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THE PROXIMATE AND MINERAL COMPOSITIONS OF FRESHWATER MUSSEL *PARREYSIA CORRUGATA* (MULLAR, 1774) FROM TUNGA RIVER IN THE WESTERN GHATS, INDIA

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Abstract

The proximate and mineral compositions of the freshwater mussel *Parreysia corrugata* (Mullar, 1774) inhabiting the upper reaches of Tungabhadra river in the Western Ghats, India have been analysed. Samples were collected from the river Tunga, at monthly intervals from March 2009 to February 2010. The average glycogen, protein and lipid contents in tissue were 9.62 mg g^{-1}, 7.73% and 6.81% respectively. The concentrations of calcium, iron, manganese, sodium and potassium were 11.78, 6.33, 9.94, 3.33 and 4.56 mg g^{-1} respectively. Copper (0.06 to 0.16 mg g^{-1}), zinc (0.37 to 0.55 mg g^{-1}), magnesium (0.98 to 2.36 mg g^{-1}) and iron (0.03 to 0.08 mg g^{-1}) content showed minor variations. The multivariate statistical analysis of ambient environmental and biochemical variables in tissue of *P. corrugata* during the study period revealed a total of 5 components which accounted for 94.02% of total variance. The hierarchical cluster analysis using complete linkage showed 2 distinct groups of biochemical variables.

Key words: River Tungabhadra, Western Ghats, minerals, proximate composition, principal component analysis.

Introduction

Freshwater bivalves provide significant ecological benefits and recognized as a source of food for human beings and also for other domestic animals from all over the world. The mussels provide high quality protein with almost all the dietary essential amino acids for maintenance and growth of human body (King *et al*., 1990). The freshwater mussels are significant contributor to the total standing crop in benthic communities and are important in the cycling of calcium (Mann, 1964; Negus, 1966; Cameron *et al*. 1979). These organisms are responsible for great deal of recycling of metals retained in abiotic compartments of the ecosystem and consequently they constitute the main pathway for export of metals from the aquatic environments to the terrestrial through the food chain by which they reach human beings. The knowledge of biochemical composition of any edible organism is extremely important since the nutritive value is reflected in its biochemical contents. The biochemical composition in the molluscs has been mainly studied to assess the nutritive status and also to supplement information on reproductive biology. The overall change in the biochemical composition during an annual cycle has been correlated to the events of the gonadal cycles of organisms. A good amount of information is available on biochemical composition of bivalve molluscs from various parts of the world (Nagabhushanum and Lomte, 1971; Ravera *et al.*, 2003; Baby *et al*., 2010; Ersoy and Sereflisan, 2010). Glycogen is the primary energy store in bivalves (Naimo *et al*., 1998) and the relative amount of glycogen stored in bivalve tissue is considered as a good indicator of body condition (Naimo *et al*., 1998; Naimo and Monroe, 1999). Glycogen has been shown to change in response to environmental perturbations such as extreme of temperature, pollutants or starvation in bivalves (de Zwann and Wijsmann, 1976; Hummel *et al.*, 1989; Haag *et al.*, 1993). The freshwater bivalves are good sources of minerals (Byrne, 2000; Ravera *et al*., 2003; Wagner and Boman, 2004) and some of the species are excellent sources of trace and minor elements needed for the proper growth and development of human being and can also be used as high-nutrient supplementary feed for domestic animals, birds and fish culture (Baby *et al*., 2010). There is a dearth of information on the biochemical composition of freshwater mussels from India.

Materials and Methods

A total of 312 individuals of *Parreysia corrugata* were collected at monthly intervals from March 2009 to February 2010 from the river Tunga at Hariharpur, near Koppa (12°43'19.66" N; 76°59'24.61" E) in Chikkamagalur district of Karnataka state. During June-September 2009 mussels could not be collected due to river flooding by south-west monsoon. The soft tissue samples were separated from shells, dried at constant temperature of 60 ^{0}C for 2 days and powdered. Estimation of glycogen of mussel was carried out based on the anthrone reagent method (Seifter *et al*., 1950). Total protein content was estimated by Lowry's method (Lowry *et al*., 1951). The total fat was extracted using chloroform-methanol-water by Bligh and Dyer method (Bligh and Dyer, 1959). The ash content of mussel was determined gravimetrically using muffle furnace (550°C for 6-8 hr) (AOAC, 1990) and was analyzed for minerals by Atomic Absorption Spectrophotometer (GBC, 932 Plus, Australia). The mean values of glycogen, protein, fat, minerals (calcium, iron, manganese, sodium, potassium, copper, zinc, magnesium, chromium) and environmental variables such as air temperature, water temperature, pH, conductivity, DO, chloride, alkalinity and total hardness were subjected to Principal Component Analysis (PCA) and Cluster Analysis (CA) (Davis 1973; Lewis-Bek 1994) to understand the relationship between the biochemical composition and environmental variables.

