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Proximate composition with dimensional relationships in *Meretrix casta* and *Villorita cyprinoides* from Neendakara part of Ashtamudi lake, Kerala, India

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ABSTRACT

Besides the augmentation of palatable value, relationship between shell characteristics and proximate composition of bivalves are significant to understand the ecological productivity of a system. Dimensional studies have great significance in fishery researches, for the improvement of current production and management. During the study period (July 2021 to May 2022), a total of 300 individuals from each clams, *M. casta* and *V. cyprinoides* were collected bimonthly from Neendakara region of Ashtamudi Lake, a Ramsar site from Kerala. The allometrical relation and the biochemical composition of bivalves were determined. The negative allometric growth showed by the length to width and thickness relationship implies that the increase in the width and thickness of the bivalves were inferior to the growth in length. Linear regression results showed that there is significant variation (p<0.05) in the allometrical relationship in the two bivalves. Various biochemical constituents such as, ash, moisture, carbohydrate, protein and fat have been also estimated. Single Factor ANOVA studies of bimonthly data reveals that, there was no significant differences (p>0.05) in the proximate compositions of the two clams. Condition Index (CI) and Edibility Percentage or Percentage Edibility (EP) were also estimated. Condition Index was recorded to be <1 during the entire study period. The results showed that the studied bivalve species could be considered as a good dietary resource, due to its high protein content as well as remarkable composition of carbohydrate and fat contents. Therefore, the present study will be helpful for implementing various sustainable management practices to increase the culturing potential of the bivalve in the estuarine beds.

Key words: Proximate analyses, Allometry, Percentage Edibility, Condition Index, Meretrix casta, Villorita cyprinoides.

Introduction

Marine food comprises various forms of aquatic biota. Among them, bivalves are known for a highly demanding proteinaceous food. India bangs second, position in capture fisheries among the Asian countries. The country has an annual production of 84,483 tonnes of bivalves and the major share is contributed from Kerala (75.8%) among the south-west coast of India. Ashtamudi Lake, a Ramsar wetland (no.1204) contributed a significant portion of these resources.

Estimation of shell dimensions for growth, biomass and total meat production are considered as a simple and non-destructive methods (Bailey and Green, 1988; Rodhouse *et al.*, 1984). Allometric studies reflects the relationship between the body parts of an organism (Reiss, 1989). It deals with the study about size and shape. Allometrical relationship act as a taxonomic aid, helps to recruit and estimation of

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stock (Spiers, 1952). It provide information for managing resources and understanding the surrounding variations (Palmer, 1990). The variation in the allometric parameters reveals the physiological status in bivalves. The physiological condition and the variation in environmental parameters can alter the meat composition of bivalves (Wilbur and Owen 1964). Various proximate factors can influenced by the allometric values. In India allometric studies on clams have been studied in several works (Algarswami and Chellam(1977); Rao (1988) and Appukuttan (1993).

As clams are one of the versatile dietary for humans, it is often a delicacy to humans. Many ecophysiological and aquaculture studies have depended on the proximate biochemical analysis (Giese, 1969; Ansell et al., 1980; Davis and Wilson, 1983). Biochemical constituents of bivalves shows variations depending on factors viz. water temperature, nutrient availability, reproductive cycle etc. (Okumus and Stirling, 1998; Orban, Di Lena et al., 2002). Shellfishes are good source of rich proteins, dietary essential aminoacids, low fat diet, and lower proportions of saturated fat, the presence of good lipids, significant amounts of omega-3-fatty acids, dietary essential amino acids, vitamin B12 and several important minerals such as iron, zinc and copper which helps to maintain the growth of the human body (Krzynowek et al., 1989; Dong, 2001; Trung tamTin hoc- Bo Thuy San, 2007; George and Gopakumar, 1995; Dincer, 2006; King et al., 1990). Various factors affecting the proximate composition of shellfishes such as spawning season (Durve, 1964), fecundity (Durve, 1964; Litaay and De Silva, 2003), depth of culture area (Ngo et al., 2006) have also been investigated (Rivonker and Parulekar, 1995).

