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Horseshoe Crab, *Tachypleus gigas* (Müller, 1785) Spawning Population at Balok Beach, Kuantan, Pahang, Malaysia

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Abstract: Local and regional decline of Asian horseshoe crabs has spurred a study on its spawning population at Balok Beach, Kuantan, Pahang, Malaysia. This location was identified as spawning site due to the occurrence of horseshoe crab spawning pairs and nests. Size-frequency, length-weight relationships, sex ratio and epibiont infestation of *Tachypleus gigas* were studied. Instar stage was estimated based on prosomal width. Condition of the horseshoe crab carapace was reported. Visual search technique of horseshoe crab was conducted during high tide of new and full moons. Prosomal, opisthosomal and telson length and weight of each horseshoe crab were measured. Largest female was recorded with mean prosomal length and width of 154.4 and 246.9 mm, respectively. About 69.8% of the males belonged to size group of 151-200 mm and 53.3% of females were grouped into 201-250 mm. All individuals were of fourteenth to sixteenth instar stages. Sex ratio varied from 0.313 to 2.5 and attributed to commercial harvest and monsoon season. Sand sediment of study site showed 93% of fine sands with grain size ranged from 120 to 250 µm. Acorn and pedunculate barnacle, conical and flat slipper shells were found on the carapace of the specimens. Most males had damaged eyes and carapaces while females with broken telsons. Body damages of about 19.9% on the specimens were likely due to nearby fishing activities. Lack of satellite male indicated low spawning population. The finding of this study showed that the species is extremely threatened by human activities and coastal development.

Key words: *Tachypleus gigas*, size-frequency distribution, length-weight relationship, sex ratio, epibiont

INTRODUCTION

Three horseshoe crab species namely *Tachypleus gigas*, *Tachypleus tridentatus* and *Carcinoscorpius rotundicauda* are reported in the coastal waters of Malaysia (Kassim *et al.*, 2008; Christianus and Saad, 2009; Zaleha *et al.*, 2010; Srijaya *et al.*, 2010a). The horseshoe crab migrates from offshore continental shelf areas to spawn on estuarine beaches (Brockmann and Smith, 2009) during high tides of new and full moons throughout the year (Sekiguchi *et al.*, 1977; Chatterji *et al.*, 1992; Chatterji, 1994; Hajeb *et al.*, 2005, 2009). Earlier studies showed that *T. gigas* can be found mostly on sandy beaches with slight influence of mud (Christianus *et al.*, 2005; Zaleha *et al.*, 2011). Most researchers suggest that the Asian horseshoe crabs are declining both locally and regionally. Therefore it is important to consider some mitigation measures to avoid the extinction of this fossil species. Anthropogenic factors are mainly responsible for

declining of horseshoe crab population in the United States and Asia (Botton and Haskin, 1984; Shuster, 1982; Itow, 1993; Chatterji, 1994; Michels, 1996; Chiu and Morton, 1999, 2003a; Widener and Barlow, 1999; Botton, 2000; Chen *et al.*, 2002, 2004; Tanacredi, 2002; Burger *et al.*, 2003; Carmichael *et al.*, 2003; Zhou and Morton, 2004; Seino *et al.*, 2004; Almendral and Schoppe, 2005; Lee and Morton, 2005, 2009; Christianus, 2006; Berkson *et al.*, 2009; Cartwright-Taylor *et al.*, 2009; Hsieh and Chen, 2009; Mishra, 2009; Nishida and Koike, 2009; Zadeh *et al.*, 2009; Shin *et al.*, 2009; Yang *et al.*, 2009; Morton and Lee, 2011).

The Asian horseshoe crab does not have multiple utilization comparing to the Atlantic species but it has potential uses in biomedical and eco-tourism (Kassim *et al.*, 2008). Quantitative data on the population of Malaysian horseshoe crabs are sparse. The population structure of juvenile and adult horseshoe crabs at the spawning ground remains unknown. Moreover, the public

is unaware of the biological and ecological importance of these horseshoe crabs.

In Asia, horseshoe crab eggs are popularly served as a local delicacy (Morton and Blackmore, 2001). At Balok beach, gravid female horseshoe crabs are harvested to make into soup, curry and salad. Horseshoe crabs are also treated as by-catch and exported to Thailand where it fetches higher price (Christianus and Saad, 2009).

