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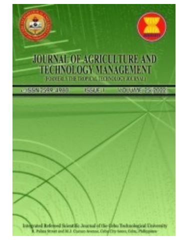

Original article

Morphometric analysis of trumpet shell, charonita tritonis, (littorinimorpha: ranellidae) from Sarangani province, Philippines

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ABSTRACT

The *C. tritonis* is an important biological indicator in coral reef ecosystem. Conservation of the species is essential to maintain the integrity of the reef. This study was conducted to investigate variations on the morphometric characters on Trumpet shell, *Charonia tritonis* Linneaus 1758, to elucidate shell developmental history. A total of 50 shells were measured for length and weight for morphometric analysis. The result on allometric relationships indicated that during the shell's development, the increase in aperture length was not proportional to the increase of the shell width. However, the increase in aperture length was significantly related to the increase in weight and total shell length. These variations on morphometric relationships could possibly be attributed to the movement and feeding behavior of the organism. The continued pressure of the foot muscle as it extends while moving and feeding pushed the aperture length causing it to increase in length. It is recommended for further study to include other landmark variations of the trumpet shell such as shell spire and body whorl for deeper understanding on shell developmental biology.

KEYWORDS: *adaptive strategy, evolution, marine, snail*

1 INTRODUCTION

Morphometry is the quantitative study on variations of biological shapes and its covariation with other parameters across various disciplines (Rohlf & Slice, 2004). In biology, morphometry allows insights into organismal evolutionary development (Roth & Mercer, 2000). In systematics, studies on morphology normally utilizes morphometric techniques (Rohlf, 1990). In studies on ontogenetic development, morphometry plays a role in understanding ecological and developmental patterns in relation to causal agents and other biological processes (Roth & Mercer, 2000).

Morphometric characteristics on the shells of gastropods contain rich sources of taxonomic information that can be used to interpret relationships between and among species (Chiu *et. al.,* 2002). The behavior-induced shell development is an adaptive strategy that allows an organism to improve its fitness to its environment (Hoverman *et. al*., 2005; Miller and Denny, 2011) Logically, the behavior of the snail can affect the growth of the shell thus understanding the shell leads to understanding the behavior of the snail (Dewitt *et. al.,* 1999).

The trumpet shell, *Charonia tritonis* (L., 1758) is a relatively large marine mollusc belonging to Family Ranellidae with a spiral shell that can reach a length of 20 inches or 50 centimeters. The trumpet shell was previously classified under the family Cymatiidae (Beu, 1970). However, the former family, Cymatiidae, has been split into two families: Ranellidae (tritons) and Personidae (Distorsios) (Beu, 1998). It is widely distributed throughout the Indo-Pacific region, Red Sea, tropical coast of Africa and in southern Japan. Due to its large size, beautiful shell design, its practical use as a ceremonial trumpet and food source made this shell as one of the most well-known and highly regarded.

Ecologically, *C. tritonis* play a role as natural biological control in coral reefs as the main predator of coral eating crown-of-thorns starfish (*Acanthaster planci)*. The shell provides protection from external threats and may influence the predatory behavior of *C. tritonis*. This study aimed to determine the morphological characters on the shell and elucidate its role in the life history of *C. tritonis.* Information on shell morphometry is important because it can capture shell shape that can establish biological structure for better understanding on the shell growth of the species. This study can be used in revisiting existing phylogenetic estimates of the genus *Charonia* and compare them based on the result of the morphometric analyses.

2 MATERIALS AND METHODS

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Sampling

The study site, located at Glan, Sarangani Province, is known for the presence of *C. tritonis* (Fig. 1) in. A total of 50 triton shells were measured for length and weight based on the morphological characters in Figure 2. There were no live samples collected for this study. All samples were collected by the local residents in the area prior to the conduct of the study. Samples were stored in their houses as ornamentals or sold to shell traders. Most of the shells were kept for months to a year; if not sold. The gratuitous permit was first secured due to the law regulating the collection of the trumpet shells. We conducted individual interviews with the locals and asked if they have trumpet shells.

