## ALLOMETRIC VARIATIONS OF HORSESHOE CRAB (*TACHYPLEUS* GIGAS) POPULATIONS COLLECTED FROM CHENDOR AND CHERATING, PAHANG, PENINSULAR MALAYSIA

# ISMAIL, N.<sup>1\*</sup>, JOLLY, J. J.<sup>1</sup>, DZULKIPLY, S. K.<sup>1</sup>, MOHD MUSTAKIM, M. K.<sup>1</sup>, NIK MOHD HAFIZ, A.<sup>1</sup>, IZZATUL HUDA, A. G.<sup>1</sup>, TAIB, M<sup>1</sup>., SHAMSUDDIN, A. A<sup>2</sup>. AND CHATTERJI, A.<sup>3</sup>

<sup>1</sup>Makmal Belangkas, <sup>2</sup>Institute of Marine Biotechnology, <sup>3</sup>The Institute of Tropical Aquaculture, Universiti Malaysia Terengganu, Mengabang Telipot, 21030 Kuala Terengganu, Terengganu, Malaysia.

\*Corresponding author: noraznawati@umt.edu.my

**Abstract:** *Tachypleus gigas* (Müller) has the widest latitudinal dispersion of the three Asian horseshoe crab species, laying out across the western Pacific Ocean from Japan to the Philippines, extending into the Indian Ocean and only found on the east coast of Peninsular Malaysia. To date, the population of this species has decreased due to active consumption as a delicacy for both domestic and international markets. A study was conducted to determine the relationship between one body parameter and the others. An allometry analysis was applied to study the allometric variability of sixty horseshoe crabs (*Tachypleus gigas*, Müller) collected from two different populations of Chendor and Cherating Pahang of Peninsular Malaysia. The values of all parameters: carapace width, total length and total weight were recorded and analysed using correlation-coefficient and regression formulations. It was found that there were linear allometric relationships between these parameters. This showed that the growth in these parameters were isometric in both populations. There were no significant differences in morphometric characteristics between both male and female and between the two populations. It can be concluded that a proportional increment in morphometric characters were seen with an increase in age.

KEYWORDS: Horseshoe crabs, allometry, morphology, growth

## Introduction

Allometry is the statistical analysis study of relationship between size and shape (Chatterji et al. 1977). This study is done to determine relationship between different body parameters and growth. Horseshoe crab, the oldest living creature on the planet, has four extant species still living Limulus polyphemus (Linnaeus), Tachypleus tridentatus (Leach), Tachypleus gigas (Müller) and Carcinoscorpius rotundicauda (Latreilli). The two species of T. gigas and C.rotundicauda are consumed as a delicacy in South East Asian countries such as Thailand and Cambodia. In addition to the above, the amoebocyte lysate from the horseshoe crab is an important source used for the detection of endotoxin in the production of biomedical devices.

The number of horseshoe crabs are is dwindling around Malaysia (Ismail *at el.*, 2011). The consumption of this animal may be the cause

Received: 24 October 2011 / Accepted: 6 August 2012

of its decline; therefore it is important to have baseline on the well-being of live specimens. The relationship between carapace width, total length and total body weight is an important indication of the horseshoe crabs' general well- being. The value of correlation coefficient (r) expresses whether there is a significant relationship between these three parameters. If there is a positive significant relationship between these three parameters, they will influence the growth of each other and vice versa.

In this paper, comparison between lengths of carapace to prosomal width, total length of ventral prosomal to telson and weight of horseshoe crabs were done on *T. gigas* from Chendor and Cherating, Pahang, Malaysia to determine the general well-being of horseshoe crabs' and its significance to each parameter. Previous studies have shown that prosomal widths, total weight and total length are correlated with age or developmental stage (Shuster, 1954). In other words, those parameters have positive significant relationship between them. Horseshoe crab also generally exhibit positive allometry between size and weight, however weight gain is found to be faster than the growth of carapace width and total length (Lee and Morton, 2005). It has been reported by other researchers that the same factors have influenced the morphometric characters and their allometric relationships (Hickman, 1979; Schaefer *et al.*, 1985; Vijayakumar *et al.*, 2000; Chatteji *et al.*, 1988).