The biological and chemical aspects, shell and meat contents, typical taste and flavor reflects the nature and quality of clams. Bivalves have nutritional, commercial, health attributes and ecological significance (Orban et al., 2002; 2006). Besides the edibility, bivalves are highly worthfull biotic component of an ecosystem due to its high conversion efficiency and rich biochemical constituents. And also, comparitively, very limited works have been done in the allometrical studies on bivalves, especially in Ashtamudi Lake. This work provide information about the nutritional qualities of the two commercially valuable bivalve species (M.casta and V.cyprinoides) of Neendakara region of Ashtamudi. Therefore, the present work has a significant practical application on human health perspective based on the shell fish consumption as well as a marketing food on bivalves cultures.

Materials and Methods

Study area

For the present study, Neendakara barmouth (Fig. 1) (8–56'09''N & 76–32' 45''E) of the Ashtamudi lake was selected as the sampling station. This area is characterized by inorganic pollution, Fishing Harbour, large scale mechanized boats/ trawler traffic, oil spillage, mining in far areas etc

Sample collection and preparation

A total of 100 specimens of *Meretrix casta* and 198 specimens of *Villorita cyprinoides* were collected bi-

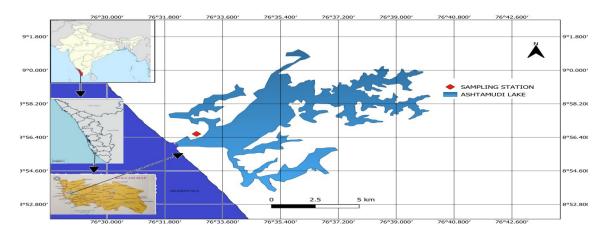


Fig. 1. Ashtamudi Lake

monthly from the sampling station, Neendakara of the Ashtamudi Lake (Fig. 1) during the study period from July 2021 to May 2022. The study site was surveyed and bivalves were collected during low tides by random free hand collection. The live samples were brought to the laboratory, cleaned to remove attached particles and kept alive for depuration. in aerated water for a day. Thereafter, the specimens were sorted out for allometrical and biochemical analyses.

Allometric analysis

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Individual bivalve specimens were measured for its whole weight (mg), shell length (mm), shell width (mm) and shell thickness (mm). Length, width and thickness were measured with the Vernier Caliper (mm) and whole body weight was measured with an electric balance (accuracy 0.01g). The allometrical parameters were used to compare the body measurements of the two populations of the stations as well as used to determine the relation with the proximate composition of the clams. Allometrical relationships were studied by testing each pair of variables potential allometric equation;

Y = a + Xb, where Y is the dependent variable (mm); X is the independent variable(mm); a is the intercept of the regression line and b is the slope regression co-efficient.

Percentage Edibility or Edibility percentage and Condition Indices were determined as per Mohite *et al.*, 2008; Okumus and Stirling (1998) respectively:

Percentage Edibilty(EP) =
$$\frac{\text{wet weight}}{\text{Total weight}} \times 100$$

Condition Index (CI) = $\frac{\text{wet weight}}{\text{Wet weight}} \times 100$

Shell weight

Proximate composition analyzes of the samples were done in triplicate for Ash, moisture, carbohydrate, protein and total fat lipid contents. Moisture was determined by oven drying at 105°C (3hrs.) to constant weight (AOAC, 2005). Samples were dried in an oven until constant weight obtained. Then cooled in dessicator and weighed to get the differential moisture content between fresh and dry weights. Ash content was estimated by heating the dried samples (550 °C overnight) in a muffle furnace and the percentage of ash was estimated by deducting the weight of the ash from the initial weight. Carbohydrate was analysed by the method of DuBois (1956). The crude protein was determined by the Kjeldahl method (AOAC, 2005). Total fat was extracted in Soxhlet apparatus using petroleum ether (AOAC, 2005).

Results

Allometry

Minimum, maximum, mean and standard deviation of shell measurements such as shell length (SL), shell width (SW), shell thickness (ST), Condition Index (CI) and Percentage Edibility (EP) were calculated (Table 1). The results showed linear models for length-width and length- thickness relationships (Graph1a to d). Linear regression analysis showed that there is a significant variation (p<0.05) observed in the allometrical relationship in the two bivalves. Table 2 showed the linear regression results for the relationships between the morphometric parameters.

