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## RESEARCH ARTICLE

# REPRODUCTIVE CYCLE AND BIOCHEMICAL CHANGES IN THE GONADS OF THE FRESHWATER CRAB, *BARYTELPHUSA GUERINI* (H. MILNE EDWARDS) (DECAPODA, POTAMIDEA)

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**ABSTRACT**

The reproductive cycle of the freshwater crab, *Barytelphusa guerini* is divided into three different periods i.e. (i) Pre-reproductive period (January to April) (ii) Reproductive period (May to August and (iii) Post-reproductive or quiescent period (September to December). The protein percentage was higher in the month of April. From June onwards, the percentage of proteins decreased and lowest was recorded in the month of December. Maximum fat content was observed in the ovaries in the month of April and May (breeding period) and in testes, it was highest in the month of May only. There was highest value of glycogen obtained in the month of April and there was significant decrease in the glycogen content during the spawning period. There was a little consistency in the glycogen content during the resting period.



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## INTRODUCTION

Studies on biochemical changes in relation to reproductive cycle in Invertebrates have investigated by some workers. Seasonal variations in different organic constituents of oysters had been shown by Rusell (1923). Okazaki and Kobayashi (1929), Sekine, *et al,* (1930), Humphrey (1941), Venkatraman and Chari (1951) and Durve and Bal (1960), Pearse and Giese (1966) have worked on the biochemical changes in relation to reproductive cycle in gastropod, *Neobuccinum eatoni* and in bivalve. *Limatula hodgsoni*. Giese (1969) in his review paper, gave a complete account of the biochemical composition of different tissue in relation to reproductive cycle in a number of molluscs. Recently, Weber (1970), studied the biochemical changes in different organs of *Haliotis crachenoidii* in relation to reproductive cycle. When compared to the investigations on marine decapod crustaceans studies on the reproductive cycle and breeding behaviour of the freshwater crabs in India have received very little attention.

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MecCann (1937) described the life history of the freshwater crab, *Barytelphusa guerini*. Chacko and Thyagarajan (1952) studied the breeding season of the crab, *Paratelphusa jacquemontii*. Very few studies has also been reported on the biochemical changes in relation to reproductive cycle in decapod crustacea in India, especially on freshwater crab, George and Patel (1956); Diwan and Nagabhusanam (1974); Farooque and Nagabhusanam (1983) were the first to study the seasonal variation in the biochemical constituent in freshwater and marine decapods. Since, little work has been done on the reproductive biology of the fresh-water crabs in India and as already the phases of reproductive cycle has been investigated in the freshwater crab, *Barytelphusa guerini* (Gangotri, *et al*., 1978). Hence, the present study was undertaken with a view to investigate the changes in the chemical composition of the gonads in this crab.

## MATERIALS AND METHODS

The adult specimens with a carapace width of 45 mm of freshwater crab, *Barytelphusa guerini*, which are sexually mature and reproductively active Gangotri *et al.* (1978) were collected from the field for a period of one year (January 2014 to December 2014).

The male and female crabs were maintained in glass troughs containing sufficient water, specimens that were not healthy conditions as well as those that had just moulted were not used for analysis. The gonads collected in each month for one year during the period January 2014 to December 2014 for histological observations. Some part of the same gonads were also used for biochemical estimation in each month. The biochemical changes were recorded in the gonads for a period of one year. For estimation of glycogen, the method recommended by Kemp, *et al.* (1954) was employed using Engel's Colorimeter. The amount of glycogen was calculated by multiplying the glucose value by the factor 0.927. The fat content was extracted from dried tissue in Soxhlet apparatus and the percentage of fat was estimated. The nitrogen was estimated by Micro-Kjeidhal Method (Hawk *et al*., 1954). The amount of total protein was calculated by multiplying the nitrogen value by 6.25. All the results were expressed in percentage of the sample on dry weight basis.

## RESULTS

**Glycogen:** The glycogen contents in the gonads were estimated monthly for one year i.e. January 2014 to December 2014. The glycogen content in case of testis was within the range of 0.563 to 0.989 mg %, whereas, in ovary, it ranged from 0.535 to 0.852 mg %. The data obtained during the year shows certain seasonal changes in glycogen contents of the male and female gonads especially during the breeding season. There was a significant decrease in glycogen content during the spawning period i.e. May to August. The glycogen content was minimum in both males and females at the end of spawning season i.e. in the month of August. During the resting stage or post reproductive season i.e. September to December, there is a little consistency in the glycogen content of the gonad. During the preparatory period or ripening period of the gonad i.e. January to April, the percentage of glycogen was increased.