## **Materials and Methods**

#### Study sites

Samples were collected from Chendor and Cherating beaches. Cherating and Chendor beaches are less than 50 km north of the city of Kuantan on the east coast of Peninsular Malaysia. There are fisherman villages nearby the beaches and horseshoe crabs were collected along the sandy clean seashores. These sites are pristine and conserved.

#### Data collection

The live specimens of horseshoe crabs (T. gigas) were randomly collected from Chendor (4°9'37"N 103°24'3"E) (1) and Cherating (4° 6'0" N, 103° 23' 0" E) (2), Pahang, Malaysia. The live samples were then kept in tanks for further use. The measurement of total length (from the tip of the carapace to the tip of the telson), prosomal width and total body weights for both sexes were recorded. The length and width measurements were done to the nearest full millimetre. The weights were measured using a single-pan electronic balance (Wh-B05, Malaysia) to the accuracy of 1 g. There were sixty samples of horseshoe crabs (T. gigas) (fifteen males and fifteen females of horseshoe crabs from each population) collected from two populations of Chendor and Cherating Pahang.

#### Analysis of data

Data collected (n=60) for both sexes were analysed using the method as described by Chatterji *et al.* (1977) were log transformed and the regression log carapace width, total length to

total body weight was calculated by least square method. Relationships of all parameters were expressed using regression equation (Y=a + bX) where 'a' and 'b' were considered as additive and multiplying constants. Log Y = a + b Log X was calculated separately for each group and a straight line was fitted to scatter diagram using Microsoft Excel 2007. The relationship between the width of carapace, total length and total body weight of horseshoe crab were determined in this analysis. It is considered significant when  $r^2 = 0.90$  and above, and it is not significant when  $r^2 = 0.89$  and below with reference to Srijaya *et al.* (2010). The p value is equal to p>0.05 if it is significant and vice versa.

## **Results and Discussion**

A summary of the total body weight, total body length and carapace width relationship along with the test of significance for the males and females specimens collected from Chendor and Cherating are presented in Tables 1, 2 and 3.

The carapace widths of female specimens were between 190 to 250mm, whereas male, 130 to 190mm. The total lengths of female specimens were between 400 to 500mm, whereas male, 300 to 390mm. The body weight for female ranged between 514 to 1145g, whereas for male 180 to 393g.

The straight line represented the calculated total body weight, total body length and carapace width of regression lines after transforming the data into their logarithmic forms (Figure 1a-b, 2ab, and 3a-b). The mean values of all measurements of males and females are shown in Table 4 and Figure 4. A regression equation was applied in order to analyse all body parameters in details. A straight line was obtained in both the populations and in all relationships either with normal values or when values were converted into logarithmic values. This indicated that the increment in body parts followed an isometric pattern. In Chendor population, the relationship between carapace width and total body weight was significant in females ( $r^2 = 0.99$ ) as well as in males ( $r^2 = 0.98$ ). Similarly, for the females and males of the same population, the relationship between total length and total body weight was also found significant

