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Estimation of the edible wet tissue weight of the mangrove clam *polymesoda erosa* harvested from the wild

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Abstract
A technique on estimating the edible wet tissue weight of *Polymesoda erosa* was established using Multiple Regression Analysis of five morphometric measurements namely, 1. Shell Length (SL), 2. Shell Width (SW), 3. Shell Height (SH), 4. Shell Volume (SV), and 5. Live Weight (LW). Multiple Regression Analysis was not directly used to test for the best estimator because of the high correlation between the 5 independent variables. Instead, it was performed to create an estimation formula that took into consideration all the contributions of the independent variables to the estimation of the Edible Wet Tissue Weight. One-Way ANOVA at 0.05 level of significance showed that there is a significant difference in terms of the contribution of the 5 estimator variables. B-values point to Shell Width and Shell Volume to have a significant contribution to the estimation formula. The Edible Wet Tissue Weight of *P. erosa* can be estimated as follows: Estimated Wet Tissue Weight= -10.689 (constant) + (0.109 x SL) + (0.122 x SW) + (0.026 x SH) + (0.008 x LW) + (0.045 x SV). This estimation method can be used by fisheries technicians without the need to sacrifice the organisms in experiments or monitoring applications.

Keywords: *Polymesoda erosa*, clam morphometry, wet-tissue-weight estimation, multiple regression analysis

Introduction
The mud clam *Polymesoda erosa* is an edible bivalve species that thrives in intertidal areas, estuaries, and rivers. They are highly tolerant to external desiccation, can sustain aerial respiration for a few days, and feed from subsurface water by means of water exchange through a narrow anterior gape of valves. Locally, it is called “tuway” by collectors in Sitio Lamintao, Barotac Nuevo, Iloilo, Philippines. Currently, the consumption of this bivalve species increased due to its high protein content and is considered a delicacy in some coastal areas (Norma and Norasma, 2016) [23]. Although it is not very popular in the Philippines because of the greater demand for the more favourite clam species, there have been several studies abroad that suggest its potential for Mari culture (eg. Morton, 1984; Meehan, 1982; Lopez, 1988; Sarong *et al.* 2015; Hamli *et al*, 2014; and Rizal, 2016) [21, 20, 19, 32, 15, 28]. There are already plenty of literature on the various techniques and procedures in shellfish aquaculture. For species such as *P. erosa*, it would be best to culture them along mangrove areas via aqua silviculture- an environment-friendly mangrove aquaculture program by the Bureau of Fisheries and Aquatic Resources (BFAR).
One of the subtle issues surrounding shellfish culture is on the assessment of growth especially of the wet edible tissue. Unlike other cultured species where you can directly assess growth before and after introducing them to the culture system, clams can be a little tricky. Determining growth in bivalves through allometric relationships can produce useful information for resource management and in understanding environmental changes and disturbances (Palmer 1990; Boulding and Hay 1993) [24, 4]. Growth is usually estimated indirectly in clams by measuring shell dimensions or the volume of the animal (Deval, 2001) [7]. These are practical and non-destructive methods that can be easily done both in the laboratory and in the field. Shell morphometric measurements are sufficient to estimate biomass and total flesh production (Ross and Lima 1994; Ravera and Sprocati 1997) [30, 27], but in reality, consumers do not eat the shell. What matters is inside the shell- the edible wet tissue and more often by experience, consumers are disappointed by a large clam with very little inside or amazed by a Medium-sized clam with relatively bigger meat on the inside.

It is thus critical to expect that shell dimensions will fail in estimating the edible flesh weight of a clam if not used carefully. Also, in grow out monitoring, technicians will have difficulty obtaining the fleshy part of the clam for initial weighting without sacrificing them. The same is true when future interventions and evaluations are planned where wet-tissue weight data is needed. At least in the experimental sense, it is a fact that it is hard to get baseline data in terms of edible wet tissue weight if the primary purpose of it is to culture the same animal to be assessed again at a later time. But theoretically, estimation is possible using the various shell dimensions (length, height, and width), shell volume, and live weight (shell + soft tissue inside). It is on this light that this simple investigation was carried out.