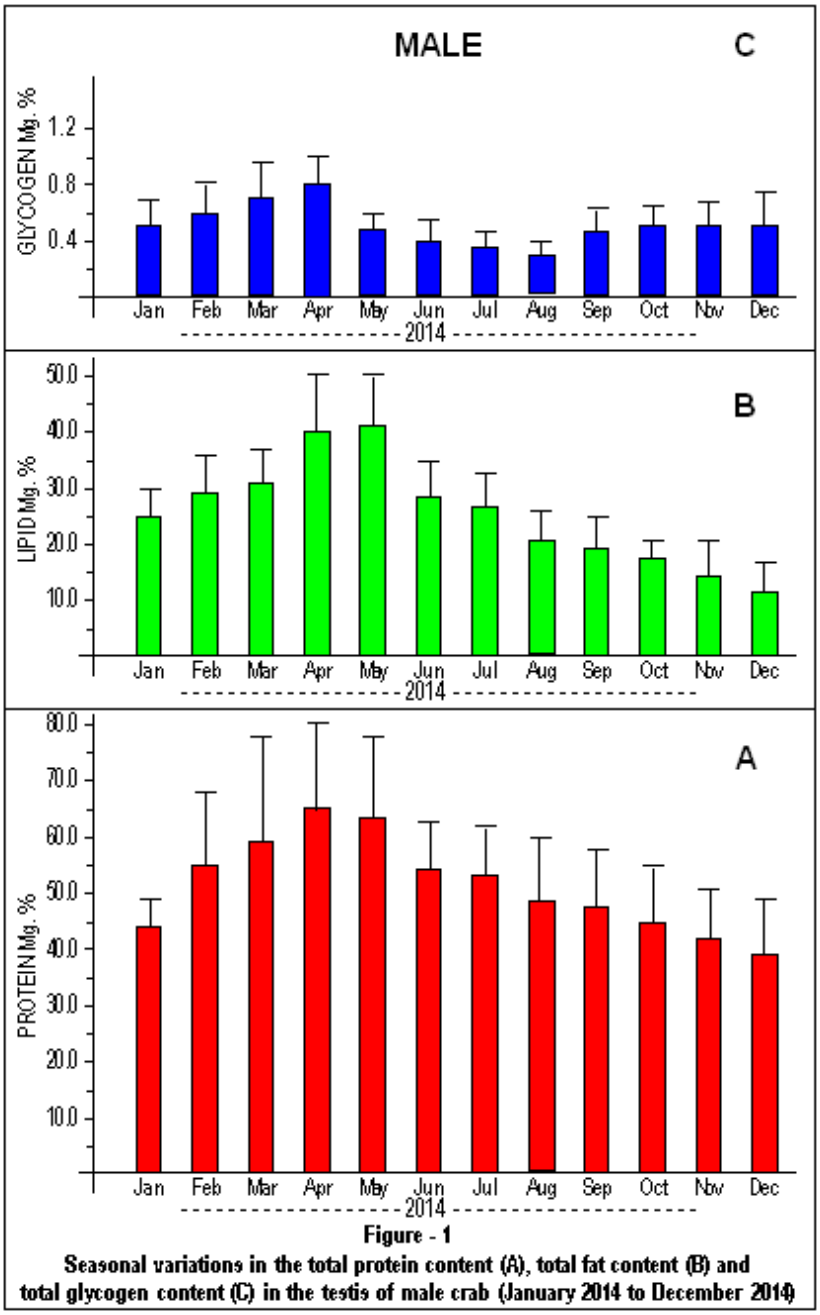


**Figure - 1**

**Seasonal variations in the total protein content (A), total fat content (B) and total glycogen content (C) in the testis of male crab (January 2014 to December 2014)**