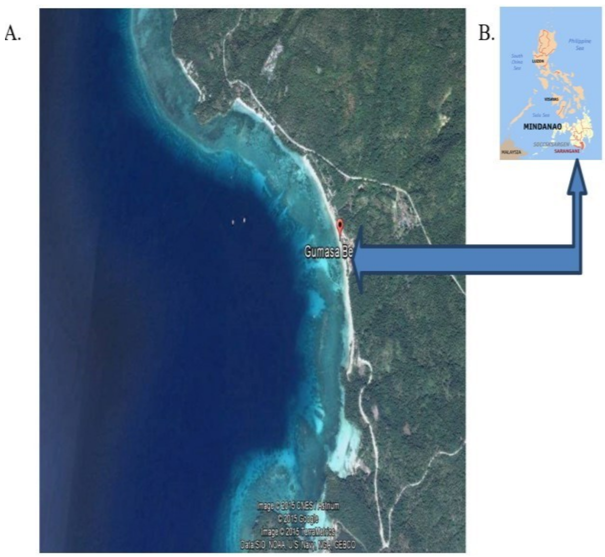

Figure 1. Map showing Barangay Gumasa, Glan, Sarangani Province where the Trumpet shells were collected. A. Map showing the reef area of the sampling site; B. The Philippine map indicates the location of the sampling site.

In this study, morphometric measurements were conducted on 50 trumpet shells, *Charonia tritonis*, sourced from Sarangani Province, Philippines. The measurement was done for weeks since most shells come from different barangays and some are not immediately available which required several visits to finish measuring all the samples.

The morphometric characteristics were the total shell length (TSL), aperture length (AL), shell width (SW) and weight (Fig. 2). The total shell length, shell width and aperture length were measured using a measuring tape to the nearest 0.01cm. Total shell length was measured from apex to the end of the siphonal canal. The shell was dry and without soft parts. The shell was weighed (shell free body weight) to the nearest 0.01gm using analytical balance (Sartorius, NTEP).

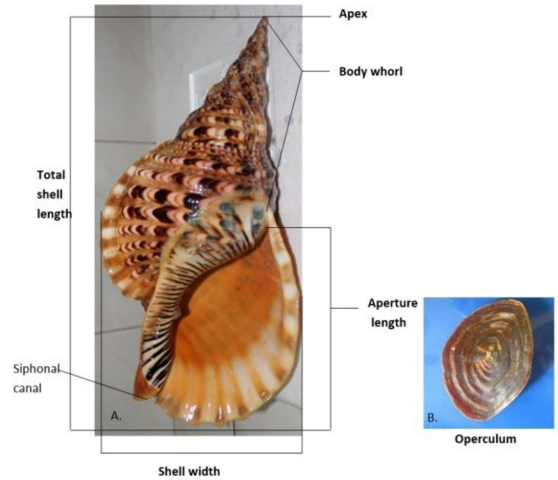

Figure 2. Morphometric characters: A. The triton shell showing the morphometric characters considered in this study: total shell length, shell width and aperture length. B. The operculum that functions as a covering of the aperture opening.

The relationship between and among these morphometric parameters were described by linear and exponential values. Total Shell length (TSL, cm) and weight (W, g) was expressed by the equation (Sokal and Rohlf, 1995):

$W=aTSL^b$.

Where *a* and *b* are the coefficients of the functional regression between TW and SL. To confirm if the values of *b* obtained in a linear regression were significantly different from the isometric value (b=3), a *t*-test (H0, b=3) with a confidence level of 95% was applied, expressed by the equation:

$t_s= (b-3)/SE_b$

Where t_s=*t*-test value, b=slope, SEb=standard error of the slope (b). The 95% confidence interval, CI of b was computed using the equation:

$CI=b\pm(1.96xSE)$

Where SE is the standard error of *b*. All the statistical analyses were considered at significance level of 5% ($P< 0.05$).

Statistical Treatment

Technical data processing was done in order to achieve the desired result of this research. Statistical analysis includes correlation, fitness models, and analysis of variance using statistical programs of SPSS and Microsoft Excel.

