077	-0.141	0.346	0.336
Cu in sediment	0.424	-0.155	0.421	-0.146
Cd in water	-0.285	-0.486	0.042	-0.253
Cd in sediment	0.148	-0.062	0.323	-0.057

*Significant at P< 0.05 (Pearson correlation test), n=9

Discussion

Environmental characteristics in the selected study sites

Exposure to environmental stressors can result in biochemical, physiological and histological (tissue) alterations in living organisms. Presence of these alterations may serve as signaling exposure to stressors or adverse effects; in the aquatic environment such stressors can constitute changes in physical parameters such as temperature, pH, salinity, conductivity, dissolved oxygen and rainfall, as well as toxic concentrations of chemical pollutants or any combination of these (Werner *et al* .,2003).

In this study temporal variations in general water quality parameters in selected sampling sites in three estuaries namely Mundalama in Mundal Lake, Marawala in Chilaw estuary, Pitipana and Pamunugama in Negombo estuary and rainfall pattern in the area was measured for nine months period starting from September 2007 to May 2008. In each site pH, water temperature in water did not vary much in the study period, but water pH level was comparatively low at the Pamunugama site. The salinity level, conductivity level and dissolved oxygen level varied widely during the study period. In the Mundalama site and Pitipana, salinity was higher than the other sites. These two sites Mundalama and Pitipana are closer to the sea area than the other two sites; Marawala and Pamunugama. This could be the reason for higher salinity level and conductivity level in these areas. Lowest level of salinity and conductivity were reported in the freshwater end of the Negombo estuary, Pamunugama. Marawala site also had lower salinity and conductivity levels due it location further to the sea.

Overall highest rainfall pattern was reported in the Negombo estuary area and Lower rainfall was observed in the Marawala site which is located at Chilaw estuary. Jayasiri and Rajapaksha (2000) showed that the salinity of Mundal lake depends on the flushing through the Dutch canal and on local evaporation, precipitation and runoff during periods of limited exchange of water, the Mundal Lake is likely to become strongly hypersaline. In this study also, the highest level of salinity was recorded in the site of Mundalama.

Shell size, (length and width) of the clams varied in the four sites. Smallest size of shell length and width was recorded in the clams collected from Mundalama site whereas largest shell length was reported in the clams collected from Pitipana site and the largest shell width was reported in the clams collected from Pamunugama site in Negombo estuary. It is not known whether age of the clams reside at the four sites are similar, as this study focused on wild populations of the clams. It is not known whether the clam populations at Mundalama site were established recently because their size is smaller than the other sites.

Fouling organisms such as barnacles, limpets, sponges, polychete worms and bivalve larvae, etc. are normal colonists of mollusc shell surfaces and do not normally present a threat to the health of the molluscs. According to Bondad - Reantaso *et al*., 2001, they can affect health directly by impeding shell opening and closing or indirectly through competition for food resources from that circumstance can weaken the mollusc. In the present study algae were observed in external shells of the clam collected from four sites during the study period. Algae attachments on the external side of the shell of the clams were higher at Mundalama, Marawala and Pamunugama sites, whereas lower algae attachment was found in the Pitipana site in Negombo estuary. Barnacles were present on the shell surface of clams collected from Pamunugama site in Negombo estuary. They attached to the clams covering their siphoning area. In a recent study on growth rate of cultured oysters *Crassostrea madresensis* showed bivalves which covered with fouling organisms had a lower growth rate than the bivalves which had cleaned shells. Weight gain of the cleaned bivalves was higher than the ones that were covered with barnacles (Piyathilaka *et al*, 2008).

Shell damage by boring organisms such as sponges and polychaete worms is normal in open water growing conditions. These are usually benign, but under certain conditions (especially in older molluscs) may reach proportions which make the shell brittle or pierce through to the underlying soft-tissues. This degree of shell damage can weaken the mollusc and render it susceptible to pathogen infections. (Bondad - Reantaso *et al*., 2001). In the present study pores in the shells were high in Marawala and Pamunugama sites. It indicate, that shell boring organisms are present in the study sites, but in the Pitipana site in Negombo estuary, pores in the shells of clams were less compared to other three study sites. The results indicate that the shell boring organisms are less in the Pitipana area compared with the other three sites.

Shell erosion of the clams was prominent in the Mundalama site in Mundal Lake, Marawala site in Chilaw estuary and Pamunugama site in the Negombo estuary. Lowest shell erosion was reported in the Pitipana site in Negombo estuary. Shell erosion was mainly occurred in the umbo region of the clams.