Percentage edibility (%) showed an increasing trend during monsoon season and highest values were recorded in July and September for *M. casta* (26.37) and *V. cyprinoides* (24.02) respectively. Lowest values were recorded in May and it was 11.85 in *M.casta* and 9.54 in *V. cyprinoides*. Maximum value of Condition Index % (CI) were recorded during November (81.52) for *M. casta* and during October (74.23) for *V. cyprinoides*. Lowest condition index

Table 1. Mean, standard deviation (S.D), minimum, maximum, Percentage edibility and Condition Index values of M.*casta* and V. cyprinoides of Neendakara during the study period from July 2021 to May 2022.

| Measurement parameters | M. casta | Minimum (mm) | Maximum (mm) | V. cyprinoides | Minimum (mm) | Maximum (mm) |
|---|-------------------------|-----------------|-----------------|-----------------------|-----------------|-----------------|
| Shell length (mm) | 27.47±3.51 | 21.64 | 43.91 | 24.34±3.16 | 20.89 | 42.49 |
| Shell width (mm) | 37.96±2.94 | 19.65 | 41.16 | 27.77±3.33 | 16.15 | 36.03 |
| Shell thickness (mm) | 17.47±3.25 | 13.98 | 23.15 | 18.49 ± 2.43 | 12.31 | 34.17 |
| Percentage edibility (%) Condition Index | 21.067±5.01 78.5±7.1 | 11.85 19.42 | 26.37 81.52 | 20.07±4.03 58±6.92 | 9.54 16.93 | 24.02 74.23 |

were observed during May in *M. casta* (19.42) and in *V. cyprinoides* (16.93) (Table 1).

Proximate Analysis

Bimonthly analysis reveals that ash content ranged from 5.00-8.30 in *M. casta* and 5.80-8.05 in *V. cyprinoides*. Amount of Ash content indicates the presence of inorganic compounds in the tissues. Ash content was found to be lowest during the month of July and highest during March (*M. casta*) and during November (*V. cyprinoides*). *M. casta* and *V. cyprinoides* showed a bimonthly mean of 7.05±1.297 and 6.9±1.670 respectively (Graph 2 and 3).

The bivalves reveals high amount of moisture followed by protein, ash, carbohydrate and fat. The main constituents of the two bivalve species was the moisture, which indicates its freshness. Moisture content was ranged from 77.4%-90% in *M. casta* and 73%-92% in *V. cyprinoides*. Moisture content was found to be lowest in the month of May and highest during September (*M.casta*) and during July (*V. cyprinoides*). *M. casta* and *V. cyprinoides* showed a bimonthly mean of 84±0.058 and 84±0.072 respectively (Graph 2 and 3).

Carbohydrate content was ranged from 3.8-6.8 in *M. casta* and 3.3-6.9 in *V. cyprinoides*. Carbohydrate content was found to be lowest in the month of March (*M. casta*) and September (*V. cyprinoides*) and highest during May. *M. casta* and *V. cyprinoides*

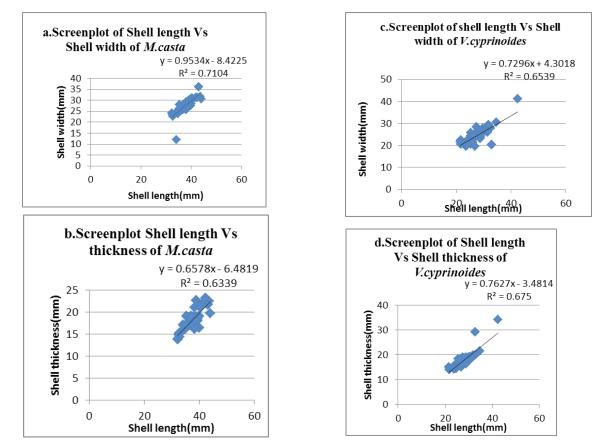
Protein content was ranged from 9.7-10.6 in *M. casta* and 8.9-10.4 in *V. cyprinoides*. Protein content was found to be lowest in the month of March and highest during January (*M.casta*) and during September (*V. cyprinoides*). *M. casta* and *V. cyprinoides* showed a bimonthly mean of 10.033 ± 0.372 and 9.80 ± 0.521 respectively (Graph 2 and 3).