Materials and Methods

This study utilized a Correlational Research Design. Various morphometric measurements such as shell width, shell height, shell length, shell volume and live weight are tested if they are statistically associated with the edible wet tissue weight of *P. erosa* clam. One hundred fifty (150) adult Polymesoda erosa clams ranging from 40-71 mm in shell length were bought from a local collector at Brgy. Tinorian, Barotac Nuevo, Iloilo, Philippines. They were cleaned and washed of sediments. They were transported and transferred to a 50-liter container containing artificial saltwater at 15ppt salinity in the Fisheries and Marine Science Lab of Iloilo State College of Fisheries, Tiwi, Barotac Nuevo, Iloilo, Philippines and were left overnight without food to clear their guts. Only 108 clams were used in the study. Too small and too large clams in the sample were not included as a contingency measure. The possibility that outliers will introduce errors in the analysis must be minimized. Clams measuring lower than 20mm and greater than 90mm were removed as samples. Also, very small clams have relatively very small fleshy tissue to be measured accurately.

Shell length, height, and width were measured using Digital Vernier calipers to the nearest 0.01mm. The shell volume was measured via simple displacement in a beaker with known volume of water initially measured using a graduated cylinder. Depending on the size of the clam, volume adjustments are done with extra water in a separate graduated cylinder. Before placing the clam in water, an initial volume is set. After the clam is submerged in the water the displaced volume is recorded. The empirical volume was also computed from the measured shell dimensions (LxWxH). If the clam is not fully submerged, an additional measured volume is added. This method is more accurate than by simply calculating the empirical volume using the length x width x height measurements because the surface of the clam is obviously not uniform. Live weight was measured directly using a digital weighting scale to the nearest 0.01 grams. After sacrificing the animal, the fleshy tissue including the clam liquor (the fluid coming out of the fleshy tissue after scraping it from the shell) is also weighted in the digital scale.

All measurements were encoded into a Statistics Software and Multiple Regression Analysis was performed at 0.05 level of significance to determine the best predictor or estimate of edible wet tissue weight. Pearson's Correlation was used to determine relationships between all the variables. Correlation analysis also justified the use of Multiple Regression Analysis because of the predictive nature of the investigation. The issue of multi collinearity among the predictor variables was ignored because all measurements were taken from the same individual organism and that is the reason why they are all correlated. The primary goal of this study is to construct a model equation where the contribution of each morphometric measurements to the estimation was established. If there is no correlation between the predictor variables and the edible wet tissue weight, it is pointless to create an estimation formula. Since correlations are significant in this study, it supported the idea that Regression Analysis can be carried out for this purpose.

Results

The following descriptive statistics came from all the measured shell dimensions, live weights and edible wet tissue weights of *Polymesoda erosa* in millimetres and grams respectively.

Table 1: Descriptive Statistics of the Various Morphometric Measurements of *P. erosa*

		Shell Length (mm)	Shell Width (mm)	Shell Height (mm)	Live Weight (g)	Shell Volume (ml)	Wet Tissue Wt (g)
N	Valid	108	108	108	108	108	108
	Missing	1	1	1	1	1	1
Mean		53.17	58.95	30.45	53.41	76.82	6.97
Std. Deviation		7.97	8.95	4.66	22.149	19.37	3.23
Variance		63.52	80.05	21.74	490.60	375.32	10.42
Minimum		40.00	44.10	21.00	20.37	52.00	1.98
Maximum		71.30	81.70	42.50	123.67	128.00	15.87

Before Multiple Regression Analysis was performed, a correlations test between all the variables was done using Pearson's R at 0.05 level of significance. As shown in Table 2. Below, all variables are correlated with each other at 0.01 level of significance. This is a clear indication that there is a strong link between all the predictor variables (shell dimensions, shell volume, and live weight) and Wet Edible Tissue Weight.