3 RESULTS AND DISCUSSION

Morphometric analysis

A total of 50 shells were measured for morphometric analysis. The morphometric characters in this study were total shell length (TSL), weight (W), width (SW) and

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aperture length (AL) of the shell of the *C. tritonis*. The values of the morphometric characters are presented in Table 2. Among the characters measured, the shell weight (W) had the highest standard deviation at 262.24gm while the aperture length (AL) had the lowest standard deviation at 1.83cm. These results mean that the weights of the shell varied greatly while that of the aperture length (AL) did not.

Table 2. The mean, minimum and maximum total shell length, shell width, weight, aperture length and standard deviation of the *Charonia tritonis*.

Characters	Mean	Std. Deviation (SD)	Min – Max
Weight (gm)	648.00	262.24	200 - 1150
Shell Width (cm)	15.14	2.18	11.8 – 20.6
Total Shell Length (cm)	33.05	3.49	27 – 39.8
Aperture Length (cm)	15.81	1.83	12.1 – 18.5

Next step would be to determine the relationships among the morphometric characters. In Fig. 4, the x axis, expressed by the line on the scatterplot, is represented by the aperture length as a dependent variable. The y axis are the shell weight, shell width, and total shell length which are the predictors. The predictors are used for the estimation of the growth of the dependent variable. The data set or "dots" were almost on the line and it followed an upward trend that signified a strong positive linear relationship.

These linear relationships, shown in Fig. 4, indicated that the increase in aperture length is influenced by the growth of the shell in weight, width and total length. To quantify or measure the extent of these relationships among the morphometric characters, a correlation regression was needed.

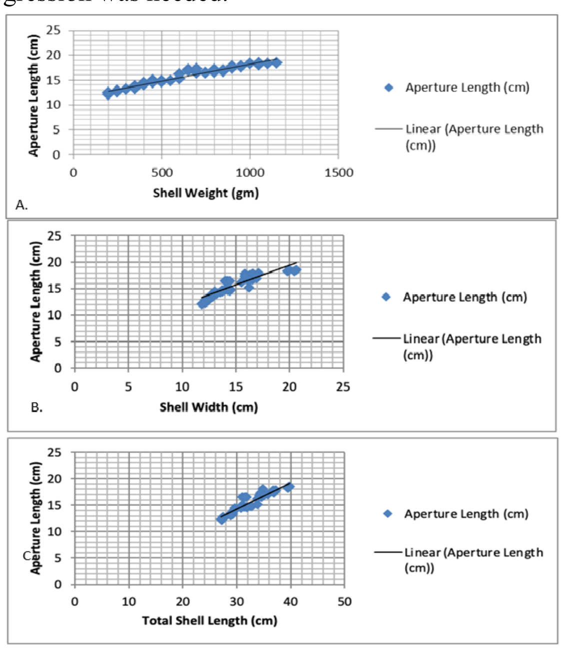

Figure 4. Correlation between aperture length and the morphometric characters of *C. tritonis*: A. shell weight of; B. shell width; C. total shell length.

The frequency distribution of the measurement values of the aperture length was conducted first prior to regression to check for distribution error. The error in distribution can affect the relationship shown on the scatterplot so it is important to determine if the distribution is normal. In Fig. 5, the measurement values of the aperture length are presented by the line on the histogram. The curve line on the histogram has validated the error of distribution as normal hence regression of the morphometric variables versus the aperture length is possible.

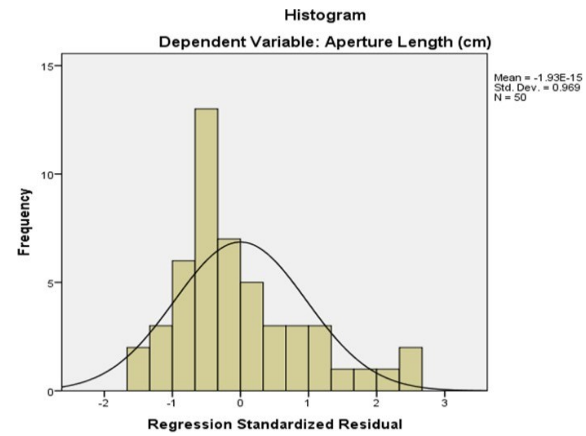

Figure 5. Frequency distribution for aperture length.