Total fat content was ranged from 0.98-1.9 in *M. casta* and 0.98-1.7 in *V. cyprinoides*. *M. casta* and *V. cyprinoides* showed a bimonthly mean of 1.496±0.456 and 1.43±0.402 respectively (Graph 2 and 3).

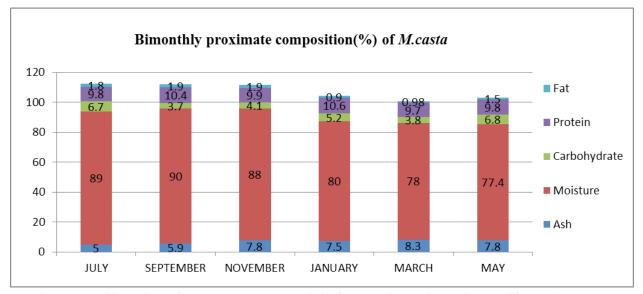
The biochemical constituents of the studied bivalves exhibited seasonal variations in Moisture, Ash, Protein, Carbohydrate and Lipid. Sesaonal analysis revealed that ash content in both M. casta and V. cyprinoides was found to be lowest during monsoon and highest during pre-monsoon. M. casta and V. cyprinoides showed a seasonal mean variation of 7.05±0.769 and 6.9±1.125 respectively. Moisture content in both M. casta and V.cyprinoides was found to be lowest during pre-monsoon and highest during post-monsoon. M. casta and V. cyprinoides showed a seasonal mean variation of 84±5.02 and 84±4.00 respectively. Carbohydrate content in both M. casta and V. cyprinoides was found to be lowest during post-monsoon and highest during monsoon. M. casta and V. cyprinoides showed a seasonal

Table 2. Linear regression results for the relationships between the morphometric parameters such as, Shell length (SL),shell width (SW), shell thickness (ST) collected from Neendakara, Ashtamudi lake during the study periodfrom July 2021 to May 2022.

| | Meretrix casta | Villorita cyprinoides | |
|--|-----------------------|-----------------------|--|
| Shell length- Shell width | a=4.301 | a=8.422 | |
| 0 | b=0.729 | b=0.953 | |
| | R ² =0.653 | $R^2=0.710$ | |
| | SE=1.047 | SE=0.0614 | |
| | P<0.05 | P<0.05 | |
| | t=4.10 | t=4.04 | |
| | 95% confidence | 95% confidence | |
| | Lower=0.0174 | Lower=0.2649 | |
| | Upper=0.0718 | Upper=0.2894 | |
| Shell length- Shell thickness | a=3.481 | a= -6481 | |
| J. J | b=0.762 | b=0.657 | |
| | $R^2=0.675$ | R ² =0.633 | |
| | SE=1.044 | SE=0.0504 | |
| | P<0.05 | P<0.05 | |
| | t=20.22 | t=4.04 | |
| | 95% confidence | 95% confidence | |
| | Lower= -0.4092 | Lower=0.1868 | |
| | Upper= -0.2751 | Upper=0.2158 | |



Graph 1. (a-d). Regression analysis of various allometrical parameters of the bivalves, M. casta and V. cyprinoides.



Graph 2. Bimonthly analysis of proximate composition (%) of *M.casta* during the study period from July 2021 to May 2022.