Table 2: Pearson's R Correlation Result of All Variables

		Length	Width	Height	Live Weight	Volume	Wet Tissue Weight
Length	Pearson Correlation	1	.938**	.924**	.812**	.951**	.923**
	Sig. (2-tailed)		.000	.000	.000	.000	.000
Width	Pearson Correlation	.938**	1	.882**	.769**	.890**	.907**
	Sig. (2-tailed)	.000		.000	.000	.000	.000
Height	Pearson Correlation	.924**	.882**	1	.816**	.912**	.876**

	Sig. (2-tailed)	.000	.000		.000	.000	.000
Live Weight	Pearson Correlation	.812**	.769**	.816**	1	.784**	.777**
	Sig. (2-tailed)	.000	.000	.000		.000	.000
Volume	Pearson Correlation	.951**	.890**	.912**	.784**	1	.904**
	Sig. (2-tailed)	.000	.000	.000	.000		.000
Wet Tissue Wt	Pearson Correlation	.923**	.907**	.876**	.777**	.904**	1
	Sig. (2-tailed)	.000	.000	.000	.000	.000	

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

However, Pearson's R can only establish only the presence or absence of a relationship or link but not the model equation for the estimation of the Wet Edible Tissue Weight. This was determined using Multiple Regression Analysis. The primary purpose of doing this is not to differentiate among the various predictor variables (which is the default use of this inferential statistic) but on the creation of an estimation formula derived from a common linear function of all predictor variables in a regression plot. This was easily derived from the resulting Model Summary Output of SPSS Software. The following tables are results of the SPSS analysis.

Table 3: Summary of the Regression Model

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.935[a]	.875	.869	1.17019

a) Predictors: (Constant), Volume, Live Weight, Width, Height, Length

As shown in the table above, the R Square value of 0.875 shows that 87.5% of the variability in the Wet Edible Tissue Weight of *P. erosa* was due to the variability of the 5 predictor variables namely Shell Length, Shell Width, Shell Height, Shell Volume, and Live Weight. This is a significant result because statistically estimating the dependent variable reliably by 87.5% is very promising. The standard error of the estimate is also noticeably low with a deviation of + 1.17.

One way ANOVA at 0.05 level of significance which is still part of the SPSS regression analysis shows that at least one of the five chosen predictors have a significantly (p=0.000) greater contribution to the estimation of the Wet Edible Tissue Weight. This can also justify why Multicollinearity has to be ignored. Despite the Correlation test results showing that all tested predictors have a strong link towards the Wet Edible Tissue Weight, One-Way ANOVA shows that the predictors are significantly different from each other.

Table 4: One-Way ANOVA of the Predictors in the Model

Model	Sum of Squares	df	Mean Square	F	Sig.
Regression	975.279	5	195.056	142.445	.000[b]
Residual	139.673	102	1.369		
Total	1114.951	107			

a. Dependent Variable: Wet Tissue Weight

b. Predictors: (Constant), Volume, Live Weight, Width, Height, Length

The question is which among the predictors really stood out when it comes to estimating the dependent variable. That was answered by the values in the Unstandardized Regression Coefficients. The B-value represents the "unit change" of the independent variables relative to the dependent variable.

Table 5: Regression Coefficients

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	-10.689	1.384		-7.720	.000
	Length	.109	.064	.270	1.713	.090
	Width	.122	.037	.339	3.316	.001
	Height	.026	.069	.037	.367	.714
	Live Weight	.008	.009	.057	.909	.365
	Volume	.045	.020	.268	2.267	.026

a) Dependent Variable: Wet Tissue Weight

Based on the B coefficients table above and the Sig. values, only Shell Width (B=0.122; p=0.001) and Shell Volume (B=0.045, p=0.026) can be considered as a significant predictor of the Edible Wet Tissue weight. But since this study must also incorporate the effects of the other predictors, the following formula for estimating the edible wet tissue weight can be established. Again, these unstandardized B coefficients tells us about how the predictor variables change to estimate the dependent variable. Thus, based on the B coefficients above estimating the wet edible tissue weight can be estimated using the following linear regression formula.

