Proc. Indian Acad. Sci. (Anim. Sci.), Vol. 97, No. 2, March 1988, pp. 151–160.

Seasonal changes in the level and content of different biochemical constituents in tropical cerithiids *Cerithidea* (*Cerithideopsilla*) *cingulata* (Gmelin 1790) and *Cerithium coralium* Kiener 1841

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MS received 16 May 1987; revised 4 February 1988

Abstract. The level and content of different biochemical constituents were investigated over a period of one year (January–December 1982) in foot, gonad digestive gland complex and viscera of two tropical cerithiids, inhabiting two different regions of backwaters. Their seasonal indices exhibited bimodal pattern of cycling and this trend was more predominant in gonad digestive gland complex and foot. In all the body components of both species, the biochemical content was found to show considerable changes when compared to their levels. In both the cerithiids, the gonad digestive gland depicted remarkable variations with the season, followed by foot. The viscera did not show much variation when compared to other body components. The carbohydrates and glycogen of both the species showed a primary peak in March and a secondary peak in September indicating maturity of gonads. This was followed by a decline in their constituents which might be due to spawning. Lipid utilisation was also observed in both the animals next to carbohydrates and glycogen. Proteins were least utilised in the process of reproduction. Thus a carbohydrate-oriented metabolism was recorded in both the species. The results were compared with the other molluscs.

Keywords. Biochemical level; biochemical content; *Cerithidea cingulata*; *Cerithium coralium*; biochemical constituents; carbohydrate-oriented metabolism.

1. Introduction

The two vital processes, reproduction and growth, of many invertebrates require energy which may be stored in reproductive or somatic body parts (Giese 1969). The existing literature (Giese 1959, 1966, 1969; Stickle 1975; Nagabhushanam and Mane 1975; Belisle and Stickle 1978; Deshpande and Nagabhushanam 1983) reveals a close relation between reproduction and changes in biochemical constituents stressing the importance to study the biochemical contents to arrive at a definite conclusion. Among molluscs, bivalves (Bayne 1976) have received much attention. Stickle (1975) and, Belisle and Stickle (1978) have made investigations on this aspect in the intertidal gastropods *Thais lamellosa* and *Thais haemastoma* respectively. Recently Uma Devi *et al* (1985) reported the changes in the composition of different biochemical constituents in an intertidal gastropod *Morula granulata*. Chitons have also been used for this study by Lawrence and Giese (1969), Himmelan (1978) and Deshpande and Nagabhushanam (1983). But no information was available on this topic in cerithiids. Therefore, in the present investigation, an attempt has been made to study the seasonal changes in the biochemical constituents of *Cerithidea cingulata* and *Cerithium coralium*. Both the level and content of different biochemical constituents have been studied and a comparison has also been made between the animals. It is also interesting that there is a difference in their distribution: *C. cingulata* lives in the middle reaches of the estuary whereas *C. coralium* occurs in the lower reaches of the estuary.

2. Materials and methods

Medium sized *C. cingulata* (shell length 27–31 mm) and *C. coralium* (shell length 23–28 mm) were collected from the backwaters of Bhimilipatnam which is 25 km from Visakhapatnam on the east coast of India. They were equilibrated to the laboratory conditions for about 24 h during which period the gut contents were cleared. Ten animals of each species were dissected and the different body components were separated. The same components of all the animals were pooled separately for further analysis. The body components of each species consist of foot, gonad digestive gland complex and viscera. As the gonad and digestive gland are inseparable, they are taken together as gonad digestive gland complex. Their wet weights were taken and dried in an air oven at 60°C. The dry tissue powder of each body component was prepared and it was stored in glass vials kept in desiccator. Collections were made for every month over a period of one year (January–December 1982). The different body component indices were calculated by using the method of Stickle (1975).

The dry tissue powder of each body component was analysed for different biochemical constituents viz., carbohydrates, glycogen, proteins and lipids. Anthrone method (Carrol *et al* 1956) was used for the estimation of carbohydrates and glycogen. Protein estimation was made by using Folin phenol reagent (Lowry *et al* 1951). The determination of lipids was carried out with chloroform : methanol (2 : 1) as suggested by Folch *et al* (1959). The total ninhydrin positive substances (TNPS) were estimated by the method of Moore and Stein (1954).