Commercial harvesting for food and production of *Tachypleus* Amebocyte Lysate (TAL), habitat and spawning area loss and coastal pollution are the major threats to Asian horseshoe crabs (Chen *et al.*, 2004; Cartwright-Taylor *et al.*, 2009; Hajeb *et al.*, 2009). To date, no conservation practices, legislation or harvest regulations have been implemented to protect the horseshoe crabs in Malaysia as they are not listed in the Malaysian Wildlife Conservation Act 2010 (Act, 716).

This study aims to determine the size frequency distribution, length-weight relationships and sex ratios of horseshoe crab population in Balok Beach, Kuantan, Pahang, which is a spawning site located at the east coast of Peninsular Malaysia. Carapace condition and infestation of epibionts on the horseshoe crabs were observed. This study provides both qualitative and quantitative baseline information on the spawning population of *T. gigas* in Malaysia, which is crucial for future implementation of conservation measures and management planning.

MATERIALS AND METHODS

Study site: Study was conducted between May 2009 and April 2010, at Balok beach (3°56.257'N, 103°22.568'E; Fig. 1), a sandy estuarine beach in Kuantan, Pahang, located at the east coast of Peninsular Malaysia. Spawning pairs of *T. gigas* were found in May and June, during high tides of new moon and full moon (Zaleha *et al.*, 2010; Kassim *et al.*, 2008).

Balok beach experiences north-east monsoon between November and February (Chua, 1984) and mix semi-diurnal tides. According to Department of Survey and Mapping Malaysia (JUPEM, 2010), the tidal amplitude is approximately 2.28 m during spring tide and reaches a maximum wave height of 3 m during new and full moon phases.

Water analysis and sediment characteristics of study site
Physicochemical parameters of Balok Beach coastal water: During each sampling, physicochemical parameters of coastal water including water temperature, salinity, dissolved oxygen content and pH were measured *in situ* at a depth of 10 cm using a water quality monitoring probe

(YSI, USA). Sediment grain size of Balok beach: Sand sediments from horseshoe crab nests collected in five cores (5 cm in diameter). Each core consisted of two measures, the surface sand sediment (0 to 5 cm depth) and sediment at a depth of 15 to 20 cm (Pooler *et al.*, 2003). Grain size of sand sediment then analyzed using sieve-pipette method (Gee and Bauder, 1986) and wet-sieving through mesh sizes of 16 to 240 μm . Meanwhile, silt-clay content was determined using pipette method (Hsieh *et al.*, 2004).

Sampling strategy: The horseshoe crabs were sampled for 6 consecutive days during every high tide of new and full moon phases between May 2009 and April 2010. Sampling was conducted during both day and night.

Random quadrat and transect technique was not suitable for this study site due to the small population size of *T. gigas*. Therefore, visual search technique used was based on sighting of horseshoe crab along the beach at high tide line. Searches were made for approximately 1.5 h prior to each high tide of new and full moons and covering a stretch of 300 m of the beach. Landing location of horseshoe crabs was recorded with a GPS (Global positioning system) device. The same location was re-visited during low tides to determine the presence of eggs nest.

During high tide, the horseshoe crab is partly buried in the sand or rapidly swimming in search for the right spot to nest. The horseshoe crab was gently handpicked out from the sand. Each horseshoe crab individual counted, sexed and measured for size (mm) and weight (g).

Determination of size-frequency distribution, length-weight relationship, age estimation and instar stage of horseshoe crab, sex ratio of horseshoe crab population at Balok beach:

Fresh body weight of each individual was measured to the nearest 0.01 kg using weighing apparatus. Prosomal sizes (length and width), opisthosomal length, total length and telson length of each individual were measured according to Yamasaki *et al.* (1988) to the nearest millimeter (mm) using a measurement tape.