Ν	Site	Sex	Regression equation	Co-relation coefficient	Significance			
1	Chendor	Male	Log Y = 1.0145 + 1.1923 X	0.982	Significant			
		Female	Log Y = 0.2069 + 2.0408 X	0.986	Significant			
2	Cherating	Male	Log Y = 0.2283 + 1.8474 X	0.986	Significant			
		Female	Log Y = -1.1385 + 3.0172 X	0.969	Significant			
	Table 2: Total length and body weight relationship of <i>T. gigas</i> of Chendor and Cherating.							
Ν	Site	Sex	Regression equation	Co-relation coefficient	Significance			
1	Chendor	Male	Log Y = 1.4163 + 0.7261 X	0.981	Significant			
		Female	Log Y= 0.1885 + 1.6809 X	0.983	Significant			
2	Cherating	Male	Log Y = -0.3745 +1.8482 X	0.927	Significant			
		Female	Log Y= -0.4724 + 2.0728X	0.979	Significant			
Table 3: Total length and carapace width relationship of <i>T. gigas</i> of Chendor and Cherating.								
Ν	Site	Sex	Regression equation	Co-relation coefficient	Significance			
1	Chendor	Male	Log Y = 0.0573 + 0.8308 X	0.981	Significant			
		Female	Log Y= 0.2355 + 0.9486 X	0.888	Not Significant			
2	Cherating	Male	Log Y = 1.4914 + 1.7474 X	0.842	Not Significant			
		Female	Log Y= 0.4406 + 1.0701 X	0.943	Significant			
Table 4: Mean of the different body measurements with their standard deviation.								
	Parameters	Parameters Chendor Cherating			g			

Table 1: Carapace width and body weight relationship of T. gigas of Chendor and Cherating.

Parameters	Chendor		Cherating	
	Males	Females	Males	Females
Carapace width	$16.5 \pm 1.22$	$21.9 \pm 1.35$	$16.5 \pm 1.53$	$21.9 \pm 1.40$
Total length	$34.4\pm2.88$	$45.8\pm2.78$	$35.5\pm1.78$	$46.2 \pm 2.68$
Total body weight	$340.2\pm21.03$	$958.7\pm97.86$	$294.0\pm29.60$	$879.3 \pm 115.07$

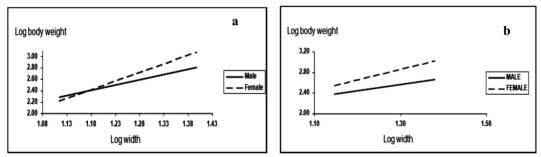


Figure 1: Relationship between log carapace width and log total body weight of male and female horseshoe crab of *T.gigas* of Chendor (a) and Cherating (b).

(females:  $r^2 = 0.98$  and male:  $r^2 = 0.98$ ). However, the relationship between total length and carapace width was not significant in males of Cherating ( $r^2$ = 0.84) and females from Chendor ( $r^2$ = 0.89).

In males of Chendor population the 'b' value was 1.19 whereas in females of the same population it was 2.04. This showed that

females of the Chendor population increased in their weight more rapidly as compared to their carapace width (Figure 1a). Similarly, the males of Cherating population showed 'b' value as 1.85 whereas females was 3.02 which indicated that the weight in female increased three times higher than the increase in carapace width (Figure 1b).

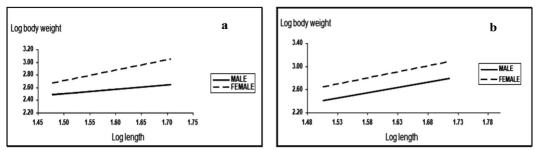


Figure 2: Relationship between log total length and log total body weight of male and female of horseshoe crab of *T. gigas* of Chendor (a) and Cherating (b).

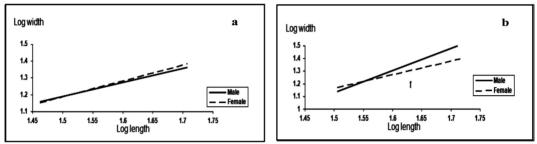


Figure 3: Relationship between log total length and log carapace width of male and female of horseshoe crab of *T.gigas* of Chendor (a) and Cherating (b).

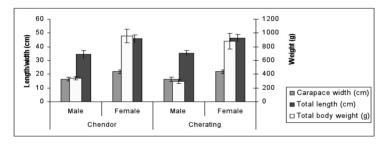


Figure 4: A comparison between carapace width, total length and total body weight of male and female horseshoe crab *T.gigas* from Chendor and Cherating.