The different biochemical constituents are presented as percentage of dry weights. Biochemical content (total present per component of a 100 g standard animal) was calculated by using the method of Stickle (1975). ANOVA (Snedecor and Cochran 1967) was carried out to determine the statistical variation in different biochemical constituents. Further analysis of the data was done by Duncan's multiple range test (Snedecor and Cochran 1967) to determine whether the variations are significant.

Results

3.1 *Field observations*

The animals, *C. cingulata* and *C. coralium*, inhabit two different regions of the backwater system where the environmental conditions fluctuate with the season (Prabhakara Rao 1980). This backwater system is extensively shallow covering about 4·5 sq. km adjoining the coast. A small river, Gousthani, and 3 freshwater creeks empty into this backwater system. The backwater system is connected to the bay waters through a narrow entrance channel.

C. cingulata is found in the middle region of the backwater system. The salinity regime at this region oscillates between 6·81 and 23·22‰. There is considerable influx of sewage and land drainage. The dissolved oxygen levels vary from 10·22–2·04 ml O_2^{-1}. The temperature fluctuations in this region are: 28·5–34·8°C. The movement of water is very slow and practically this is void of wave action and presents a stagnant brackish condition. The organic matter is plentiful in this region. The substratum is composed of medium sized grains of sand (0·350–0·250 mm).

C. coralium lives near the lower region of the backwater. Here the conditions are almost marine with frequent disturbances. The salinity of the water varies between 20·23 and 34·02‰. *C. coralium* occurs in marine conditions and was never found to extend into the upper reaches of backwaters. The temperature in this region ranges from 25·5–34·8°C. The dissolved oxygen levels vary from 5·656–3·7 ml O_2^{-1}. The organic matter is comparatively lower in this region than in the habitat of *C. cingulata*. Coarse type sand grains (0·710–0·500 mm) are present in this region.

There are two spawning periods in these cerithiids: one occurring in June and July and another in December and January. During these periods, the animals were found to lay eggs in the form of ribbon on any substratum available for them.

3.2 *Body component indices*

All the different body component indices were found to be higher in *C. cingulata* than *C. coralium* (figure 1). However, both species exhibited a bimodal seasonal pattern in their indices with primary and secondary peaks. In foot, a primary peak value was observed in March and a secondary peak in September for *C. cingulata*. The highest

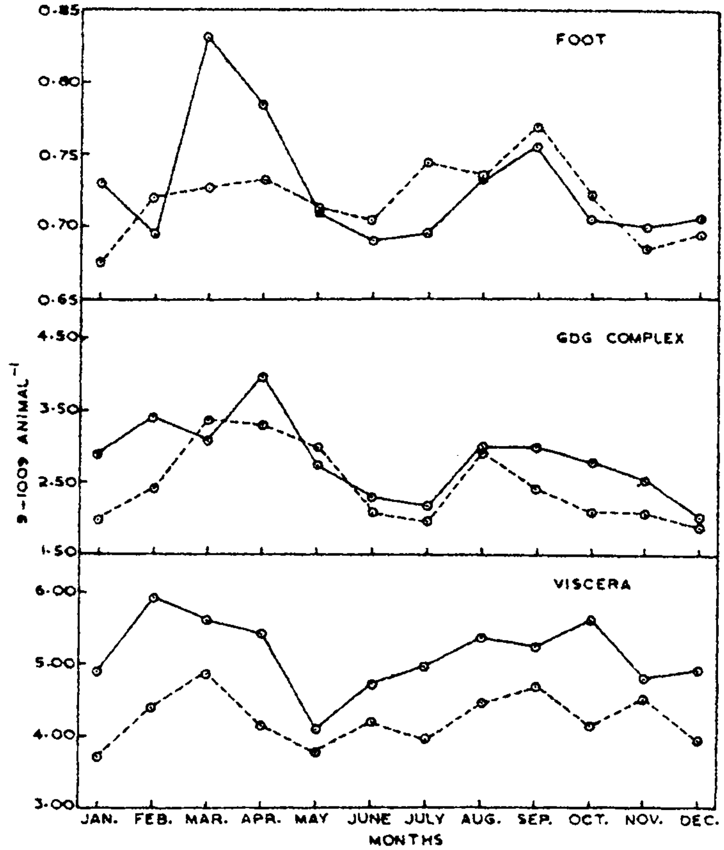

Figure 1. Seasonal changes in the body component indices of *C. cingulata* (———) and *C. coralium* (- - - -).