Each horseshoe crab was classified according to prosomal width size classes. The size-frequency distribution was expressed in bar chart with different interval size classes of 0-50, 51-100 mm, 101-150 mm and so on. Prosomal width of horseshoe crab was employed as a morphometric proxy to estimate age and instar stage based on size-age-growth relationship established by Sekiguchi *et al.* (1988a). Length and weight data analyzed according to Le Cren (1951) using hypothetical formula: $W = aL^b$, where W is weight (g), L is carapace



Fig. 1: The study site at Balok Beach, Kuantan, Pahang, Malaysia

length (mm), parameter ‘a’ is proportionality constant and parameter ‘b’ is the allometric coefficient. The logarithm equation $\log W = \log a + b \log L$ calculated separately for both sexes and a straight line fitted to scatter diagram. Parameters ‘a’ and ‘b’ estimated based on logarithmically transformed expression where weight is the dependent variable and length is the independent variable (Cherif *et al.*, 2008). Sex ratio determined according to morphological characteristics following Debnath (1991) and calculated for each month throughout the study period.

Epibiont infestation on horseshoe crab: Each horseshoe crab examined individually for the presence of epibionts. Each epibiont was removed using a scalpel and fixed in 10% formalin. The collected epibiont then identified to genus level in the laboratory.

Observation of horseshoe crab carapace condition: The condition of carapace, lateral eyes and telson of each horseshoe crab individual examined and summarized. Upon completion of data collection, all horseshoe crab individuals were return to the water.

Statistical analysis: Mean value for each morphometric parameter was compared between sexes using Student’s t-tests and a significant level of 95% probability was

adopted (Chiu and Morton, 2003b). Normal distribution within each prosomal size group examined using Kolmogorov-Smirnov test. Student’s t-test was used to determine the significance at 95% probability level. Covariance analysis was employed to describe difference in the regression of logarithmic value of weight on logarithmic value of carapace length (Debnath and Choudhury, 1988; Srijaya *et al.*, 2010b). Chi-squared test was applied to determine deviation of sex ratios from 1 (Cartwright-Taylor *et al.*, 2009).

RESULTS AND DISCUSSION

Species determination, size-frequency distribution, length-weight relationship, age estimation and instar stage of horseshoe crab, sex ratio of horseshoe crab population at Balok beach: Based on morphologically characteristics following Yamasaki *et al.* (1988), the horseshoe crab species found at Balok Beach was identified as *Tachypleus gigas*. In total, 161 *T. gigas* (86 males and 75 females) were sampled throughout the study period. The total number of horseshoe crabs collected varied every month. Male individuals outnumbered the females because they tend to re-visit the spawning beach more often than the female (Lovel and Botton, 1992). Hence, males were the dominant sex at the spawning site.

Table 1: Mean and range (bracketed values) of various morphometric parameters of *Tachypleus gigas* examined at Balok Beach, Kuantan, Pahang, Malaysia

| Morphometric parameter | Male | Female | t-value |
|--------------------------|------------------|-------------------|---------|
| Body weight (g) | 332.91 (160-590) | 824.44 (260-1350) | 25.911* |
| Prosomal length (mm) | 118.67 (88-2180) | 154.4 (110-1800) | 72.687* |
| Opisthosomal length (mm) | 84.2907 (72-117) | 105.84 (75-1450) | 90.122 |
| Telson length (mm) | 175.57 (80-2260) | 218.76 (165-260) | 79.105* |
| Prosomal width (mm) | 191.20 (160-241) | 246.87 (190-320) | 74.893* |
| Carapace length (mm) | 202.97 (160-310) | 260.24 (185-305) | 82.006* |

*Significant differences between the mean values of the two sexes using Student's t-tests at 95% probability level

Most horseshoe crabs came ashore in amplexus pairs. There was occurrence of solitary males and two mating pairs in tandem where two males were coupling with one behind the other male and a female was at front of the males. The satellite male was not observed at this site. This is probably due to the small population size therefore lack of satellite male and group spawning (Botton *et al.*, 1996; Brockmann and Smith, 2009).