A comparison of the carapace widths and body weights between the population of Chendor and Cherating was made which clearly indicated that both sexes of Cherating showed better growth in their carapace widths and weights as compared to the population of Chendor. However, the regression analysis showed that the carapace width and weight relationships in male specimens were significant in Cherating compared to Chendor, whereas for the female specimens, the regression analysis was significant in Chendor compared to Cherating. In both populations of Chendor and Cherating, result showed that females were heavier than the male specimen populations.

According to Srijaya *et al.* (2010), the ratio of carapace width and total length in *T. gigas* 

increased with the increase of age of the horseshoe crab which suggested that, as the animals grew older, they become wider and values of carapace width exceeded carapace length. However, in the male population of Cherating, carapace width and total body weight increased more rapidly than the population of Chendor. In contrast, total body weight and carapace width for females from Chendor increased more rapidly than females from Cherating. In both males and females population of Chendor and Cherating, a significant difference in these morphometric characters were observed which showed that in both sexes the allometric relationship was isometric but the pattern in growth increment was different. In comparison, the males of Chendor population showed 'b' value as 0.73 whereas females showed 1.68. This clearly indicated that the weight in female increased more rapidly than the increase in carapace width (Figure 2a). Similarly in males of Cherating population the 'b' value was 1.85 whereas in females of the same habitat it was 2.07 which showed that females of the Cherating population increased in their weight more rapidly as compared to their total length (Figure 2b).

A comparison of the total body lengths and body weights between the population of Chendor and Cherating indicated that both sexes of both sites showed similar growth of their total body lengths and weights. Regression analysis showed that the total body length and weight relationships in male specimens were significant ( $r^2 = 0.981$ ) in Chendor compared to Cherating  $(r^2 = 0.927)$ , whereas for the female specimens, the regression analysis was significant ( $r^2 = 0.983$ ) in Chendor compared to Cherating. In both populations, results obtained showed that females were heavier than the male specimens in both Chendor and Cherating populations. The same phenomenon was reported by Vijayakumar et al., (2000), regarding the linear growth of total length and body weight parameters.

However, in comparison with males and females from Chendor, the 'b' values were 0.83 and 0.95 respectively. This showed that the increase in their carapace width was almost the same rate as an increase in their total body length (Figure 3a). Similarly in Cherating populations, the 'b' value of male was 1.75 whereas in females of the same habitat it was 1.07. This showed that females of the Cherating population increased in their weight more rapidly as compared to their carapace width (Figure 3b).

A comparison between the total body lengths and carapace widths of the two populations of Chendor and Cherating was done which clearly indicated that both males and females of Cherating showed better growth in their total lengths and carapace widths as compared to the population of Chendor. Regression analysis showed that the total body lengths and carapace width relationships in male is higher ( $r^2 = 0.981$ ) in Chendor compared to male in Cherating  $(r^2 = 0.842)$  and female specimens were significant in Cherating  $(r^2 = 0.943)$  compared to Chendor  $(r^2 = 0.888)$ . In Cherating population, males were heavier than the female specimens which contradicts with the specimens from Chendor. Carapace width-total length relationship was found to be proportionate and uniform as indicated by the linear relationship.

In the present study, the allometric relationship between carapace widths, total body weight and total length were almost linear for all three parameters examined. The growth pattern of the carapace width, total body length and total body weight were isometric in both populations (Table 4; Figure 4). In most aquatic animals the ratio between body parts and the increment of body length does not show a constant growth.

The carapace width, total body length and total body weight in T. gigas has been reported to increase with age of the horseshoe crab, which suggested that as the animals grew older, they become wider, longer and heavier. Chatterji et al. (1988) stated that the allometric relationship between carapace length and soft body parts provides a better understanding of the growth of different species of horseshoe crabs C. rotundicauda. This phenomenon is proven through the growth pattern as analysed from the data above. According to Chatterji et al. (1988) and Vijayakumar et al. (2000), different diet and food supply will influence the growth patterns for species of horseshoe crabs, T. gigas and C. rotundicauda. According to Srijaya et al. (2010), in another species of horseshoe crab, the measurements of all morphometric parameters in males and females of C. rotundicauda of Setiu population have been recorded higher as compared to Gelang Patah (Johor) population. The total lengths of males and females were higher by 9.30 and 1.98%, respectively as compared to Gelang Patah (Johor) population. The carapace length was higher by 2.65% whereas carapace width (1.26%) and telson length (16.26%) in males of Setiu population as compared to males of Gelang Patah (Johor). Similar trend was observed in female population of Setiu where the carapace length was higher by 9.46%, carapace width (7.90%) and telson length (13.34%) as compared