foot value was recorded in *C. coralium* in September. The gonad digestive gland complex index of *C. cingulata* was highest in April and a secondary peak was noticed in August. In *C. coralium*, the gonad digestive gland complex exhibited a peak value in March and another peak in August. The viscera of both species did not show any particular trend but highest value was observed in February for *C. cingulata* and in March for *C. coralium*.

3.3 *Seasonal changes in biochemical composition*

The changes in the levels of different biochemical constituents in *C. cingulata* and *C. coralium* are presented in figure 2 for foot, figure 3 for gonad digestive gland complex and figure 4 for viscera. It is clear from ANOVA that the changes (both

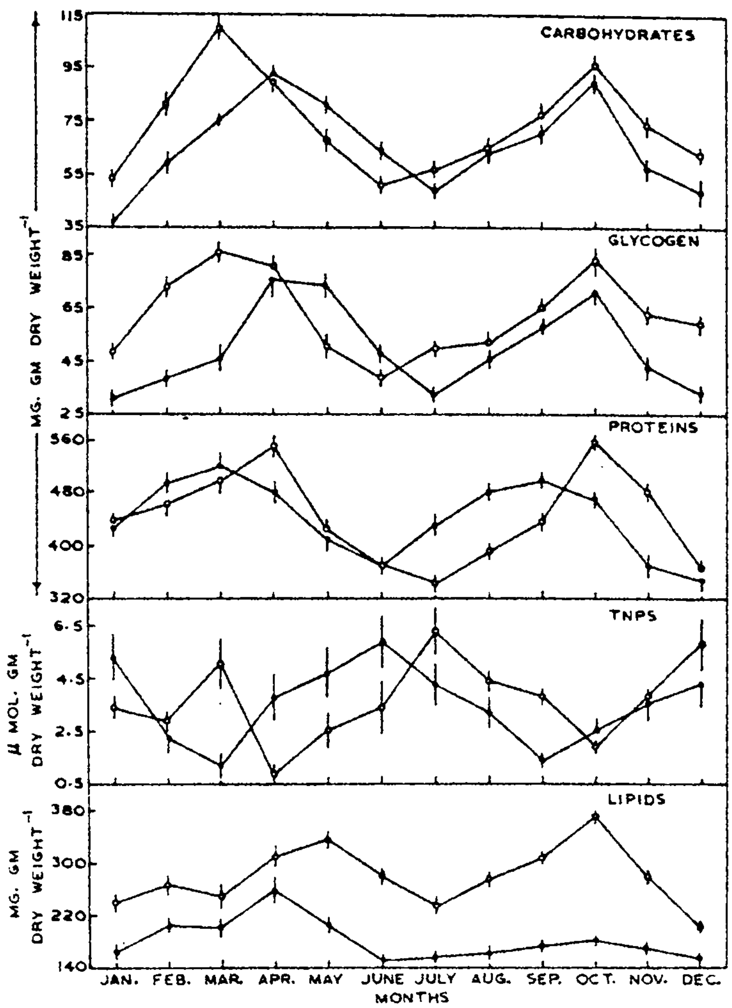

Figure 2. Seasonal changes in the levels of different biochemical constituents in foot of *C. cingulata* (○) and *C. coralium* (●).