The largest prosomal width sampled male and female were 241 and 320 mm, respectively. Sexual dimorphism characteristic of horseshoe crab is obviously on the size difference. Females are prominently bigger than the males (Yamasaki *et al.*, 1988; Key *et al.*, 1996; Brockmann and Smith, 2009). The females were bigger in size than the males with mean prosomal length and width of 154 and 246 mm, respectively (Table 1). Matured females were heavier than the males with mean body weight of 824 g (Table 1) due to greater growth in each molt (Gerhart, 2007) and numerous egg masses in prosomal cavity (Graham *et al.*, 2009). Table 1 showed significant differences ($p < 0.05$) in prosomal size (length and width), telson length, carapace length (prosomal length) and body weight between the two sexes.

Size-frequency distribution of *Tachypleus gigas* at Balok Beach, Kuantan, Pahang: Size-frequency data indicated size groups of 151-200 mm for males and 201-250 mm for females (Fig. 2). Male *T. gigas* is smaller than the respective female. Based on prosomal widths, approximately 69% of male horseshoe crabs ranged from 151-200 mm and 53% of females were ranging from 201-250 mm. Mean values obtained from prosomal widths of both sexes showed significant differences ($p < 0.05$). The size-frequency distribution between both sexes was not normally distributed.

Length-weight relationship of horseshoe crab population at Balok Beach, Kuantan, Pahang: The parabolic equations were determined from the length-weight data:

- **Male:** $W = 0.0018 L^{2.557}$
- **Female:** $W = 0.0005 L^{2.837}$

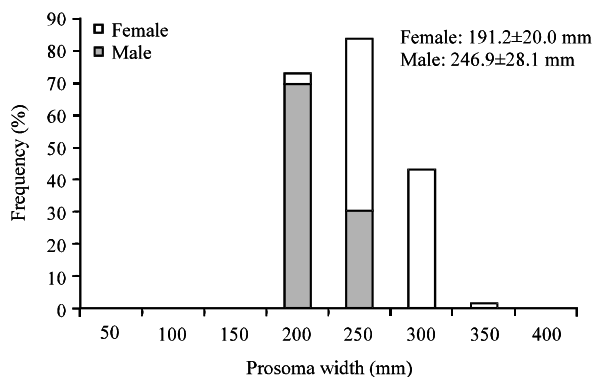


Fig. 2: Frequency of size classes of *Tachypleus gigas* at Balok Beach, Kuantan, Pahang

The length-weight relationships were linear for both sexes. The equations were shown in the following:

- **Male:** $\log W = 2.557 \log L - 2.750$
- **Female:** $\log W = 2.837 \log L - 3.303$

The values of the slopes (b) for both sexes were more than 1 thus exhibited a positive allometry where increment of weight was more rapid as compared to prosomal length. Covariance analysis revealed significant difference between the slopes of both sexes ($F = 3085.391$ with df 1,160; $p < 0.05$). Weights of males and females increased with exponentials of 2.557 and 2.837, respectively.

Length-weight relationship varies with water temperature, food availability, habitat type and reproductive activities. Fish weight and b-value increase with sufficient feeding and gonad development. The b-value is also an indicator of food intake and growth pattern (Arslan *et al.*, 2004). An animal living in inadequate water temperature and feed insufficiently would demonstrate a negative allometric growth (Arslan *et al.*, 2004).

Age estimation of horseshoe crabs at Balok Beach, Kuantan, Pahang: All horseshoe crabs examined at Balok Beach were adult individuals. Based on size-age-growth relationship established by Sekiguchi *et al.* (1988b), the

horseshoe crabs were grouped into fourteenth and fifteenth instar stages, with prosomal width sizes ranged from 154-194 mm and 194-244 mm, respectively. Individual classified into fourteenth instar stage was estimated to have survived for 10 to 11 years whereas the fifteenth instar stage group was estimated to have lived for 11 to 12 years. However, individual with prosomal width of more than 244 mm was regarded as being in the sixteenth instar stage and presumed to have lived for more than 12 years. Approximately 64% of male individuals with prosomal widths ranged between 154 and 194 mm and 55% of female individuals belonged to prosomal width size group ranging from 194 to 244 mm (Table 2).

In previous study conducted by Hajeb *et al.* (2005), a total of 54 males and 49 females *T. gigas* were sampled and measured. About 74% of male individuals categorized into prosomal widths group ranging from 154-194 mm hence grouped into fourteenth instar stage and estimated to have lived form 10 to 11 years. Meanwhile, 71% of female individuals in prosomal width size group of 194-244 mm were fifteenth instars and presumed to have lived for 11 to 12 years. There was no record of horseshoe crab individual in size group of more than 244 mm. According to Morton and Lee (2011), locality and salinity affect the increment of prosomal width of horseshoe crab species.