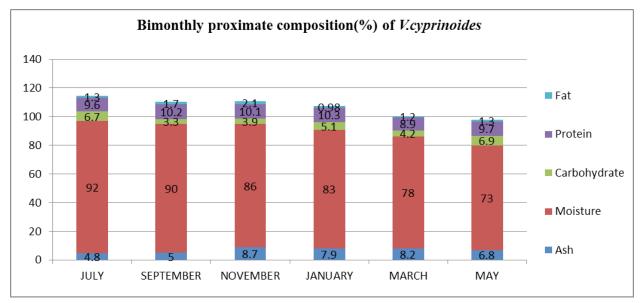
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mean variation of 5.05 ± 1.502 and 5.01 ± 1.63 respectively. Protein content in both *M. casta* and *V.cyprinoides* was found to be lowest during monsoon (*M. casta*) and pre-monsoon (*V. cyprinoides*) and highest during pre-monsoon and post-monsoon in *M. casta* and during post-monsoon in *V. cyprinoides*. *M. casta* and *V. cyprinoides* showed a seasonal mean variation of 10.033 ± 0.372 and 9.75 ± 0.217 respectively. Total fat content in both *M. casta* and *V. cyprinoides* was found to be lowest during pre-monsoon and highest during post-monsoon. *M. casta* and *V. cyprinoides* was found to be lowest during pre-monsoon and highest during post-monsoon. *M. casta* and *V. cyprinoides* showed a seasonal mean variation of 1.49 ± 0.49 and 1.43 ± 0.420 respectively (Table 3 and

4). Analysis of variance (ANOVA) was used to evaluate the proximal composition (ash, moisture, carbohydrate, protein and fat) data and there is no significant differences (p > 0.5) were observed among the two bivalve species.

Discussion

M. casta (Chemnitz) a Venerid edible clam, act as a part of sustenance level fishery in both east and west coast of India. This species contributes to a large part of livelihood along the Ashtamudi estuary (Laxmilatha *et al.*, 2006 a, b). The abundance of



Graph 3. Bimonthly analysis of proximate composition (%) of *V.cyprinoides* during the study period from July 2021 to May 2022.

| Table 3. Seasonal analysis o | f proximate comp | position (%) of <i>M.casta</i> | during the study | v period from Jul | v 2021 to May 2022. |
|------------------------------|------------------|--------------------------------|------------------|-------------------|---------------------|
| | | | | | |

| | | M. casta | | | |
|--------------|------|----------|--------------|---------|------|
| | Ash | Moisture | Carbohydrate | Protein | Fat |
| Pre-monsoon | 7.9 | 79 | 4.5 | 10.15 | 0.94 |
| Monsoon | 6.4 | 83.20 | 6.75 | 9.8 | 1.65 |
| Post-monsoon | 6.85 | 89 | 3.9 | 10.15 | 1.9 |

 Table 4. Seasonal analysis of proximate composition (%) of *V.cyprinoides* during the study period from July 2021 to May 2022.

| | V. cyprinoides | | | | |
|--------------|----------------|----------|--------------|---------|------|
| | Ash | Moisture | Carbohydrate | Protein | Fat |
| Pre-monsoon | 8.05 | 81 | 4.65 | 9.6 | 1.09 |
| Monsoon | 5.8 | 83 | 6.8 | 9.65 | 1.3 |
| Post-monsoon | 6.85 | 88 | 3.6 | 10 | 1.9 |

M.casta is influenced by several environmental factors (Tanyaros and Tongnunui (2011). It has been listed as Vulnerable (A 1c, 1d) of International Union for Conservation of Nature (IUCN), due to the population decline in the estuarine beds, habitat quality and number of mature individuals (Ananda-Rao et al., 1998). Villorira cyprinoides (Gray, 1825), the black clam found in the fine sand, clay and silt regions of backwaters. It contributes 45,000 t to the annual landings of clams of the Kerala State (Narasimhan et al., 1993). Hydrological conditions and productivity of the estuary have an important role on the growth of the clams (Abraham, 1963; Seshappa, 1971). Various ecological factors are identified for their effect on bivalve shell shape: wave impact, trophic conditions, water depth, density etc.

The demand for proteinaceous food has been increasing globally and expected to rise in the near future. The molluscan meat, especially the meat of bivalves has been regarded as a promising source to meet this need. The meat of the bivalves are good sources for the provision of high quality protein with all the essential amino acids for the growth and maintenance of human body.

Studies about the cyclical changes in biochemical composition of tissue help to assess the nutritive status of an organisms. Accumulation of protien, carbohydrate and lipid will be high during the proliferation phase of gonads (George, 1980). Proximate analysis showed that bivalves are rich in protein, carbohydrate and moisture levels. Many research works have already reveals about the protein, carbohydrate and lipid contents of the bivalves (Asha *et al.*, 2014; Dupcic *et al.*, 2014, Jagadis, 2005; Babu *et al.*, 2012; Salaskar and Nayak, 2011 and Laxmilatha, 2009) and its relation with various environmental as well as gametogenic activities (Orban *et al.*, 2002; Smolders *et al.*, 2005).

