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Original Article

# Influence of environmental variables on the abundance of estuarine clam *Meretrix casta* (Chemnitz, 1782) in Trang Province, Southern Thailand

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# Abstract

The estuarine clam *Meretrix casta* (Chemnitz, 1782) is an edible bivalve that burrows into bottom sediment to protect itself from environmental fluctuations and predation. This study examined the relationship between clam populations and abiotic environmental conditions in order to develop an understanding of the biological and ecological processes that could be used to improve their management and exploitation. The study site was located in the estuary of the Palian River, Kantang District, Trang Province, on the Andaman coast of Thailand. Six zones were established along the estuary. Data were collected from January 2008 to December 2008. The results showed that salinity (Sal) and concentrations of total suspended solids (TSS) were the water quality parameters most closely related to population densities of estuarine clams. The combination of those variables gave the strongest correlation to clam densities (Spearman rank correlation,  $p_w = 0.504$ ). Among bottom sediment parameters, pH and ferrous and ferric iron concentrations gave the strongest correlations to clam densities (Spearman rank correlations to clam densities (Spearman rank correlations to clam densities (Spearman rank correlations to clam densities (PH, ferrous and ferric iron) were the main significant parameters influencing the abundance of estuarine clam *M. casta*.

Keywords: Estuarine clam, Meretrix casta, bivalve, population, environment

# 1. Introduction

One aim of marine biologists is to understand ecological processes by examining the interrelationship between abiotic parameters and biotic structures. Estuarine ecosytems are ideal for examining such interactions due to their wide range of abiotic parameters. This is usually accomplished by inferential analysis of empirical data by multivariate techniques, and by conducting manipulative experiments (Brown *et al.*, 2000; Edgar and Barrett, 2002). Work on the influence of environmental parameters on population densities of bivalves has been performed elsewhere (Franz, 1976; Boonruang and Janekarn, 1983; Absalão, 1991; Baron and Clavier, 1992; Soares-Gomes and Pires-Vanin, 2005; McLeod and Wing, 2008). However, there is great variation among geographical areas. Estuarine clam *M. casta* fishing in the coastal area of Trang Province, Thailand, has been conducted by local villagers for more than a decade. A population of the clam *M. casta* from the estuarine system in this area has been studied (Songrak *et al.*, 2009) but no work has been done on its relationship to water and sediment quality. The purpose of this study was to gather quantitative information on *M. casta* from that area in an attempt to relate community structure to water and sediment parameters. Knowledge of the environmental parameters affecting the population structure of that bivalve is required for sound management of their exploitation.

# 2. Materials and Methods

#### 2.1 Description of study site

\*Corresponding author. Email address: stanyaros@gmail.com The study was conducted in the estuary of the Palian River, located on the coast of the Andaman Sea between



Figure 1. Map of the study site in the Palian River Estuary, Kantang District, Trang Province, Thailand.

 $7^{\circ}15' - 7^{\circ}45'$  N and  $99^{\circ}15' - 99^{\circ}45'$  E, Kantang District, Trang Province, southern Thailand (Figure 1). The climate of the area is influenced by seasonal displacements of the southwest monsoon and the northeast monsoon. The dry season extends from December to April, and the rainy season from May to November. The estuary receives major freshwater discharges during the rainy season. The tidal range is about 3 m during spring tides, and 1 m during neap tides. *M. casta* is naturally distributed along the estuary and has been fished by local fishermen for a century.

# 2.2 Clam sampling

The study was conducted at six zones along the estuary (Figure 1). Each zone was comprised of three stations. Data were collected at monthly intervals from January to December 2008. Sampling occurred on the same day at every sampling station. The stations were reached by boat and positions were checked using GPS. A clam drag was used to collect clam samples. The drag was made from stainless steel with an opening, 35 cm in width and 15 cm in height. The net (mesh size  $1.8 \times 1.8$  cm) was connected to the tail of the drag. Clams were collected at each station by drawing the drag by hand for a distance of 15 m. The number of clams were then counted and clam density calculated per square meter.