Effects of Allometric Growth on Shell Morphology

Regression was conducted using Pearson correlation to measure the extent of the relationships between the morphometric characters as shown in Table 3. The correlation coefficients (*r*) ranged from ±0.897 - ± 0.969 which indicated a very high to near perfect relationships and these relationships are significant even at the 0.01 level. All the p-values are 0 which suggests that the weights, widths, total shell lengths and aperture lengths of the shell are significantly correlated (Table 3).

The shell weights and aperture lengths almost have a perfect positive relationship with a Pearson *r* value of 96.9% ($p < 0.00$). This means that the heavier the shell weight, the longer is its aperture likewise the lower is its weight, the smaller is its aperture length.

Table 3. Pearson Correlation among the four morphometric characters: weight, shell width, total shell length and aperture length.

Characters		Weight (gm)	Shell Width (cm)	Total Shell Length (cm)	Aperture Length (cm)
Weight (gm)	Pearson Correlation	1	.912**	.939**	.969**
	Sig. (2-tailed)		.000	.000	.000
Width (cm)	Pearson Correlation	.912**	1	.955**	.897**
	Sig. (2-tailed)	.000		.000	.000
Total Shell Length (cm)	Pearson Correlation	.939**	.955**	1	.937**
	Sig. (2-tailed)	.000	.000		.000
Aperture Length (cm)	Pearson Correlation	.969**	.897**	.937**	1
	Sig. (2-tailed)	.000	.000	.000	

**. Correlation is significant at the 0.01 level (2-tailed).

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N=50

Similar interpretations can be made with the following pairs: shells' weights and widths, shells' weights and total lengths, shells' widths and total shell lengths, shells' widths and aperture lengths and total shell lengths and shell's widths, and shell's total shell lengths and aperture lengths.

Since the shells' measurements are highly correlated, it would also be interesting to investigate whether the length of the shell's aperture is a function of its weight, width, and total length. Thus, regression analyses were performed using aperture length as the response variable, and the weights, widths, and total lengths as predictor variables.

The Analysis of Variance (ANOVA) Table 4 for Model 1 shows that when the aperture length is regressed using shells' lengths, widths, and weights, the significance level is 0.00. As reflected from the table, the p-value of the Model 1 is significant at the 0.01 level.

Table 4. ANOVA for Regression Model 1 for aperture length using total shell length, weight, and shell width as predictors.

Model		Sum of Squares	Df	Mean Square	F	Sig.
1	Regression	155.047	3	51.682	267.088	.000[b]
	Residual	8.901	46	.194		
	Total	163.948	49			

a. Dependent Variable: Aperture Length (cm)

b. Predictors: (Constant), Total Shell Length (cm), Weight (gm), Shell Width (cm)

Based on the results found in Table 5, regression of the aperture length shows that weight and total shell length have p-values of 0.00 and .023, respectively, which are significant at 0.05 level. However, the shell width has p-values of .334 which is not significant in relation to the aperture length.

Table 5. Regression Model 1 for shell's aperture length.

Model		Unstandardized Coefficients				95.0% Confidence Interval for B	
		B	Std. Error	t	Sig.	Lower Bound	Upper Bound
1	(Constant)	8.101	1.344	6.026	.000	5.395	10.807
	Weigth (gm)	.005	.001	7.559	.000	.004	.007
	Shell Width (cm)	-.096	.099	-.976	.334	-.295	.102
	Total Shell Length (cm)	.173	.073	2.357	.023	.025	.320

Based on the results of Table 5, the aperture length can be predicted using the following model:

Aperture Length = 8.101 + 0.005 Weight – 0.96 Width + 0.173 Total Length.