Sex ratio of horseshoe crab population at Balok Beach, Kuantan, Pahang: Fewer horseshoe crabs were sighted during north-east monsoon (November to March) due to

occurrence of heavy precipitation, strong surf and flooding at the study site. Strong offshore winds and low water temperature deter the horseshoe crabs from approaching the nesting beach (Gerhart, 2007). Horseshoe crab favors low wave height and swash velocity to increase spawning and hatching success of the eggs (Jackson *et al.*, 2010).

Most horseshoe crabs came ashore at Balok Beach in amplexed pairs. There were two occurrences of tandem amplexus. Neither unattached female nor satellite male was sighted. However, there was sighting of unattached males swimming nearshore. According to Zaleha *et al.* (2010), horseshoe crab spawning season varies with latitude and spawning peaks reported in May and June at the east coast of Peninsular Malaysia. However, spawning peaks observed in this study for 2009 were in May and September (Table 3). There was no large spawning aggregation at Balok Beach and spawning activity occurred year round except during monsoon season.

As compared to female, higher proportion of returning males were observed. The sex ratio (Table 3) was not significantly different from 1 except in September 2009 and March 2010 ($p < 0.05$). The null hypothesis on the sex ratio was equivalent to 1 per month of the year cannot be rejected except in July, September, December 2009 and January to April 2010 where the sex ratios were not more than 1 when males outnumbered females and thus suggesting an active breeding period. Sex ratio of 1:1 suggests a consequence of low population density (Botton *et al.*, 1996) or a maximized reproduction

Table 2: Age and instar stage estimation using prosomal widths (mm) of *Tachypleus gigas* examined at Balok Beach based on size-age-growth relationship established by Sekiguchi *et al.* (1988b)

| Prosoma width (mm) | No. of individual | | Instar stage | Age (year) |
|--------------------|-------------------|--------|--------------|------------|
| | Male | Female | | |
| 154-194 | 55 | 1 | 14 | 10-11 |
| 194-244 | 19 | 42 | 15 | 11-12 |
| >244 | 11 | 33 | 16 | >12 |

Table 3: Male to female ratios of horseshoe crab *Tachypleus gigas* sampled at Balok Beach between May 2009 to April 2010

| Year | Month | N | M:F Ratio | χ^2 | p-value |
|------|-------|----|-----------|----------|---------|
| 2009 | May | 28 | 1.333 | 0.693 | 0.405 |
| | Jun | 21 | 0.909 | 0.594 | 0.441 |
| | Jul | 21 | 0.313 | 0.007 | 0.936 |
| | Aug | 19 | 0.900 | 0.597 | 0.440 |
| | Sept | 23 | 2.286 | 0.121 | 0.729 |
| | Oct | 14 | 1.0 | 0.798 | 0.372 |
| | Nov | 7 | 1.333 | 0.843 | 0.359 |
| | Dec* | 0 | - | - | - |
| 2010 | Jan | 1 | - | 0.350 | 0.554 |
| | Feb* | 0 | - | - | - |
| | Mac | 20 | 1.857 | 0.299 | 0.585 |
| | Apr | 7 | 2.500 | 0.339 | 0.560 |

*No horseshoe crab sighting

and a full reproductive capacity of horseshoe crabs (Reyes, 2009). Fisher's principle on 1:1 (male to female ratio) is evolutionarily stable but it is not common in horseshoe crab population because male tends to re-visit the nesting beach more often than the female. Sex ratio can be influenced by the harvesting of females horseshoe crab as delicacy by the local community at Balok Beach.

Tachypleus and *Carcinoscorpius* show insignificant or no reproductive competition. Asian horseshoe crab lacks of satellite male and spawning aggregation (Brockmann and Smith, 2009) thus demonstrates monogamous mating pattern where operational sex ratio is 1:1 (Mattei *et al.*, 2010).