The present study exhibited seasonal variation in the proximate composition. Moisture content of the bivalves reflects the onset of spawning period (Singh *et al.*, 2012) and showed a negative correlation with salinity conditions (Maqbool, 1993). In the present study also, moisture content showed its highest content during monsoon period. Moisture content is an indication of flesh value (Ojea *et al.*, 2004; Dridi *et al.*, 2007; Ovissipour *et al.*, 2013). As an indication of the amount of inorganic nutrients, ash content of the tissue has a determining role in the biochemical quality of the tissues. Ash content in both the tissue samples were highest during postmonsoon and lowest during pre-monsoon period.

During the present study, a remarkable protein levels were observed in *M. casta* and *V. cyprinoides*, reached to a mean value of 10.15 and 10.1 respectively. A gradual increase in the protein content of the bivalves were observed during post monsoon season (maturity period) and then a slight decreasing trend during monsoon (spawning period). Similar results were observed in various studies (Jayabal and Kalyani, 1986; Berthelin *et al.*, 2000). Many studies showed a positive correlation of protein content with reproductive state (Eble, 1969). In contrast, part wise protein content in some clams remains without any remarkable variation in relation with reproductive cycle (Easterson and Kandasamy, 1988).

Amount of protein and carbohydrate is an important aspect to assess the quality and texture of seafoods (Loje *et al.*, 2007). Elevation in the level of protein and carbohydrate were in the gametogenesis period and a decrease in spawning phase. While carbohydrate shows a decline trend during reproductive phase (Ojea *et al.*, 2004; Dridi *et al.*, 2006). In bivalves, carbohydrate contents shows a reduction during monsoon periods. This may be due to the exploitation of carbohydrate stores during conditions such as stress, low salinity, gametogenesis stage etc. (Eble, 1969). Gradual increase in the carbohydrate content could be happen and may be due to the development of gonads (Shettigar and Seetharamaiah, 2013)

Fat contents have various important biological functions such as energy reserves, structural constituent of membranes, bio-signalling. Lipid utilization starts when all carbohydrate resources are exhausted. Temperature is an important factor which augment the fat consumption by the clams for energy and shows a negative correlation with temperature. Several research reports reveals the same results (Dridi and Romdhana, 2017).

Several studies have demonstrated that the Edibility Percentage (EP) and Condition Index (CI) as the main tools for the commercialization of bivalves depending various endogeneous and exogeneous conditions (Gabott, 1975; Lagade *et al.*, 2015). Condition Index (CI) serve as an indication of physiological status. During the present study, CI and EP have peak values during July to November and then showed a rapid decline. It indicates the best time for consumption on nutritional aspects. The Percentage Edibility denotes the quality of meat and it undergoes seasonal variations. It may be due to the energy

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utilization for the meat production and gametogenesis during the unfavourable seasons (Biandolino *et al.*, 2008). During spawning period percentage edibility declined (Mohite, 2009; Venkataraman and Chari, 1951). Besides economically very cheap, bivalves remains always a nutritional source, which are highly significant to humans a balanced diet. Therefore, a seasonal exploration and a sustainable use of the bivalves are recommended for the demand of a nutritional food as well as exploitation of bivalves at juvenile state (low nutritional stage).

Conclusion and Recommendation

Since the bivalves have remarkable concentrations of the essential biochemical constituents such as ash, moisture, carbohydrate, protein and fat, these are highly recommended for a rich nutritional diet. Allometric parameters have greater influence on its biochemical constituents. A scientific and continuous monitoring system is needed for enhancing the potential of bivalve cultures to overcome the problem of unscientific exploitation on clam beds. Therefore, this study suggested for an immediate implementation of strict scientific guidelines for culturing and conservation against overexploitation of these bivalves.

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Conflict of Interest

All the authors hereby declare, there is no conflict of interest on this matter.

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