Estimated Wet Tissue

Weight= -10.689 (constant) + (0.109 x SL) + (0.122 x SW) + (0.026 x SH) + (0.008 x LW) + (0.045 x SV)

This formula can now be used to be able to assess the growth of adult clams after a period of time without sacrificing them. There are also conditioning programs where clams are loaded with high-quality feeds and microalgal food. For a short period of time, there will be little increase in the shell and volume measurements but using the estimated weight of the wet edible tissue prior to the program, we can check if there is a significant increase in the weight of the edible tissue after comparing the estimated value with the actual value. Again, for experimental purposes it will be possible to study short-term feeding programs by focusing on the edible wet tissue weight alone even if there is no significant increase in the shell measurements, live weight, and shell volume.

Discussion

In any particular environment, the growth of an organism can be a direct determinant of fitness. There is what we can call as "absolute growth" in bivalves where we can attribute growth directly with the age of the organism- a condition where

cumulative biomass increase is expected through time. The percentage increase in biomass per unit time is "relative growth" (Seed, 1976) [33]. Utilizing clam shell measurements only for measuring growth is obviously insufficient because soft tissue growth is also happening inside it. Soft tissue growth is also determined by a variety of factors that may not be similar to the factors contributing to shell growth. The soft organs/tissues also carry out the living processes of the animal, not the shell. Allometric relationships between shell measurements (including body weight) and soft tissue biomass are often developed in order to non-destructively estimate the latter on living bivalves (Dame, 1996). Shell growth is a primarily determined by calcium availability in the water. Other factors include the amount of water pump across the bivalve tissues, pH, metabolism, and temperature. Shell size can only be an indirect estimator of wet tissue weight, because it is determined by other factors that may not be useful in increasing soft-tissue weight. A few findings in literature point to various environmental factors that are known to determine shell morphology and relative proportions of bivalve species, such as latitude (Beukema and Meeha, 1985) [3], depth (Claxton *et al*., 1998) [5], currents (Fuiman *et al*., 1999) [12], water turbulence (Hinch and Bailey, 1988) [17], wave exposure, type of bottom and type of sediment (Claxton *et al*., 1998) [5]. Burrowing behaviour (ability and efficiency) also influence the relative growth of some bivalve species (Seed, 1980) [34].

Aside from studies on morphometric aspects of bivalve populations from the Indian coast (Parulekar *et al*., 1986) [25], information on morphometry of *P. erosa* is not well-established. Worldwide, some morphological aspects of this species from Hong Kong mangroves were reported by Morton (1976) and from Australian mangroves by Gimmin *et al*. (2004) [14]. Food supply composition of the environment have differential effects on the biochemistry of soft tissues in different clam species (Gabbott, 1976; Baker and Hornbach, 1999) [13, 1]. Some clams also store energy in various ways biochemically. How they allocate these to produce shell or soft tissues can also be dependent on what is needed by the organism at any given moment or condition. For example, when need arises, they can shift from shell growth to soft tissue growth or vice versa (Lewis and Cerrato, 1997; Eversole, 2001) [18, 8]. In mussels, quahogs and scallops, shell growth can be measured directly and rapidly in transplanted juveniles (Evgenidou and Valiela, 2002; Shriver *et al*., 2002; Weiss *et al*., 2002) [9, 35, 37]. Indirectly, estimation can be done using established models such as the von Bertalanffy growth model.