#### 2.3 Water sampling and analytical methods

Dissolved oxygen (DO) concentrations, pH, and water temperature were measured *in situ* using a YSI DO meter (model 85) and pH meter (Hanna model HI 991002), respectively. Water salinity was measured with a hand refractometer. Three replicate samples of water were collected from 20 cm below the water surface in plastic bottles. Samples were kept on ice and analyzed soon after returning to the laboratory. Total suspended solids (TSS) were analyzed by the GF/C filtration method (APHA, 1980). Samples were filtered through pre-washed GF/C filters before determining ammonia (NH<sub>3</sub>), nitrite (NO<sub>2</sub><sup>-</sup>), nitrate (NO<sub>3</sub><sup>-</sup>), and filterable reactive phosphate (PO<sub>4</sub><sup>3-</sup>) concentrations. NH<sub>3</sub> of water samples was analyzed by the indophenol blue method (Koroleff, 1983). NO<sub>2</sub><sup>-</sup> was analyzed by the diazotization method (Strickland and Persons, 1972). NO<sub>3</sub><sup>-</sup> was analyzed by cadmium reduction (Strickland and Persons, 1972). NO<sub>3</sub><sup>-</sup> in water samples was reduced to NO<sub>2</sub><sup>-</sup> and the original and reduced NO<sub>2</sub><sup>-</sup> were analyzed using the same diazotization method (Strickland and Persons, 1972). PO<sub>4</sub><sup>3-</sup> was analyzed by the ascorbic acid method (Strickland and Persons, 1972), and chlorophyll *a* (Chl *a*) was analyzed by the acetone extraction method (APHA, 1980).

#### 2.4 Sediment sampling and analytical methods

Three sediment cores were taken per station using an acrylic corer (5 cm in diameter, 20 cm length) at low tide. The cores were brought to the laboratory and sectioned 15 cm from the surface. Half the sediment was used to determine soil pH, organic matter (OM), and sediment respiration rate (SRR). Soil pH was taken using a pH meter (Hanna model HI 991002). OM was analyzed by the ignition method (Ben-Dor and Banin, 1989). A laboratory SRR experiment was conducted to determine CO, production rates in the soil. CO, released during aerobic respiration was analyzed by the respiratory chamber method (Boyd, 1992). The other half of the sediment slice was transferred to double centrifuging tubes and centrifuged (1,500 rpm, 10 min) to obtain pore water for determination of hydrogen sulfide ( $H_2S$ ), ferrous (Fe<sup>3+</sup>) and ferric (Fe<sup>2+</sup>) iron, and ammonia (NH<sub>2</sub>). Samples for determination of H,S were fixed with 100 µl of 100 mM Zinc acetate per 1 ml sample and stored frozen until analysis.  $Fe^{3+}$  and  $Fe^{2+}$ irons were determined by the spectrophotometric method (Stookey, 1970; Sørensen, 1982), and NH, in pore water was determined by the indophenol blue method (Koroleff, 1983). H<sub>s</sub>S was determined by the spectrophotometric method (Cline, 1969).

#### 2.5 Data analysis

The one-way analysis of similarity permutation test (ANOSIM) proposed by Clarke and Green (1988) was used to test the null hypothesis of monthly differences in estuarine clam abundance. The degree of similarity of clam density on each sampling was calculated using the Bray-Curtis similarity coefficient, based on the number of individuals. The resultant similarity matrix was subjected to cluster analysis and nonmetric multidimensional scaling (MDS). A stress value was calculated for the MDS procedure. It is a measure of how well the relationship among the samples is represented by the MDS. A value < 2.0 is considered to provide a good representation (Clarke and Warwick, 1994). Analyses were performed using the CLUSTER program in the PRIMER 5 computer package (Clarke and Warwick, 1994). Abiotic environmental data, both water quality variables (DO, pH, Salinity, water temperature, TSS, TIN (NO<sub>2</sub> + NO<sub>3</sub> + NH<sub>3</sub>), PO<sub>4</sub><sup>-3</sup>, and Chl *a*) and bottom sediment variables (pH, SRR, OM, NH<sub>3</sub> in pore water, Fe<sup>3+</sup> and Fe<sup>2+</sup> iron, and H<sub>2</sub>S), were expressed as means, and then the structure of clam abundance and environment matching analyses were performed using the BIOENV program in PRIMER 5 (Clarke and Warwick, 1994).