to the females of Gelang Patah (Johor). The relationship between total length and carapace width has been reported to be highly significant (0.0639+0.96) in males of population of Johor. Srijaya *et al.* (2010) summarised that there could be substantial physiological differences among the two populations of *C. rotundicauda* due to changed habitats depending to individual acclimatisation of species or genetically-fixed adaptations of the same species.

From these findings, it is very clear that comparison between lengths of carapace to prosomal width, total length of ventral prosomal to telson and weight of horseshoe crabs done on *T. gigas* from Chendor and Cherating, Pahang, Malaysia showed similar patterns of growth in the allometric variations in the populations from these two populations, which strongly suggest that these specimens may be derived from the same independent stock. This could be due to the close proximity of the two locations due to similar local environmental conditions and food sources.

#### Acknowledgements

We thank Ministry of Science, Technology and Innovations Malaysia for funding this project through Institute of Pharmaceutical and Nutraceutical Malaysia.

#### References

- Chatterji, A., Siddiqui A. Q., & Khan, A. A. (1977). Length-weight relationship of a carp, Labeo bata (Ham.) Proc. Inidan Acad. Sci. 88(B): 189-195.
- Chatterji, A., Vijayakumar, R., & Parulekar, A. H. (1988). Growth and morphometric characteristics of the horseshoe crab *Carcinoscorpius rotundicauda* (Latreille) from Canning (West

Bengal), India. Pak. J. Sci. Ind. Res. 31(5): 352-353.

- Hickman, J. C. (1979). The basic biology of plant numbers. In Tropics in plant population biology (Eds. O.T. Solbrig, S.K. Jain, G.B. Johnson and P.H. Raven). New York: Columbia University Press. 232-263.
- Ismail, N., Taib, M., Shamsuddin, A. A., & Shazani, S. (2011). Genetic variability of wild horseshoe crab, *Tachypleus gigas* (MÜller) in Tanjung Dawai, Kedah and Cherating, Pahang of Peninsular Malaysia. *European Journal of Scientific Research*. 60 (4): 574-583.
- Lee, C. N., & Morton, B. (2005). Journal of Experimental Marine Biology and Ecology. 318: 39–49.
- Schaefer, R., Trutschler, K., & Rumohr, H. (1985). Biometric studies on the bivalves Astarte elliptica, A. borealis and A. montagui in Keil bay (Western Baltic Sea). Helgolender Meeresunters. 39: 245-253.
- Shuster C. N. Jr. (1954). A horseshoe "crab" grows up. *Ward's National Science Bulletin.* 28: 3.
- Srijaya T. C., Pradeep, P. J., Mithun, S., Hassan, A., Shaharom, F., & Chatterji, A. (2010). A New record on the morphometric variations in the populations of horseshoe crab (*Carcinoscorpius rotundicauda*) obtained from two different ecological habitats of Peninsular Malaysia. *Our Nature.* 8: 204-211.
- Srijaya, T. C. (2011) Morphometric variabilities in the adult populations of mangrove horseshoe crab *Carcinoscorpius rotundicauda* (Latreille), embryonic development and their larval behavior under different culture conditions. Ph.D. thesis, University Malaysia Terengganu. 326.
- Vijaykumar, R., Das, S., Chatterji, A., & Parulekar, A. H. (2000). Morphometric characteristics in the horseshoe crab, *Tachypleus gigas* (Athropoda: Merostomata). *Ind. J. Mar. Sci.* 29: 333-335.