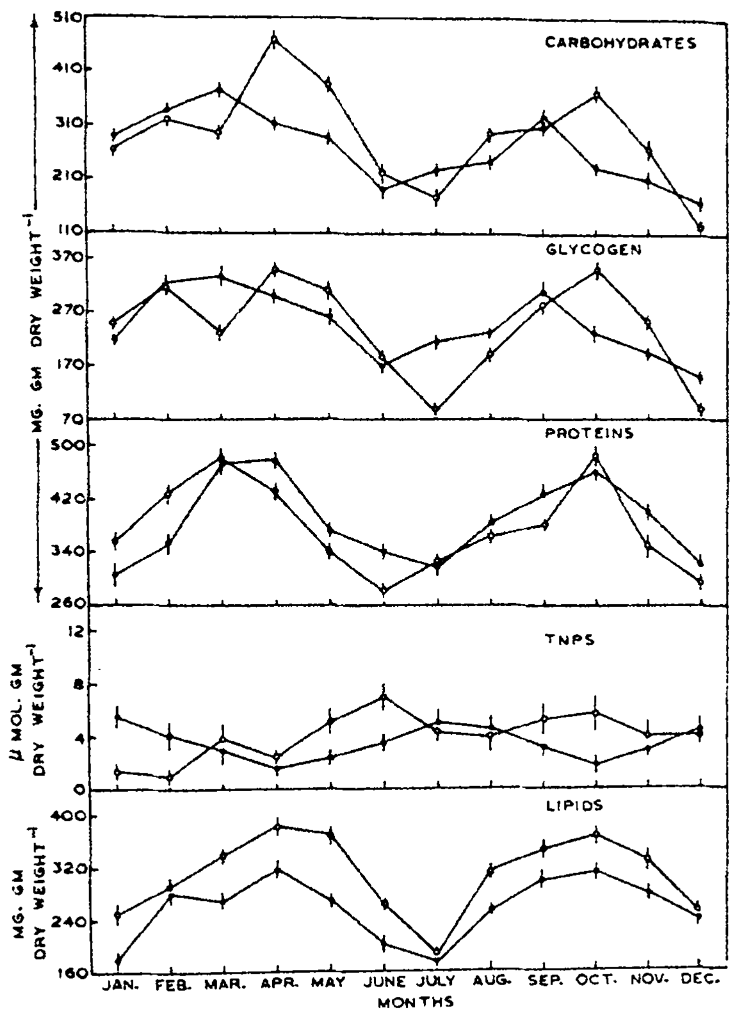

Figure 3. Seasonal changes in the levels of different biochemical constituents in gonad digestive gland complex of *C. cingulata* (○) and *C. corallum* (●).

level and content) in biochemical constituents for different body components i.e. foot, gonad digestive gland complex and viscera were found to be significantly different ($P < 0{\cdot}05$) for both the species.

4. Discussion

The present investigation demonstrates seasonal changes in different body component indices and also in the biochemical constituents (level and content) of both species. The bimodal pattern of indices observed in the present investigation correlates well with spawning period of the animals. During the months of February and March, the increase in the index values can be attributed to the addition of new cellular elements in the form of gonad maturation. The secondary maximum appeared