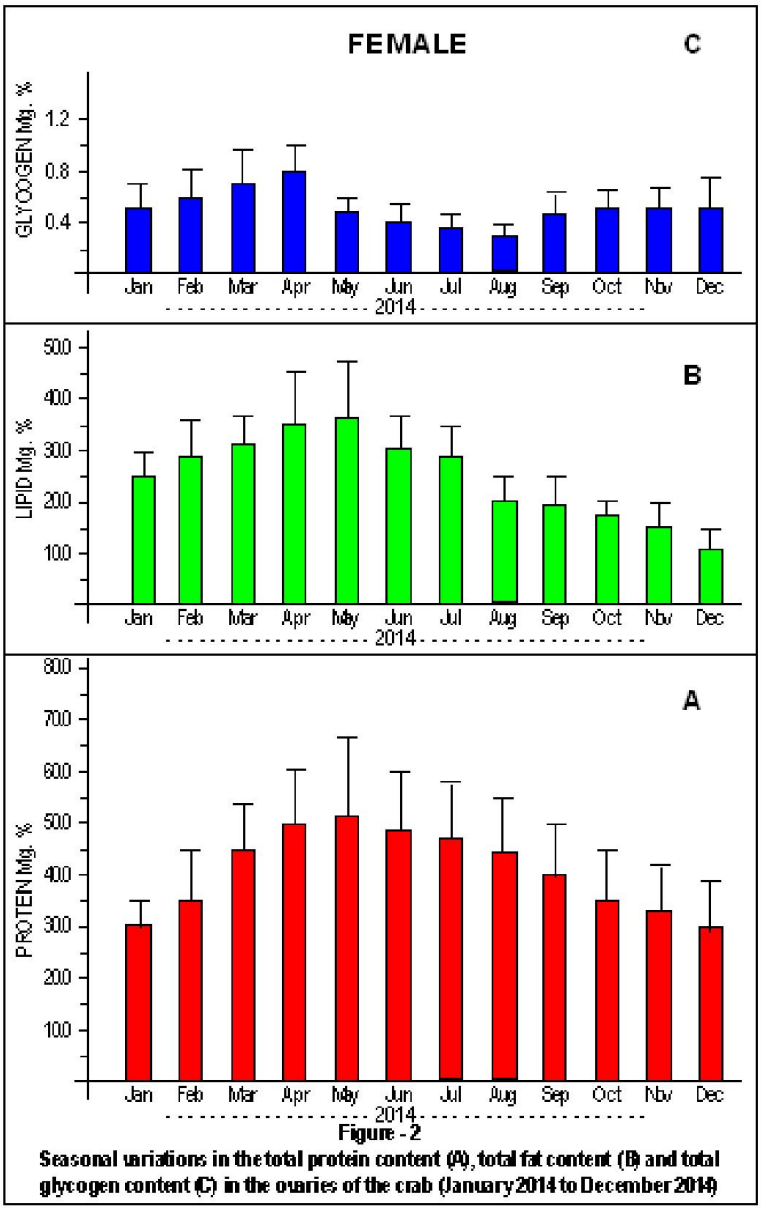


**Figure - 2**

**Seasonal variations in the total protein content (A), total fat content (B) and total glycogen content (C) in the ovaries of the crab (January 2014 to December 2014)**

The highest value was noticed in the month of April in both the male and female gonads (Figure - 1 and 2).

**Lipids:** The total fat contents of the gonads was estimated in each month during the year January 2014 to December 2014. The total fat content in the testes and ovaries was in the range of 22.5 % to 35.3 % and 15.5 % to 40.5 % respectively. The maximum fat content was observed in the ovaries in the month of April and May; and in testes, it was highest in the month of May only. There was a steady increase of fat content in the gonads was noticed during the maturation of the gonads i.e. January to April. During the spawning period i.e. June to August, the fat content in both sexes decreased. (Figure - 1 and 2)

**Protein:** The proteins were also estimated in every month during the year from January 2014 to December 2014. The results obtained show that the protein percentage was in the range of 40.50 mg % to 65.42 mg % in tastis and 30.52 mg % to 50.35 mg % in ovaries.

From the results, it was seen that, the protein percentage was highest in the month of April. From June onwards, the percentage of protein decreased and the lowest was recorded in the month of December (Figure - 1 and 2).