#### 3. Results

#### 3.1 Water quality parameters

**DO**: The mean of DO concentrations among all stations varied from 4.81 to 7.31 mg L<sup>-1</sup>. The highest value was found in December, and the least in April (Figure 2a). **pH**: The variation of pH values is shown in Figure 2b. Mean pH values ranged from 6.14 to 7.46. **Salinity**: High salinity was found in the dry season from January to April and low salinity in the rainy season from May to December (Figure 2c). The maximum value was recorded in surface water on March (23.94



Figure 2. Mean ( $\pm$ SD) values of dissolved oxygen (a), pH (b), salinity (c), water temperature (d), chlorophyll *a* (e), total inorganic nitrogen (f), orthophosphate (g), and total suspended solids (h) in each month at the sampling sites over the year 2008.

ppt), and the minimum value in September (5.27 ppt). Water temperature: Water temperatures did not vary greatly, and followed a typical seasonal pattern. Mean water temperature values over the study period varied from 27.58-31.18°C (Figure 2d). Chl a: Mean chl a concentrations over the study period varied between 2.16 to 8.35 mg  $L^{-1}$  (Figure 2e). The highest chl a value was found in March during the dry season, and the least in November during the rainy season. **TIN**: The variation in TIN ( $NO_2^{-} + NO_3^{-}$  and  $NH_3$ ) concentrations is represented in Figure 2f. The maximum value (0.70 mg L<sup>-1</sup>) was recorded in July during the rainy season, and the minimum mean value (0.23 mg L<sup>-1</sup>) in January during the dry season.  $PO_4^{3-}$ : The variation in  $PO_4^{3-}$  is represented in Figure 2g. The mean of concentrations among all stations varied from 0.009 to 0.013 mg L<sup>-1</sup>. TSS: The variation in TSS concentrations is shown in Figure 2h. Mean TSS values ranged from 11.87 mg  $L^{-1}$  in January to 49.11 mg  $L^{-1}$ , in April.

# 3.2 Bottom sediment quality parameters

**pH**: The overall pH of bottom sediment during the dry season was higher than during the rainy season (Figure 3a).

The maximum mean pH value was recorded in February (7.18) and the minimum mean pH value in December (6.42). **SRR**: The highest mean SRR value was found in January (38.19 mg cm<sup>-2</sup> h<sup>-1</sup>) and the lowest in July (12.73 mg cm<sup>-2</sup> h<sup>-1</sup>) (Figure 3b). **OM**: The mean OM content of bottom sediment among all stations varied from 1.13% to 2.35% (Figure 3c). **NH**<sub>3</sub> **in pore water**: Lowest ammonia-N concentrations in sediment pore water were found during June to September (0.46-0.61 mg L<sup>-1</sup>), and were highest in November (6.96 mg L<sup>-1</sup>) (Figure 3d). **Ferrous and ferric iron (Fe):** Mean concentrations of ferrous and ferric irons in bottom sediments varied over the study period (Figure 3e) from 29.00 mg L<sup>-1</sup> in June to 55.73 mg L<sup>-1</sup> in February. **H**<sub>2</sub>**S**: Mean concentrations of hydrogen sulfide did not show much variation (Figure 3f). Mean values over the study period varied from 14.04-16.96 µg L<sup>-1</sup>.

#### 3.3 The abundance of estuarine clams

Populations of the estuarine clam *M. casta* varied in a clear seasonal pattern (Figure 4). Compositions of clam densities were significantly different among months (Global R = 0.18, p < 0.007). Higher densities were found during the



Figure 3. Mean (±SD) values of pH (a), soil respiration rate (b), organic matter (c), ammonia-N in pore water (d), ferrous and ferric irons (e), and hydrogen sulfide (f) in each month at the sampling sites over the year 2008.



Figure 4. Mean (±SE) densities of estuarine clams from monthly sampling.



Figure 5. Dendrogram of cluster analysis based on estuarine clam densities from January to December 2008.

dry season than during the rainy season. Peak densities occurred in March (32 ind.  $m^{-2}$ ), and were lowest in October (8 ind.  $m^{-2}$ ). A cluster analysis based on abundances of estuarine clams showed that the monthly sample densities could

be placed into three major periods with a similarity level of 78.01% (Figure 5). The first period showed moderate densities in January and February. The second period showed high densities between March and May. The last period showed low densities between June and December

# 3.4 Relationship between environmental factors and estuarine clam populations

# 3.4.1 Water quality

From linking population of the estuarine clam to water quality variables, the results found that salinity (Sal) and total suspended solids (TSS) were produced the strongest correlation with estuarine clam densities. The combination of those two variables were gave the optimal Spearman rank correlation value,  $p_w = 0.504$  (Table 1).