Results

The total glycogen content varied from 1.81 (January) to 16.3 mg g^{-1} (April) (Figure 1a). The mean value was 9.62 mg g^{-1}. The values of glycogen content increased gradually from March to May. From October onwards the glycogen decreased gradually and reached the lowest level during January and thereafter it increased again. Total protein content varied from 6.12 (February 2010) to 8.51% (March) with a mean value of 7.73% (Figure 1b). The variations in protein content of tissue showed minor fluctuations throughout the study period except during January and February when low values were recorded. The temporal variability in the values of fat content of *P. corrugata* is presented in Figure 1c. The total fat content varied from 6.2 (October) to 7.8% (February) on dry weight basis. The mean value of fat was 6.81%. The values of fat content were higher during the post-monsoon season when compared to pre-monsoon season. The ash content varied from 11.45 (May) to 22.2% (January) (Figure 1d). The mean value was 15.66%. The value of ash content showed decreased trend from March to May. From October onwards the ash content increased with the highest peak during January and thereafter again decreased.

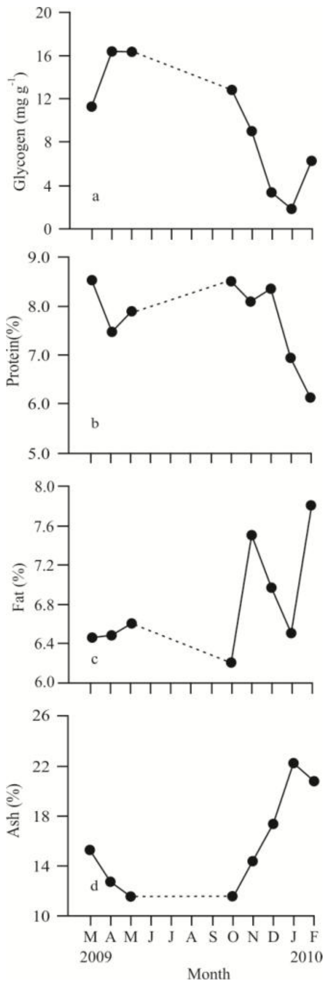

Figure 1: Monthly variability in glycogen (a), protein (b), fat (c) and ash (d) contents in *Parreysia corrugata*

The data on the concentration of micronutrients such as calcium, manganese, iron, potassium, sodium, magnesium, zinc, copper and chromium is presented in Table 1. The calcium values ranged between 9.76 (October) and 15.7 mg g^{-1} (January) with a mean value 11.78 mg g^{-1} of dried tissue. The calcium content was maximum during the post-monsoon season. The buildup of calcium was started during October onwards and reached the peak value during January. Thereafter the values showed decreasing trend. Manganese, iron, potassium and sodium contents in tissue were present in moderate quantity and the average values were 9.94, 6.33, 4.57 and 3.32 mg g^{-1} respectively. The manganese values varied from 6.38 (March) to 13.45 mg g^{-1} (November) with a mean value of 9.94 mg g^{-1}. The higher values of manganese were recorded during the post-monsoon season where as the lower values were recorded during pre-monsoon season. The iron content was highest (8.76 mg g^{-1}) in November 2009 and lowest (3.85 mg g^{-1}) in May 2009. The mean value of iron content was 6.33 mg g^{-1}. The low value of iron content was noticed during the pre-monsoon period and also during October. Thereafter the values were increased gradually and the peak was appeared during the post-monsoon season (Table 1). The potassium values in tissues were ranged between 3.11 (April) and 6.27 mg g^{-1} (November). The difference between the maximum and minimum values was 3.16 mg g^{-1}. The mean value of potassium was 4.56 mg g^{-1}. The potassium content was high during the post-monsoon season. The high value (4.38 mg g^{-1}) of sodium was noticed in November and the low value (2.69 mg g^{-1}) was in April. The difference between the minimum and maximum values of sodium content was 1.69 mg g^{-1}. The mean value was 3.32 mg g^{-1}. The values of sodium content in tissue were lower during March to May 2009, October 2009, January and February. The values of sodium were higher during November-December.