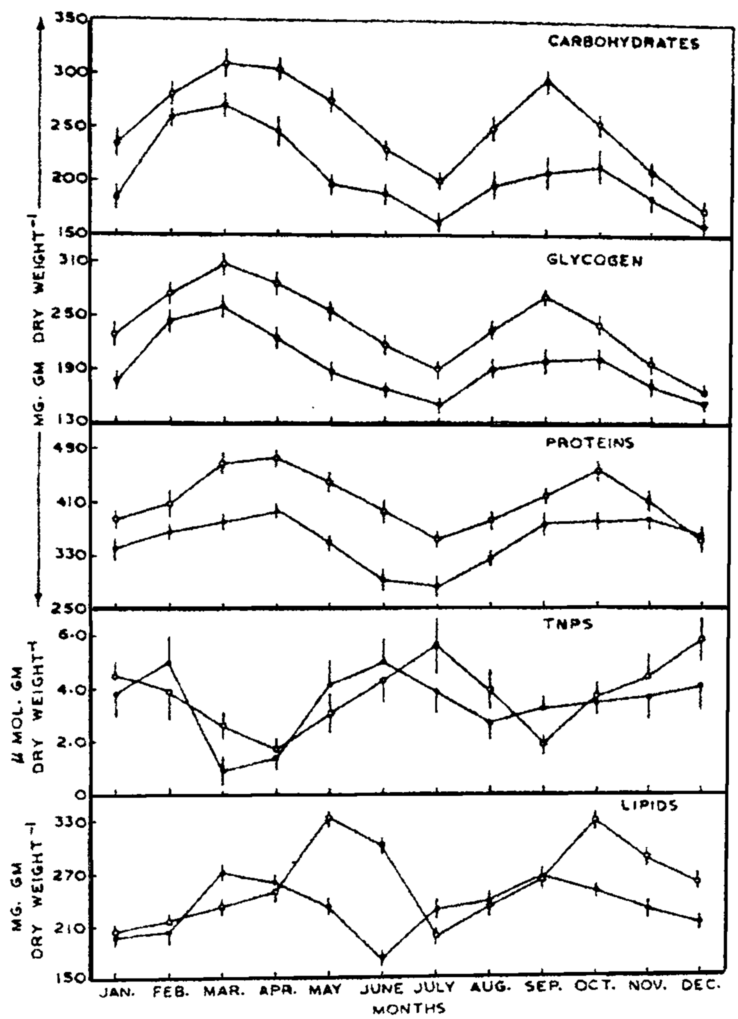

Figure 4. Seasonal changes in the levels of different biochemical constituents in viscera of *C. cingulata* (○) and *C. coralium* (●).

in the months of September and October which may denote the preparation period for the minor spawning season. Similar increase in gonad index during reproductive season was reported in several molluscs (Giese and Pearse 1974). Lawrence *et al* (1965) have noticed this type of cycling of gut and gonad indices in *Katherina tunicata* and *Cryptochiton stellari* respectively during reproductive phase. A rapid increase in the gonad index value was due to accumulation of ripe gamates which are to be released during spawning season (Giese 1969). The low values observed in July and December for both the species indicate the period of spawning. Nagabhushanam and Deshpande (1982) also reported seasonal variation of testis, gonad and digestive gland indices in *Chiton iatricus*. No cyclic pattern was noticed in the digestive gland of abalone *Haliotis cracheroidii* (Webber 1970). However, the higher indices observed in *C. cingulata* than *C. coralium* might be due to greater accumulation of gamates or to a thinner shell in *C. cingulata*.

A close observation of the results indicate that of all the 3 body components, major changes were observed in gonad digestive gland complex and to some extent in foot. According to Giese (1969) that there are no discrete biochemical storage organs in molluscs as occurred in vertebrates. Therefore, during the period of reproduction which requires a great amount of energy, gonad digestive gland complex contributes much and then followed by foot. In foot also, reserve food material may be stored for use but only a small amount of utilisable biochemical constituents are channeled out during reproductive period. It is clear from the results that the carbohydrates and glycogen played a major role during reproduction when compared to other biochemical constituents in both species. The changes observed in biochemical levels indicate their synthesis and utilisation during different periods, but their contents depict a clear picture of storage and utilisation (Stickle 1975). The content values observed here, thus, denote remarkable changes showing their accumulation and utilisation.

