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Growth Indices and Reproductive Pattern of Freshwater Mussel Lamellidens Marginalis (Lamarck, 1819) in Northwest Bangladesh

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GROWTH INDICES AND REPRODUCTIVE PATTERN OF FRESHWATER MUSSEL *LAMELLIDENS MARGINALIS* (LAMARCK, 1819) IN NORTHWEST BANGLADESH

A THESIS SUBMITTED TO THE UNIVERSITY OF RAJSHAHI, BANGLADESH FOR THE DEGREE OF

DOCTOR OF PHILOSOPHY (PhD)

Submitted by

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Dedicated

To

My Beloved Parents

Declaration

I hereby declare that the whole work submitted as a thesis entitled GROWTH INDICES AND REPRODUCTIVE PATTERN OF FRESHWATER MUSSEL *LAMELLIDENS MARGINALIS* (LAMARCK, 1819) IN NORTHWEST BANGLADESH to the Department of Fisheries, Faculty of Agriculture, University of Rajshahi, Rajshahi-6205, Bangladesh for the degree of Doctor of Philosophy (PhD) is the result of my own investigation which has been carried out under the supervision of Professor Dr. Saleha Jasmine and Professor Dr. Md. Mostafizur Rahman Mondol**,** Department of Fisheries, Faculty of Agriculture, University of Rajshahi, Rajshahi-6205, Bangladesh. I further declare that thesis has not been submitted elsewhere for any other degree.

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CERTIFICATE

This is certify that the entitled **GROWTH INDICES AND REPRODUCTIVE PATTERN OF FRESHWATER MUSSEL *LAMELLIDENS MARGINALIS* (LAMARCK, 1819) IN NORTHWEST BANGLADESH** has been prepared by Dil Afroz Nahar (Reg. No. 10037; Roll No. 13602; Session: 2013-2014) under my supervision for submission to the Department of Fisheries, Faculty of Agriculture, University of Rajshahi, Rajshahi 6205, Bangladesh for the degree of Doctor of Philosophy (PhD). All the data presented in this thesis are based on her own observation and no part of this work has been previously published or submitted for any other degree.

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The authoress

Summary

Freshwater bivalve *Lamellidens marginalis* (Lamarck, 1819) (Bivalvia: Unionidae) is an important species in the aquatic ecosystem of Bangladesh. The present study investigated the growth indices and reproductive pattern of freshwater mussel *L. marginalis* in northwest Bangladesh. The present study was carried out in the freshwater lake located at 24º 22´ 21.56´´ N and 88º 38´ 11.16´´E. Monthly specimens of *L. marginalis* were collected from the study area for a period of 19 months during January, 2014 and July, 2015. Additionally water quality parameters of the study area were also monitored monthly.

Chapter I describes the general introduction and objectives of the study and Chapter II describes the review of literature.

Chapter III describes the water quality parameters of the study site. During this study all the water quality parameters were within the suitable range and showed seasonal some fluctuations. The water temperature ranged from 18.5 to 34.5 ^{0}C, transparency varied from 19.8 cm to 31.2 cm, pH ranged from 6.6 to 7.8, dissolved oxygen fluctuated from 4.8 to 7.0 mg/l, free carbon dioxide was ranged from 4.3 to 6.8 mg/l and total alkalinity ranged from 65 to 95 mg/l.

Chapter IV describes the growth pattern of the freshwater bivalve mollusk *L. marginalis* from the northwest Bangladesh. During the study the mussel showed a significant linear growth pattern for length-length relationships and non-linear growth pattern for length-weight relationships ($P<0.05$). The length-breadth and length-width relationships for the entire study period were $B = 0.179 + 0.475L$ and $W =0.196+ 0.3163L$ respectively. The length-total weight, length-wet tissue weight, length-dry tissue weight, length-shell weight, length-dry shell weight relationships were $W = 0.1241L^{2.9066}$, $W = 0.0524L^{2.7377}$, $W =0.0116L^{2.7849}$, $W = 0.0533L^{2.9178}$ and W =

$0.0391L^{3.0386}$ respectively. The values of *b* indicated the relative growth in body weight and better physiological condition of the mussels in the study area.