# 3.4.2 Bottom sediment quality

The bottom sediment variables that produced the strongest correlation with estuarine clam densities were pH and the concentration of ferrous and ferric iron (Fe). The overall optimum value for Spearman rank correlation value,  $p_{\rm w}$  of 0.716, was obtained from both variables in combination (Table 1).

# 4. Discussion

#### 4.1 General environmental conditions

The overall water quality in the study area was assessed as good. The least DO concentration was 4.81 mg/l. That value is higher than the criteria (<4 mg/l) recommended

Table 1. Correlations between estuarine clam population densities and combinations of water quality and bottom sediment quality variables, taken k at a time, to find the best matches of biotic and abiotic similarity matrices for each k, as measured by weighted Spearman rank correlation (ρ<sub>w</sub>). Bold type indicates overall optimum. (Sal.: Salinity; Chl a: Chlorophyll a; TSS: Total Suspended Solids; DO: Dissolved Oxygen; Fe: Ferrous and Ferric irons; H<sub>2</sub>S: Hydrogen Sulfide; OM: Organic Matter).

k	Best variable combination Spearman rank correlation ( $\rho_{\!_{W}}\!)$	
	Water quality variables	Bottom sediment variables
1	Sal.	pH
	(0.322)	(0.401)
2	Sal., TSS	pH, Fe
	(0.504)	(0.716)
3	Sal., TSS, Chl. a	pH, Fe, H <sub>2</sub> S
	(0.498)	(0.642)
4	Sal., TSS, Chl. a, DO	pH, Fe, H,S, OM
	(0.480)	(0.560)

by the Pollution Control Department (1994) for the conservation of natural habitats, such as mangrove, nursery areas, and the reproduction and nutrition zones of marine organisms. Mean pH values during the dry season (December-April) were higher than during the rainy season (May-November) because of the low pH of freshwater run-off. The mean pH values in this survey (6.14-7.46) were lower than the range of normal seawater, which is in the range 7.5 to 8.4 (Zeebe and Wolf-Gladrow, 2001). Mean salinity was higher in the dry season than in the rainy season as rainfall had a majority effect on salinity. Water temperatures varied seasonally. Concentrations of chl a fluctuated over the study period and were not correlated with nutrient concentrations. The results of this study do not clearly indicate that changes in the trophic state can occur simply as a result of changes in TIN and  $PO_4^{3-}$  content. To date, those variables have never been taken into account in this kind of study. In this survey, elevated TIN levels were found at the end of the dry season (April) and into the rainy season, suggesting perhaps that the TIN sources to the estuary may have come from both river inputs and sewage from the nearby resident area. However, the concentrations of TIN  $(NO_2" + NO_3", + NH_3)$  are in an acceptable range recommended by the Pollution Control Department (1994). Low  $PO_4^{3-}$  concentrations were found over the study period. This suggests that some  $PO_4^{3-}$  may be taken up by marine phytoplankton. Phosphate uptake by natural phytoplankton in coastal regions has been reported (Aubriot et al., 2004). TSS concentrations fluctuated over the study period. A major contributor of TSS to estuarine water in this area is river discharge. As is well-known, high amounts of solids in estuarine waters are potentially one of the most important environmental problems confronting benthic communities. However, TSS concentrations in this study were lower than environmental criteria recommended by the Pollution Control Department (1994).

Mean pH values in bottom sediments showed a similar trend to pH values in overlying water during the study. Sediment respiration results in CO<sub>2</sub> evolution from the oxidation of organic matter by micro-organisms (Jingyun et al., 1998), but the SRR in this study was shown to be non-correlated to the organic matter content of bottom sediment. Increasing temperature during dry season may be a key factor increasing rates of sediment respiration in this study. The estuarine clam habitat was found to contain a high percentage of sand. The OM content in bottom sediment was low when compared with the OM content in the soils of mangroves (Lovelock, 2008) and seagrass beds (Tanyaros, 2009). Lower NH, concentrations were observed in pore water during the rainy season than during the dry season. Tidal processes strongly influence NH, concentrations in the pore water of intertidal sediments. During low tides, air exposure increases the rate of degradation of organic matter, causing the NH, concentration in the sediment to increase between tidal inundations (Rocha, 1998). At high tide, NH<sub>2</sub> is often released from the inundated sediment into the water