The biochemical constituents, mostly carbohydrates and glycogen, were high during February–April indicating the maturation of gonads followed by spawning in May–July in which months low values were recorded. These constituents again increase during prespawning period (August and September) and there is second maxima in October. Then, during November–January, there appeared a second spawning season with low values of biochemical constituents indicating the shed down of male and female gamates. Thus, a bimodal pattern of reproductive cycle with accumulation and utilisation of different biochemical constituents, mostly carbohydrates and glycogen, was observed in both species. A similar bimodal pattern was reported in gastropod *Littorina irrorata* but with peaks in May and August (Bistransin 1976). However, a unimodal pattern is not uncommon in cold water gastropods as observed by Stickle (1973) in *Thais lamellosa* in which the TDG, ODG and CAG were found to cycle with the season. Belisle and Stickle (1978) studied seasonal changes both in level and content of carbohydrates, protein and lipid in different body components of *T. haemastoma* and they found a bimodal pattern of seasonal change. They have also reported that the changes in different biochemical constituents were predominant due to component indices but not to fluctuations in constituent levels. Bistransin (1975) observed a bimodal pattern of seasonal changes in the RVM of *L. irrorata* with peaks in May and August. In *Clypeomorus clypeomorus*, an intertidal cerithiid, high amounts of biochemical constituents were observed from June–August and December–February due to development of gonads (Manmadha Rao 1977). The increase in different biochemical constituents viz. glycogen, protein and lipid during gonad development was reported in several molluscs, *Parreysia corrugata* (Nagabhushanam and Lomte 1971), *Paphia laterisulca* (Nagabhushanam and Dhamne 1977), *Mytilus viridis* (Nagabhushanam and Mane 1978), *Villorita cyprinoides* var. *Cochenensis* and *Meretrix casta* (Lakshmanan and Krishnan Nambison 1980). Deshpande and Nagabhushanam (1983) while studying the seasonal changes in the biochemical composition of *C. iatricus* and *K. tunicata*, suggested an increase of the glycogen content with the ripening of testes and ovaries in both the species indicating its synthesis and utilisation during reproductive period. Recently, Uma Devi *et al* (1985) reported a progressive increase of all biochemical constituents during May–October and a decrease during November–April in *M. granulata* and they have correlated these changes to reproductive season and feeding of the animal.

Food availability is also one of the factors that influence the biochemical constituents of the animals (Ansell 1974). In the present investigation, the food availability in the habitat of *C. cingulata* is more when compared to *C. coralium*. Both of them feed on algae (*Ulva* and *Enteromorpha* sps.) and myriads of benthic diatoms. Because of frequent disturbances, the algal growth is found to be not much in the lower reaches. This may be one of the probable reasons for the presence of greater amounts of biochemical constituents in *C. cingulata* than *C. coralium*. However, the run-off from the river may also bring nutrients for *C. cingulata*. It was also reported (Annapurna 1978) that organic composition was found to be higher in the habitat of *C. cingulata*. As the environmental conditions in the habitat of *C. cingulata* show wide variations, the animal has to withstand all these problems and reproduce. Therefore, they need more energy for propagation and the reproductive material should also be produced much to counter the environmental disturbances. Hence, these animals of *C. cingulata* have stored more quantities of biochemical constituents than *C. coralium*.

It is interesting to observe a carbohydrate-oriented metabolism in these cerithiids as the glycogen and carbohydrates are the major biochemical constituents utilised during reproduction. These cerithiids also exhibited a carbohydrate-oriented metabolism when they were subjected to starvation stress (Prabhakara Rao *et al* 1987) and oxygen lack (Prabhakara Rao and Prasada Rao 1983). Thus, whenever, these animals are in need of energy, they switch on to carbohydrate reserves. Similarly carbohydrate-oriented metabolism was reported in several molluscs (Emerson 1967; Ramamurti and Subrahmanyam 1976; Manmadha Rao 1977; Nagabhushanam and Mane 1978). Next to carbohydrates, these animals preferred lipids and lastly proteins are utilised. According to Giese (1959), only 5% of the total lipids constitute for the structural lipids and the rest might be a storage for utilisation. Moreover, Lawrence and Giese (1969) also reported increased utilisation of lipids during gamatogenesis. However, protein content was not much utilised in cerithiids but in some of the other molluscs, proteins appeared to be the major biochemical reserves for utilisation during reproduction (Giese and Araki 1962; Stickle 1975; Deshpande and Nagabhushanam 1983). The TNPS values of both the animals showed decline in March and April because of utilisation for the formation of proteins. Another set of minimum values occurred in September and October. The highest values in June and July and, December and January might be due to proteolysis and they are in synchronisation with protein depletion.

Acknowledgements

The authors are (YPR, VUD) thankful to the authorities of Council of Scientific and Industrial Research, New Delhi for financial assistance. Thanks are also due to the Head of the Department for providing necessary facilities.

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