Table 1: Monthly variation in mineral content (mg g^{-1}) of *P. corrugata* from the study area

Minerals \ Month	Mar 2009	Apr	May	Oct	Nov	Dec	Jan 2010	Feb	Mean
Ca	12.86	9.88	9.8	9.76	10.94	12.56	15.7	12.77	11.78
Mn	6.38	6.84	9.71	7.13	13.45	12.16	13.19	10.67	9.94
Fe	4.92	4.4	3.85	4.58	8.76	7.81	8.71	7.63	6.33
K	3.22	3.11	4.54	5.44	6.27	5.46	4.37	4.13	4.57
Na	3.05	2.65	2.89	3.18	4.38	3.86	3.18	3.38	3.32
Mg	1.26	0.98	1.97	2.24	2.36	2.17	2.23	2.15	1.92
Zn	0.48	0.43	0.45	0.37	0.42	0.46	0.44	0.55	0.45
Cu	0.13	0.16	0.08	0.06	0.1	0.13	0.09	0.09	0.11
Cr	0.03	0.03	0.04	0.08	0.08	0.06	0.06	0.06	0.06

The concentration of magnesium, zinc, copper and chromium in the whole tissue of *P. corrugata* is presented in Table 1 and the average values were 1.92, 0.45, 0.11 and 0.05 mg g^{-1} respectively. The magnesium content was highest (2.36 mg g^{-1}) during November and lowest (0.98 mg g^{-1}) during April (Table 1). The higher values were recorded during the post-monsoon season and also during pre-monsoon season (May). The average value was 1.92 mg g^{-1}. The values of zinc fluctuated between 0.37 (October) and 0.55 mg g^{-1} (February). The average value of zinc was 0.45 mg g^{-1}. The values were more or less same throughout the study period. The maximum (0.16 mg g^{-1}) and minimum (0.06 mg g^{-1}) values of copper were recorded in April and October respectively (Table 1). The average value of copper during the study period was 0.11 mg g^{-1}. The values of copper showed two peaks during April and December. The chromium content in tissue was minimum (0.03 mg g^{-1}) in March and maximum (0.08 mg g^{-1}) in October and November. The average value of chromium was 0.05 mg g^{-1} during the study period. The chromium content was highest during the post-monsoon season than that of summer season (Table 1).

The application of Principal Component Analysis (PCA) has showed the possible correlation between the ambient environmental (air temperature, water temperature, pH, conductivity, dissolved solids, chloride, alkalinity, dissolved oxygen, total hardness, calcium and magnesium) and biochemical variables (glycogen, fat, protein, ash, calcium, magnesium, copper, iron, sodium, potassium, chromium, zinc, manganese) in tissue of *P. corrugata.* The eigen values and their contribution to the total variance indicated a small number of components that could adequately explain the observed correlations among the large numbers of observed variables. The plots of principal component axis 1 and axis 2 showed 4 different groupings of environmental and biochemical variables in tissues of *P. corrugata* (Figure 2).

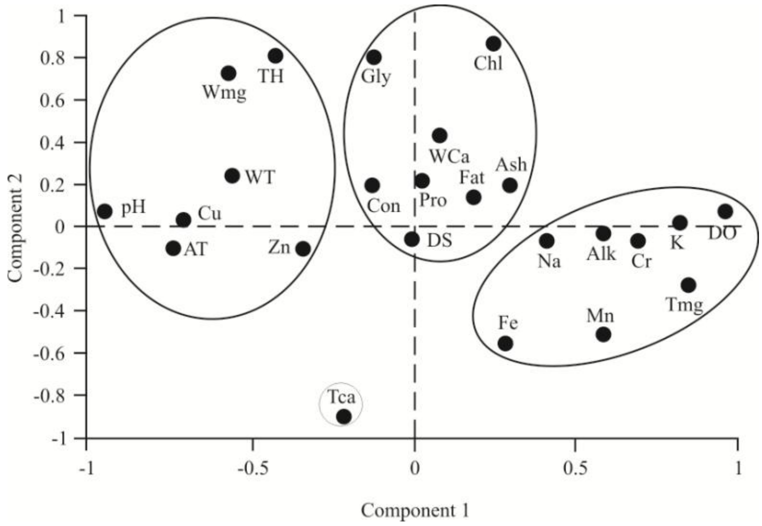

Figure 2: Principal component plots of component 1 and component 2 of ambient environmental and biochemical variables of *Parreysia corrugata.* Abbreviations: AT, air temperature; TH, total hardness; Chl, chloride; Ca, calcium; Mg, magnesium; WT, water temperature, DO, dissolved oxygen; Con, conductivity; DS, dissolved solids; Alk, alkalinity; Wca, water calcium, WMg, water magnesium; Gly, glycogen, Pro, protein; TCa, tissue calcium; TMg, tissue magnesium, Cu, copper; Cr, chromium, Mn, manganese; Na, sodium; K, potassium; Fe, iron; Zn, zinc.