column by diffusion and convection (Rocha, 1998; Cabrita *et al.*, 1999). Increasing and decreasing of Fe<sup>3+</sup> and Fe<sup>2+</sup> iron concentrations over the study period were closely related to organic content because of the reduction of iron by electron accepting processes coupled to the oxidation of organic matter. Postma (1994) stated that up to 50% of organic carbon oxidation in marine sediments may proceed by the reduction of Fe<sup>3+</sup> iron. In coastal sediments, the dominant process of, hydrogen sulfide production is by the reduction of sulfate by anaerobic bacterial respiration. However, 90% of all sulfide produced may be lost by re-oxidation at the sediment surface (Jørgensen, 1977). That process may have resulted in the low variation in hydrogen sulfide concentrations found in the sediment in this study.

# 4.2 Effect of water quality on estuarine clam density

In this study, the water quality parameters that appeared to have the greatest influence on the abundance and population structure of the estuarine clam M. casta were salinity and TSS. The mean values of salinity and estuarine clam abundance showed similar patterns over the study period. Changes in water salinity affect a wide variety of biochemical and physiologic processes in marine bivalves. An increase or decrease in salinity often results in an increase or decrease of free amino acid (FAA) levels in the tissues of marine bivalves, which are often monitored as a stress indicator (Powell el al., 1982; Lee et al., 2004). Variations in FFA in two marine bivalves, Macoma balthica and Mytilus spp., have been related to salinity changes by Kube et al. (2007). A study in oyster found that changing water salinity affected the sensitivity and activity of cilia and cirri on the ctenidia (Dean and Paparo, 1983). The effects of salinity changes on the feeding physiology and growth of bivalves had been reported by Sara et al. (2008). TSS concentrations were the second most important factor influencing the abundance of estuarine clams in this study. Suspension-feeding bivalves are particularly vulnerable to the effects of elevated levels of suspended solids (Shin et al., 2002). Studies on the physiological responses of bivalves to increasing suspended sediment concentrations have shown a decrease in clearance rates (Bricelj and Malouf, 1984; Ward and MacDonald, 1996; Bacon et al., 1998; Shin et al., 2002), oxygen consumption (Grant and Thorpe, 1991; Alexander et al., 1994), and growth (MacDonald et al., 1998), along with increased gill damage (Shin et al., 2002; Cheung and Shin, 2005). However, the component of TSS in the present study comprised of phytoplankton as the mean values of TSS and chl. a concentrations showed similar patterns. Positive correlation between bivalve biomass and phytoplankton abundance has been reported by Foe and Knight (1985) and Dame (1996). On the other hand, bivalves are important recyclers of nitrogen in coastal marine systems, releasing ammonium and dissolved organic nitrogen that can be taken up directly by phytoplankton (Dame, 1996).

# 4.3 Effect of bottom sediment qualities on estuarine clam density

The pH and iron concentrations (Fe) in bottom sediments were most closely related to the abundance of estuarine clams in this study. The pH values in sediment during the rainy season were lower than 6.7. When pH is lower than 6.8-6.9, calcium loss to the external environment exceeds gains (Vindograv et al., 1993). Thus, the calcium necessary for normal metabolic functions is lost, resulting in mortality. Sensitivity to decreased calcium may be due to its importance in physiology, as calcium is involved in muscular contractions, cellular cohesion, nervous functions, and the maintenance of acid-base balances (Chetail and Krampitz, 1982). Low pH may also impair ion exchange (Hunter, 1990) and affect glutamate catabolism in the mitochondria of the bivalve mantle (Moyes et al., 1985). Therefore, low pH in the sediment may have been the most important reason for the low abundance of clams during the rainy season in the present study. The concentration of Fe irons in bottom sediments was the second factor likely influencing the abundance of estuarine clams in the present study. Larger quantities of iron are capable of reducing the assimilation efficiency of Mytilus edulis and Macoma balthica (Griscom et al., 2002). However, no study has reported the effects of iron on bivalve distribution and mortality.

From the investigations carried out in this study, the results revealed that salinity (Sal) and concentrations of total suspended solids (TSS) were the water quality parameters influencing on the abundance of estuarine clams, while pH and ferrous and ferric ions (Fe) were the bottom sediment parameters gave the strongest correlations to clam densities.

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