Infestation of epibiont on *Tachypleus gigas*: A total of 45% of male individuals were fouled. Adult male was prone to epibiotic infestation as compared to female (Table 4) due to more molt frequent of females before reaching maturity (Sekiguchi *et al.*, 1988a) and nesting behavior makes females less vulnerable to epibiont. Only sexually matured adult horseshoe crabs are infested by epibiont as they have stopped molting (Botton, 2009). However, in culture condition, this kind of epibiotic infestation was not observed on larvae and juvenile of *T. gigas* (Faizul *et al.*, 2011). Horseshoe crabs are generally burrowing shallowly in the sediment substrates when resting, foraging or spawning (Sekiguchi and Shuster, 2009). In amplexus position, the prosoma carapace of female is buried at the level of lateral eyes into the sand during spawning and opisthosoma carapace is covered by the male holding onto the female. Because spawning horseshoe crabs remain amplexed for a considerable time therefore male body is vulnerable to epibiont colonization (Patil and Anil, 2000).

Throughout the study period, four epibiont group were found on dorsal and ventral carapaces of

Tachypleus gigas namely acorn barnacle *Balanus*, pedunculate barnacle *Octolasmis*, conical slipper shell *Calyptraea* and flat slipper shell *Crepidula*. Barnacle, polychaete and mollusk are common epibionts infesting horseshoe crabs (Botton, 2009; Tan *et al.*, 2011).

Observation of horseshoe crab carapace condition: The condition of carapaces, lateral eyes and telsons of all horseshoe crab samples were examined and classified accordingly (Table 5). Approximately 8% of males had covered eyes and damaged carapaces and 15% of females with broken telsons (Table 5). Meanwhile, damaged carapace and broken telson was 19%, this is possibly due to collision with boats since the study site is near to boat landing jetty.

At the study site, males with encrusted prosoma and eyes were found attached to females as amplexus pairs. This is in contrast with the report by Brockmann and Penn (1992) that an attached male is less likely to have its prosoma and eye encrusted by fouling organisms. Duffy *et al.* (2006) also speculated that a male horseshoe crab with deteriorated lateral eyes is not fully excluded in mating. The male is either an attached or satellite individual. There were attached males with severely damaged eyes in present study. Poor eye condition is defined if one or both eyes are rough to be touched and blocked by fouled organisms (Schwab and Brockmann, 2007). In this study, only 9% of males were observed with poor eye condition.

Carcasses of horseshoe crabs were found onshore as the animals are treated as by-catch. The fisherman removes the horseshoe crabs entangled in fishing net and tosses the animals onshore. The animals usually die due to desiccation after prolonged exposure to high temperature and fail to overturn. Male in good condition is more likely to right itself using telson and at low risk to be stranded when the tide recedes (Brockmann and Smith, 2009). As

Table 4: Number and percentage of horseshoe crab individuals infested by epibiont

| Character | Male | Female | t-test | Total |
|-----------------------------|------|--------|--------|-------|
| No. of individual collected | 86.0 | 75.0 | | 161 |
| Individual collected (%) | 53.4 | 46.6 | | 100 |
| No. of individual fouled | 39.0 | 13.0 | 0.295 | |
| Individual fouled (%) | 45.3 | 17.3 | | |

Table 5: Individual horseshoe crab *Tachypleus gigas* with notable physical condition

| Condition of body part | Male (n = 86) | Female (n = 75) |
|------------------------|---------------|-----------------|
| Carapace | | |
| Damaged | 7 | 9 |
| Eye | | |
| Missing | 1 | 0 |
| Covered | 7 | 0 |
| Telson | | |
| Deformed | 1 | 2 |
| Broken | 4 | 11 |
| Missing | 0 | 1 |

Table 6: Mean and standard deviation and range of physicochemical water parameters at Balok Beach, Kuantan, Pahang, Malaysia

| Parameters | Ranges | Mean± SD |
|--------------------------|-------------|------------|
| Temperature (°C) | 28.41-32.73 | 30.27±0.93 |
| Salinity (ppt) | 27.70-31.93 | 30.69±0.88 |
| DO (mg L ⁻¹) | 5.13-7.860 | 6.63±1.00 |
| pH | 6.73-7.650 | 7.30±0.42 |