The Model Fit is 94.2% (Table 6) which means that 94.2% of the variance in the shell's aperture length can be explained by the variance of its weight, width and total length.

In other words, the shell's weight, width and total lengths contributed 94.2% of the shell's aperture. Now, interpreting the individual parameters:

1. Shell weight – for every 1 gram increased in the weight of the shell, aperture length is increased by an average of 0.005 centimeter when shells' width and total lengths are fixed (p = 0.000).

2. Total Shell Length – For every 1 centimeter increased in the shell's length, aperture length is increased by an average of 0.073 centimeter when the shell's width and weight are held constant (p = 0.023).

3. Shell's Width – No interpretation is made regarding the shell's width

Table 6. Model 1 Fitness based on Coefficient of Multiple Determination.

Model	R	R Square	Adjusted R Square
1	.972[a]	.946	.942

a. Dependent Variable: Aperture Length (cm)

b. Predictors: (Constant), Total Shell Length (cm), Weight (gm), Shell Width (cm)

Presented in Table 7 are the Analysis of Variance tables for Model 2 and Model 3. Model 2 uses only the shell weight as predictor while Model 3 uses the shell's weight and total length as predictors. Note that the p-values of both models are 0.00 which is less than 0.01. This implies that the two models are also useful in predicting the shell's aperture length.

Table 7. Analysis of Variance (ANOVA) for Regression Models 2 and 3 for aperture length.

Model		Sum of Squares	Df	Mean Square	F	Sig.
2	Regression	153.799	1	153.799	727.398	.000[b]
	Residual	10.149	48	.211		
	Total	163.948	49			
3	Regression	154.863	2	77.431	400.555	.000[c]
	Residual	9.086	47	.193		
	Total	163.948	49			

a. Dependent Variable: Aperture Length (cm)

b. Predictors: (Constant), Weight (gm)

c. Predictors: (Constant), Weight (gm), Total Shell Length (cm)

Based on Table 8, Model 1 the aperture coefficient is 11.43 and weight has coefficient of .007 with a p- value of .000, suggesting that there is significant relationship with regard to shell weight and the growth of the aperture length. The result for Model 1 can be written as:

*Aperture Length = 11.428 + 0.007*Weight*

Table 8. Parameter estimates for Model 1 and Model 2 for aperture length.

Model		Unstandardized Coefficients		Standardized Coefficients			95.0% Confidence Interval for B	
		B	Std. Error	Beta	T	Sig.	Lower Bound	Upper Bound
1	(Constant)	11.43	.175		65.357	.000	11.077	11.780
	Weigth (gm)	.007	.000	.969	26.970	.000	.006	.007
2	(Constant)	8.367	1.316		6.359	.000	5.720	11.014
	Weigth (gm)	.005	.001	.749	7.501	.000	.004	.007
	Total Shell Length (cm)	.123	.052	.234	2.345	.023	.017	.228

a. Dependent Variable: Aperture Length (cm)

As given in Table 8, the Model Fit is 93.7% as shown

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in Table 8. This means that 93.7% of the variability in the shell's aperture length can be explained by the variability in its weights. In other words, the weight of the shells contributed 93.7% of its aperture length. More specifically, for every one gram increased in the weight of the shell, its aperture length is increased by an average of 0.007 centimeters.

Furthermore, Model 2 can be presented in the form:

*Aperture Length = 8.367 + 0.005*Weight +0.123*Total Length*

The Model Fit is 94.2% as reflected also in Table 9. This follows that 94.2% of the variances in the shell's aperture lengths can be explained by the variances in the shells' weights and total lengths. In terms of its parameters:

1. Shell weights – For every 1 gram increased in the weight of the shell, its aperture length is increased by an average of 0.005 centimeters holding total length constant;
2. Shell Total Lengths – For every 1 centimeter increased in the total length of the shell, its aperture length is increased by an average of 0.123 centimeters holding its weight fixed.

Table 9. Model Fitness of Model 1 and Model 2.

3. Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.969[a]	.938	.937	.4598
2	.972[b]	.945	.942	.4397

a. Predictors: (Constant), Weight (gm)

b. Predictors: (Constant), Weight (gm), Total Shell Length (cm)

c. Dependent Variable: Aperture Length (cm)

The correlation coefficient has shown significant relationships of all the parameters. However, the result of the regression of the morphometric characters may suggest that the shell width may have reached optimum expansion since it is no longer expanding even if the shell is still increasing in weight and in length. The relative increase of the aperture length without affecting the shell width has shown an allometric growth of the shell.

Gastropods, such as *C. tritonis*, appeared to be bilaterally symmetrical hence projecting a seemingly symmetrical growth. While shell growth of gastropods may be morphologically isometric (growth in length is accompanied by weight increase) other shells are allometric (unequal growth rates between and among length and weight variables). There are some shells that indicate a positive allometric growth which could mean that weight increase is superior to the growth in length and negative allometric growth when growth in length is superior to weight increase (Jokinen, 1982).

This allometric growth of the shell was reported in the study of length-weight relationship of Rapa whelk, *Rapana venosa,* by Saglam and Düzgünes (2014). In their study, the aperture length and shell width of Rapa has shown a negative relationship while the shell weight and shell length has shown positive relationship with the aperture length. Medeiros (2015) also found the same on the morphological variability of shells in populations of *Leptinaria unilamellata*. In this study, the allometric relationship between shell aperture width and shell width, as well as the relationship between the shell aperture height and shell width, indicate that, during the snails' development, the increase in shell width is not proportional to the increase of the shell aperture length.

This high to near perfect relationship of the Trumpet shell's weight to aperture length may be attributed to the growth and movement of the body of the Trumpet shell. When the body is inside the shell, the body contracts thus its size is smaller. However, when the body tends to exit through the aperture, the foot muscle expands and it moves from place to place then feeds. As the foot muscle expands, the aperture may have to expand in length to accommodate the body of the Trumpet shell. The relationship of the total shell length to the aperture length can be attributed to the increasing growth of the shell by accretion. As the shell length grows so also is the aperture length since it is part of the total shell length.

This observation of the body movements in relation to shell growth was reported in the study of Trumpet shell by Paterson and Poulsen (1988) on the Great Barrier Reef in Australia. They have observed that the size of the shell's body whorl is smaller than its aperture and when the Trumpet shell is disturbed it retracts inside the whorl thus, it has to contract its muscle to fit its body. However, when the triton starts to move, the body attaches to the ground and its muscle extends the outer boundary of the aperture hence the body has expanded much larger than when it is hiding inside its shell.

The method of attacking the prey, employed by *C. tritonis*, may also affect the growth of the shell as observed by Percharde (1972) in their study conducted in Trinidad and Tobago. *C. tritonis* exhibited a creeping approach on its prey with tentacles being swept from side to side. After contact is made with the prey, the large proboscis injects the acid saliva on the prey causing immediate paralysis. The foot muscle stretches its length, larger than the aperture length, and then wraps the prey. While engaged in feeding, the Trumpet shell holds its prey firmly by its foot muscle. This feeding behavior may have caused the continued expansion of the aperture length as it is subject to constant pressure when the foot muscle of *C. tritonis* extends its length.

The result on scatterplot has indicated a positive linear relationship on all of the morphometric characters. In quantifying the extent of the relationship, the morphometric characters were subjected to correlation regression. The result on regression has indicated that the increase in aperture length was not proportional to the

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increase of the shell width. However, the increase in aperture length was significantly related to the increase in weight and total shell length of the Trumpet shell.

4 CONCLUSION AND RECOMMENDATION

The growth allometry of the shell could have been influenced by the movement and feeding behavior of the Trumpet shell. The body mass of a Trumpet shell can affect the size of its shell since it has to increase in size as the body increases. It can also push the aperture length causing it to increase in length whenever it moves and feeds. It is recommended for further study that other landmark variations on the shell of trumpet shall be considered such as shell spire and body whorl to have a deeper understanding on developmental biology of *Charonia tritonis*.

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