## DISCUSSION

The protein level in the gonads during the month of the year i.e. January 2014 to December 2014) was studied in *Barytelphusa guerini*. In the testis and ovary protein level showed marked variations during the reproductive cycle. The level was highest in the month of April, when the gonads were in developing condition. The highest percentage of protein in testis was 65.10 mg. whereas in ovary it was 50.12 mg. The protein level declined to minimum level during the spawning period. The lowest value noted in December for the testis was 40.30 mg and for ovary 30.25 mg. From January onwards, the level of protein was increased till the complete maturation of the gonads. Giese, *et al.* (1958), while working on the organic productivity in the reproductive cycle of the sea urchin,

*Stronglylocentrotus purpuratus* observed the increase of protein and RNA with the maturation of ovary, while increase of DNA proportionally was less. In testis protein and DNA increased more, while RNA increased less than the increase in the total bulk. The present observation regarding the protein level in *Barytelphusa guerini* confirms the findings of Durve and Bal, (1960), in Oystar, *Crassostrea graphoides* and Giese, (1969) in *Katharina tunicata* (Greenfield, *et al.,* 1958); *Barytelphusa cunicularis,* (Diwan and Nagabhushanam, 1974); Webber, (1970) in *Haliotis* found that the protein level did not show much variation during the reproductive cycle.

The total fat content was increased in testis and ovary during the ripening of the gonads. The highest value noted was 40.10 mg % in testis and 35.5mg % in ovary. During spawning period, the percentage of fat was decreased. Thiele (1959), determined the total fat content of *Helix* and found only slight seasonal variation. It would appear probable that as the gonad grew, it received fat from the other parts of the body and accumulated in developing gonads. The fat appeared to be one of the most important sources of energy metabolism during the breeding season. With greater catabolism of fat during the spawning period, the fat content was lowered to its minimum value. Giese (1969), while studying the reproductive cycle of *Katharina tunicata* found the accumulation of lipids in the ovary during the formation of ova preceding spawning and the lipid level remained essentially the same from month to month in mantle, testis and gut.

In another study on *Catharina* (Lawrence and Griese, 1969). It was found that the partition of lipid between the neutral lipids and phospholipids was about the same in the ovary, whenever, examined during the reproductive cycle, but the relative amount of phospholipids increased with increase in size of the testis, while neutral lipid fraction declined. This shows increased storage of phospholipids with increase in size of the testis. Results of the present study on *Barytelphusa guerini* are in agreement with the findings reported by Giese, *et al.,* (1958), in *Strongylocentrotus purpuratus*. In three species of echinoderms, *Pisaster Ochraces*, *Pisaster giganteus* and *Strongylocentrotus franciscanus*, (Greenfield, *et al.,*1958); Durve and Bal (1960), in Oyster, *Grassastrea gryphoides*. Pearse and Giese, (1966), in *Haliotis Cracheroidii* and *Barytelphusa cunicularis* (Diwan and Nagabhushanam, 1974). The variability of chemical constituents of the gonads of *Barytelphusa guerini* during the year, may be just another index to confirm the spawning period of this species. Carbohydrate level probably represents storage of food (Chiefly glycogen) in tissues and might be expected to vary during the reproductive period. A striking change in the glycogen level was seen during the course of reproductive cycle in testis and ovary. The level reached lowest value at the time of spawning period and after spawning the level was increased till the complete growth of the gonad. This suggest that the stored glycogen might be utilised for the formation of reproductive elements and this counts for the decrease in the glycogen content during the breeding season.

It is known that the hepatopancreas serves as a storage organ, the principal storage nutrients being fat and glycogen (Yonge, 1924). It is further suggested that the organic material may be transferred from the storage organ to gonad as the animal matures (Ferguson, 1964), Okazaki and Kubayashi, (1929), while working on the Japanese Oysters have stated that glycogen was at minimum level when breeding occurred. Humpray (1941) and Hatanka (1940) also suggested that glycogen acted as a reserve food material and utilized for the formation of gonad products. Similar observations made by Giese, *et al.* (1958), on purple Sea Urchin, *Strongylocentrotus* purpuratus and Durve and Bal (1960) on Oyster, *Crassostrea gryphoides*. The gonads of *Katharina tunicata* showed lowest value of glycogen during the spawning period and the level rose again after spawning (Giaese, 1969). The same results were also reported in *Barytelphusa cuniculasis* (Diwan and Nagabhushanam, 1974). It would be thus clear from the foregoing account that the glycogen shows seasonal changes in its percentage in association with the sexual and metabolic activity.

In conclusion, the results obtained on histological and biochemical observa-tions in the three different periods of reproductive cycle i.e. (i) Preproductive period (January to April) (ii) Reproductive period (May to August) and (iii) the Post-reproductive period (September to December) in *Barytelphusa guerini* suggests that these three periods are characterised by the variations in biochemical composition.

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