Changes in food supply can also upset overall bivalve growth (Lewis and Cerrato, 1997) [18]. This suggest that shell and wet tissue formation respond differently to changes in food quantity and quality. Since bivalves can reallocate energy to support different types of growth under different conditions (Eversole, 2001) [8], soft-tissue growth changes also mean variations in physiological condition of clams. To be able to allow increased stocking of commercial bivalves, it is suggested to determine whether the growth of the soft tissue portion of clams is via a different biochemical pathway than the growth of the non-edible shell. One observable effect of high population density on the shape of benthic suspension feeders is found in exposed mounds formed by high-density barnacle populations (Bertness *et al*. 1998) [2]. Crowding is usually seen as the presence of physical interference among population members. Experiments on population density in some infaunal bivalves have shown no evidence of physical interference (Peterson, 1982) [26]. This is also why there are noticeable change in shape after food depletion in dense bivalve beds or when food is regulated artificially (Wildish and Kristmanson, 1984, Fréchette and Bourget, 1985a, b) [38, 10, 11]. Thus, the effect of high population density on mussel shape (e.g. Seed 1968) could be influenced by food regulation, physical interference and their interactions.

The results of this study are not only limited to *P. erosa* as a species. Proper management of commercial clams in general requires that growth is assessed accurately. This will facilitate successful interventions for quality control and stock enhancement. An increase in size is always associated with growth, but if aquaculturists rely only on the external morphometric measurements, it will remain unreliable unless backed up by statistical evidence. Based on the descriptive data, the mean wet tissue weight is only 9% of the live weight indicating that there is a large difference between shell mass and wet tissue mass. Those who reported this lower soft tissue weight of *Polymesoda erosa* hypothesized that it was due to desiccation, salinity fluctuations, and low pH as seen in the shape of the shells that looked eroded by acidity (Morton, 1988). Another reason for the decreased soft tissue weight is that the growing shell makes the clam heavier. We can always attribute this to increased shell mass or the ability to store water (Currey, 1988) [6]. This focused utilization of energy on the shell limits the growth of the organism because of the need for strong shells in adverse environments. It is therefore unclear whether growth of the soft tissues inside will eventually cope up even after shell growth already stopped. The organisms used in this study were also taken from the wild. It would be an interesting line of inquiry if the shell-to-wet-tissue weight ratio can be improved if these animals are cultured in the lab or in an intensive culture system where those so-called environmental factors or stresses no longer exist.

The strong correlation between shell variables in *P. erosa* is similar to the previous findings in other bivalves, such as *Mercenaria* (Hibbert 1977) [16], *Mytilus edulis* (Rodhouse *et al*. 1984) [29], *Dreissena polymorpha* and *D. burgensis* (Ross and Lima 1994) [30], and *Chamelea gallina* (Deval 2001) [7]. It was also noted that shell variables fail to estimate soft tissue weight due to a variety of reasons. It can be the reproductive state of the animal (Gimin *et al*., 2004; Rueda and Urban, 1998) [31], population density (Seed, 1968) [33], and physico-chemical variables of the habitat (Thorarinsdittir and Johannesson, 1996) [36]. The estimation technique conceptualized by this study is more relevant in experiments done in a controlled environment such as a laboratory or a mariculture facility. Since it was strongly established in literature that shell variables, volume and live weights has a strong link with the edible wet tissue weight, the use of regression analysis should come almost natural to every aquaculturists when it comes to a specific clam species that is being studied or managed. Measurements can change between different batches of clams but it is only a matter of sampling and some adjustments in the computations to provide a batch-specific formula for assessment. The procedure can be standardized and statistical software always comes in handy.

Conclusion and Recommendation

This study was able to verify strong correlations between Shell Dimensions (length, width, and height), Shell Volume, Live Weight, and Wet Edible Tissue Weight of *Polymesoda*

erosa. The various morphometric measurements can be used as predictors of edible wet tissue weight, but only Shell Width and Shell Volume has a significant contribution if we combine all their effects in a Multiple Regression Model. This simple method used to derive a formula for estimating the edible wet tissue weight can be applied to all commercial clams in general. One potential application of this estimation technique is in feeding experiments. Clams can be fed for a period of time and their estimated wet-tissue-weight is compared with the actual wet-tissue-weight to be able to assess the possible impact of a feeding treatment. It is also felt that estimated measurements are more robust in controlled conditions that in the wild thus the results of this study are really relevant to individuals who are into clam aquaculture.

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