The hierarchical cluster analysis using complete linkage showed two major groups of biochemical and environmental variables of *P. corrugata* (Figure 3).

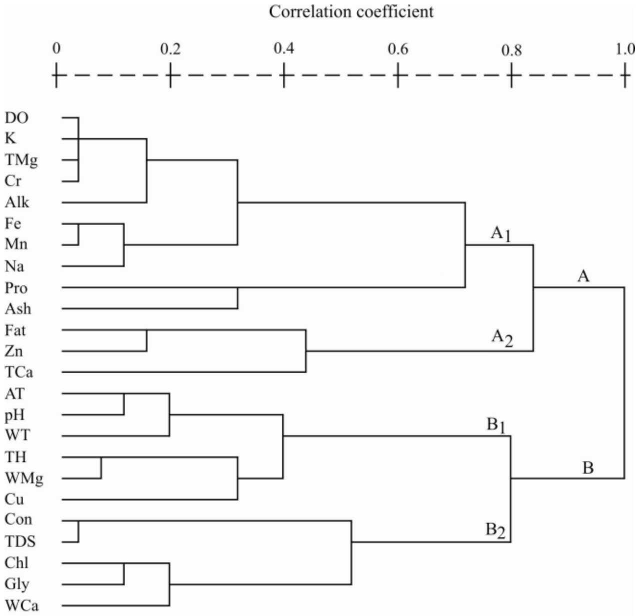

Figure 3: Dendrogram (complete linkage) of biochemical parameters of *Parreysia corrugata.* Abbreviations: AT, air temperature; WT, water temperature, DO, dissolved oxygen; Con, conductivity; TH, total hardness; Cl, chloride; Wca, water calcium, WMg, water magnesium; DS, dissolved solids; Alk, alkalinity; Gly, glycogen, Pro, protein; TCa, tissue calcium; TMg, tissue magnesium, Cu, copper; Cr, chromium, Mn, manganese; Na, sodium; K, potassium; Fe, iron; Zn, zinc.

Discussion

Measuring of macromolecules (carbohydrate, protein, lipid) revealed the biochemical function and energy storage in freshwater Unionids (Greseth *et al*., 2003). Glycogen, the primary energy reserve in bivalves, drives many important physiological processes and could be used to ensure short-term exposure to anoxia, emersion and reduced food supplies. The highest glycogen content in *P. corrugata* was 16.3 mg g^{-1} (April) and the lowest was 1.81 mg g^{-1} (January) during the present study period. The values of glycogen increased slowly from March to May due to build up of gonads. The fall in glycogen content from October onwards could be due to spawning activity. The gradual increased content of glycogen from March onwards could be due to the development of the gonads. An intimate association of glycogen with the period of sexual activity was also observed in *Corbicula* sp. (Greseth *et al*., 2003). Baby *et al.* (2010) observed that the glycogen content in *Lamellidens margianalis* was 4.94%. Nagabhushanam and Lomte (1971) in *P. corrugata* observed that the glycogen content varied from 4.57 to 5.73% of dry weight in various size groups. They further observed that the variation in the glycogen content was not size dependent. Histochemical preparation showed that maximum glycogen concentration was noticed in the mantle, muscles, gills, gonads and digestive diverticula of mussels (Nagabhushanam and Lomte, 1971). Significant reductions in Unioid glycogen greatly reduce their ability to cope with natural stressors present in the new environment. Glycogen storage fluctuates seasonally during ebb periods of gametogenisis and decrease rapidly in response to reduced food availability and environmental stress (Williams and McMohan, 1989; Patterson *et al*., 1997, 1999).

The average value of total protein content during the present study was 7.73% and the values ranged from 6.12 to 8.51%. The present observations were similar with the observations of earlier workers from India. Baby *et al.* (2010) reported that the protein content in *Lamellidens marginalis* was 6.46%. The total protein content in *P. corrugata* from Kham river near Aurangabad, India varied from 6.50 to 8.00% of dry weight (Nagabhushanam and Lomte, 1971). Further they reported that the protein content of smaller individuals was higher than that of the larger forms.

The mean value of total lipid was 6.81% during the present study period. The similar observations were made by Hori (1954) on *Corbicula* sp. and Nagabhushanam and Lomte (1971) on *P. corrugata* and in these organisms the fat content was considerably reduced during the spawning season. However in case of *Unio terminalis* (2.55%), *Potamida littoralis* (1.05%) and *Lamellidens marginalis* (0.507%) the fat content was very low (Ersoy and Sereflisan, 2010; Baby *et al*., 2010) when compared to the values of fat content in the present study. Nagabhushanam and Lomte (1971) observed that the total fat content in *P. corrugata* ranged from 3.30 to 4.60% of dry weight and accumulated in the gonads and the mantle. Bascinar (2003) observed that the lipid content of *Anodonta cygnea* was 6.39% in lake Cildir, Turkey. The fall of lipid in January and February in the present study could be due to the discharge of the gametes during the post-monsoon season.