Shell erosion was severe in some of the clams collected from Pamunugama site and in Pamunugama site relatively low pH in water in the study period may be one reason for higher shell erosion observed in the clams collected from that site.

In the present study abnormal internal shell colour was observed in the clams collected from four study sites. But the severely affected shells were observed in the Pamunugama site and Mundalama site. But in the Negombo estuary at Pitipana site, occurrence of this abnormal internal shell colour among the clams was less. That dark colouration was due to the deposition of chonchiolin layer. Chonchiolin is secreted by bivalves when they are under stress conditions. Chonchiolin deposition in clams also occurs when they are affected by the brown ring disease caused by vibriosis (Paillard, 2004). But it has been reported that the brown ring disease is absent in areas with high temperature. In this study, temperature in all four study sites was higher than the 21^0C. It indicates that this abnormal colour and deposition of chonchiolin layer occurred due to some other reason. Wateriness in clam tissues was also observed in the four study sites. In the month of March 2008, all sites had 100% wateriness in the clam tissues.

The condition index (the ratio of soft body weight to shell weight) is a quantitative measure of the relative physical condition of the molluscs. Since the metabolic energy remaining after reproduction and daily maintenance is converted to biomass, a bivalve stressed either by its water quality or by disease has less energy for growth consequently a comparison of bivalve condition index among the bivalves should be indicative of bivalve health and influence environmental and contaminant stress. Condition indices can be affected by multiple abiotic factors and physiological activities, among them spawning is one of the most important. While seasonally regulated in temperate climates, this is apparently not so in the tropics (Rebelo *et al*., 2005).

Condition Index (CI) of clams collected from four study sites during the study period of September 2007 to May 2008 was significant. Highest CI was reported in the clams collected from Pitipana site and Pamunugama site in Negombo estuary where as lowest CI was reported in the clams from Mundal Lake. It is not known whether the low Condition index of the clam collected from Mundal Lake is due to age differences or due to environmental factors.

Influence of environmental parameters on the health status of the clams residing in the study sites

Environmental conditions have a significant effect on mollusc health both directly (within the ranges of physiological tolerances) and indirectly enhancing susceptibility to infections. This is especially important for species grown under conditions which bear little resemblance to the wild situation. Water temperature, salinity, turbidity, fouling and plankton blooms are all important natural factors for molluscan health. Extremes and or rapid fluctuations in any one of these can compromise mollusc health (Bondad-Reantaso *et al*., 2001). Climate change is also recognized as an important factor of the change in distribution of pathogens and diseases. Change in environmental conditions is believed to be responsible for the expansion of some diseases such as the diseases due to *Perkinsus marinus*. Sea level rise could drastically alter the coastal environment such as salinity and as a result, may favor local diseases or emergence of new pathogens (Berthe, 2008).

Correlations between condition Index and the environmental parameters showed that conductivity and salinity in Mundalama site in Mundal Lake was significantly correlated with the Condition Index. In Pamunugama site dissolved oxygen and pH of water correlated with the condition index. These results could be related to the larger environmental variations occurred in these sites. Lower Condition index values observed in clams collected from Mundalama could be a result of high salinity due to low freshwater supply and high evaporation (Arulananthan *et al*., 1995) in that water body or may be due to environmental pollution in that area. Because Mundal Lake located in area where shrimp farms practiced and their effluents discharged in to that water without treatments and this water body has very limited mixing (Corea *et al*., 1998).

Lowest CI was reported in the clams from Mundalama site in Mundal Lake but the clams collected from the other three sites namely Mundalama, Marawala and Pitipana April 2008 had the lowest CI values. Clams from Pamunugama site had lowest CI value in the month of March 2008 and this sudden drop of CI was recovered in short period of time. So this sudden drop of CI values may be due to the spawning of the clams in that period. These CI fluctuations can govern by seasonal factors including gamete production, food availability and temperature or by toxins (Rebelo *et al*., 2005).

Conclusion

In conclusion the present study revealed the clams in selected study sites are safe for human consumption and their CI is affected by the environmental parameters. However, there should be a constant monitoring programme on the estuarine environments because there is a tendency to change the General health due to increasing urbanization, agricultural and industrial development of the area. Considering the health status of the clams although several abnormalities were observed in relation to the appearance of the shells and soft body. However, site–specific changes were found in relation to CI, which measures the overall health; of the clams. Further studies on health aspects of bivalve resources in Sri Lanka are needed for sustainable exploitation of these resources as food and nonfood sources.

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