Table 7: Comparison between type of sediment and grain size of spawning sites of different horseshoe crab species

| Species | Type of sediment | Range of grain size | Reference |
|-------------------------------------|-----------------------------|---------------------|--|
| <i>Limulus polyphemus</i> | Fine sand with broken shell | 0.30 mm | Botton (1984) |
| | Mixed sand and gravel | | Penn and Brockmann (1994), |
| | Medium to coarse sand | | Nordstrom <i>et al.</i> (2006) |
| <i>Carcinoscorpius rotundicauda</i> | Muddy | 0.35-0.50 mm | Avissar (2006) |
| | | | Rudloe (1980), |
| <i>Tachypleus gigas</i> | Fine sand | 0.182-0.203 mm | Debnath and Choudhury (1989), Li (2008) |
| | | 2.18-2.46 phi | Rudloe and Herrnkind (1976), |
| | | 0.182-0.221 mm | Chatterji and Abidi (1993) |
| | | 0.063-0.125 mm | Chatterji <i>et al.</i> (1996), Chatterji and |
| | | 0.12-0.25 mm | Shaharom (2009), Mishra (2009), Current study |
| <i>Tachypleus tridentatus</i> | Medium to very coarse sand | 0.4-1.8 mm | Chen <i>et al.</i> (2004), |
| | Medium fine sand | | Almendral and Schoppe (2005) |

described by Botton and Loveland (1989), a female horseshoe crab sampled at study site was found with a stubby telson and a male has a curved telson. Horseshoe crab with telson abnormality is at risk of become stranded once overturned by waves.

A female horseshoe crab was found with damaged fifth walking appendage. The walking appendages have higher tendency of damage because horseshoe crab uses the appendages to push itself into the sand sediment (Duffy *et al.*, 2006).

Physicochemical parameters of Balok Beach coastal water: Physicochemical parameters of water including temperature, salinity, dissolved oxygen content and pH were measured *in situ* at Balok coastal water during May 2009 and April 2010. Table 6 showed the measured physicochemical parameters at the study area.

Sediment grain size of Balok Beach: The sand sediment of study site comprises 93.78% of fine sands with mean grain size ranged from 120 to 250 µm and silt-clay content is 5.58%. Each horseshoe crab species prefers a specific range of sediment grain size for nesting site (Table 7). Medium grain size is most ideal for the aeration of fertilized eggs (Rudloe and Herrnkind, 1976). It is reported that the moderate sorted sediment promotes hatching and survival of larvae (Chiu and Morton, 2003c). Shuster (1985) suggested sediment size and sorting affect both water infiltration and exfiltration through the beach. While, Rudloe and Herrnkind (1976) noted that an increase in the grain size does not favor horseshoe crab nesting.

CONCLUSION

The sand sediment of Pantai Balok mainly is made of fine sands with mean grain size ranged from 120 to 250 µm. Horseshoe crab nests can be found at study site after the tide recedes.

Out of the 86 males and 75 female horseshoe crabs *T. gigas* sampled, 49 were amplexus pairs. The size of the largest male and female measured were 241 and 320 mm, respectively.

More males were found ashore hence the sex ratio was male biased. In comparison to the Atlantic horseshoe crab, occurrence of satellite male was not observed. Local community harvests the female horseshoe crabs for two purposes, own consumption and income source (sold to middleman for exports to Thailand). Based on prosomal width size classes, the horseshoe crabs were found to be fourteenth to sixteenth instars.

Fewer horseshoe crabs were sighted during north-east monsoon due to the occurrence of heavy precipitation, strong surf and flooding at the study site. Spawning peaks were in May and September. Males were more prone to epibiont infestation than females. This is basically due to its nesting behavior, whereby males carapace were exposed to the environment of epibiont. Four epibiont species found on the carapaces of these horseshoe crabs were acorn barnacle *Balamus*, pedunculate barnacle *Octolasmis*, conical slipper shells *Calyptrea* and flat slipper shells *Crepidula*. Considering the study site is near to fish landing jetty, it is not a surprise to discover horseshoe crabs with damaged eyes, carapaces and telsons, possibly due to collision with boats.

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