The mean value of ash content in *P. corrugata* during the study period was 15.66%. In *L. marginalis* little ash was reported (Baby *et al*., 2010). The analysis of the minerals in whole tissue of *P. corrugata* showed that the mussels are good source of minerals. Calcium was the most prominent element detected (9.76-19.02 mg g^{-1}) in tissue of *P. corrugata.* The other elements such as manganese (6.38-13.45 mg g^{-1}), iron (3.85-8.76 mg g^{-1}), potassium (3.11-6.27 mg g^{-1}), sodium (2.69-4.38 mg g^{-1}), magnesium (0.98-2.36) and zinc (0.37- 0.69 mg g^{-1}) were also in higher concentrations. The similar observations have been reported in soft tissues of *Anodonta woodiana* (Liu *et al*., 2010) and *A. cygnea* and *Unio pictorum mancus* (Ravera *et al*., 2003). However in *Lamellidens marginalis* the concentration of these elements were less (Baby *et al*., 2010) when compared to the present study. Manganese, iron, and zinc are well-known essential elements for all living organisms (Mertz, 1981; Klassen, 2001). Manganese and iron acts as a cofactor for numerous enzymatic reactions (Goyer and Clarkson, 2001).

Principal component analysis applied for ambient environmental and biochemical variables of *P. corrugata* revealed a total of 6 components, which accounted for a total variance of 97.49%. The component 1 (AT, WT, DO, pH, Cu, TMg, K, Cr, Mn) accounted for as much variance as possible (39.19%) followed by component 2 (Cl, TH, WMg, Gly, Fe, TCa) accounted for 21.49% of variance, component 3 (Alk, Fe, Na) accounted for 13.03% of total variance. The component 4 (Con, DS, WCa) accounted for 11.39% of variance. The component 5 and 6 together accounted for 12.39% of total variance. The component 5 (Fat, Pro, Zn) accounted for 7.82% of total variance and component 6 (Ash) accounted for 4.57% total variance. The plots of principal component axis 1 and axis 2 showed 4 different groupings of environmental and biochemical variables in tissues of *P. corrugata* (Figure 2). Group 1 consisted of biochemical variables such as Fe, Mn, Mg, Na, K and Cr and ambient environmental variables such as alkalinity, dissolved oxygen. The variables dissolved oxygen and potassium showed positive correlation on both axes where as other variables showed positive correlation on component axis 1 and negative correlation on component axis 2. All these variables showed medium to high correlation on component axis 1 and low correlation on component axis 2. Group 2 included biological parameters such as (glycogen, protein, fat, ash) and environmental parameters such as chloride, calcium, conductivity and dissolved solids. The variables chloride, calcium, protein, fat and ash showed positive correlation on both component axes, conductivity and glycogen showed negative correlation on component axis 1 and positive correlation on axes 2 where as dissolved solids showed negative correlation on both component axes. All these variables showed low significant correlation on both axes except glycogen and chloride showed high significant correlation on component axis 2 and low correlation on component axis 1. Group 3 consists of biological variable (Cu, Zn) and environmental variable such as (air temperature, water temperature, pH, total hardness, magnesium). All the variables showed negative correlation on component axis 1 and positive correlation on component axis 2 except air temperature and zinc showed negative correlation on both component axes. The variables pH, air temperature, water temperature and copper showed high correlation on component axis 1 where as low correlation on axis 2. The environmental variables magnesium and total hardness showed medium correlation on component axis 1 and high correlation on axis 2. The mineral zinc showed medium correlation on component axis 1 and low correlation on component axis 2. Group 4 included only one biological parameter such as calcium and showed significant negative correlation on component axis 2 where as low negative correlation on component axis 1 (Figure 2). The hierarchical cluster analysis showed two major groups, group A and group B (Figure 3). Group A with 2 subgroups A_1 (DO, Alk, K, TMg, Cr, Fe, Na, Mn, protein and ash) and A_2 (Fat, Zn, TCa) and group B also with 2 subgroups B_1 and B_2. The subgroup B_1 included AT, WT, pH, TH, WMg and Cu. The subgroup B_2 consisted of conductivity, dissolved solids, chloride, calcium and glycogen.

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