Chapter V describes seasonal variation in condition index and meat yield index of the freshwater mussel. Condition index is an important parameter, which shows the physiological status of bivalves and its seasonal variation linked with the reproductive activity as well as the seasonal energy storage of bivalves. In the present study the highest condition index was observed in June (23.35) and lowest in November (10.44). Condition index increased during April and June and then sharply decline during June and August. Meat yield index also showed similar monthly fluctuation as condition indices. The higher condition index and meat yield index of the mussel during winter and summer indicated a better period of commercial exploitation of the mussel from the natural ground.

Chapter VI describes the seasonal variation in reproductive pattern of the freshwater mussel. During the study sex of the mussel was differentiable in all the months. Out of 620 specimens collected 45.97% were female and 54.03% male. The overall male to female sex ratio was 1: 0.82, which was statistically different from the expected sex ratio of 1:1 (X^2 = 5.90, P < 0.05). Histological observation revealed five gonad development stages as early developing, late developing, ripe, spawning and spent both for male and female mussels. During this study sexually undifferentiated and hermaphroditic individual were not found. Patterns of gonadal development, maturation and spawning were similar both for males and females. The mussel *L. marginalis* spawned throughout the year with peak spawning during July and August when most of the mussels (up to 70%) participated in spawning. In the present study the 6-7 cm size group mussels were more potential group in respect to reproductive performance. Seasonal changes of gonadal index also clearly related with maturity stages: GI increased progressively with the development of maturity to reach a maximal value (9.41± 1.23) in June indicates the maximum ripeness of the gonad of the mussel. The GI value sharply declined from July to afterwards at reaching a minimal value of 4.83±0.96 in the month of September, indicates the spawning of the mussel population during this time.

Chapter VII describes the seasonal variation in biochemical composition (water content, protein, carbohydrate, and lipid contents) of the mussel. During the study the water content ranged from 74.66 to 81.88%, carbohydrate level ranged from 19.83%-21.25%, protein ranged from 14.04-38.88% and lipid content ranged from 5.3%-12.06%. The biochemical composition showed several monthly fluctuations and the fluctuations of carbohydrate and protein level was inversely related to each other. Higher carbohydrate observed during January and March; indicate higher energy accumulation of the mussel during this period. A higher protein level observed during June and July could be linked with the ripe gamete of the mussel and lower level of protein during August to onwards could be linked with the spawning of the mussel.

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CHAPTER I

General Introduction

Bivalve mollusk constitutes a significant part of the world fishery today. Bivalves alone contributed 11.6 % to global aquatic production during 2016 and ranked 3rd position just next to finfish and seaweeds (FAO, 2018). Mollusks have been exploited worldwide for food, ornamentation, pearls, lime, and medicine (Nayar and Rao, 1985). They are of major interest to man and many species can play a great role in the national economy. From the standpoint of food alone mollusks have been the tremendous importance to man since prehistoric times. Mollusks meat is extensively used as human food. In south East Asia fish and mollusks provide over 50% of all consumed animal protein (Shabuddin et al., 2010). Beside this mollusks are also used for variety of other purposes as pearl production, bio-indicator in pollution monitoring, in controlling of harmful algal blooms, preparation of lime, poultry, shrimp and fish feed (Shabuddin et al., 2010).

In the past five decades, global fisheries and aquaculture have grown steadily, and seafood consumption per capita has increased from an average of 9.9 kg in the 1960s to 19.2 kg in 2012 (FAO, 2012). This development is impressive, and probably driven by multiple factors, such as population growth, rising income, and urbanization (Diana, 2009). Molluskan shellfish has traditionally been a major component of world aquaculture. For example, the molluskan shellfish production in 2012 (15.2 million metric tonnes) accounted for about 22.8% of the total (inland and marine) aquaculture production and 60.3% of the world marine aquaculture production (FAO, 2014).

Worldwide, the major aquaculture molluskan species groups include clams (together with cockles and arkshells), oysters, mussels, scallops, abalones, and other miscellaneous mollusks. Based on their production in 2012, clams, oysters, mussels, and scallops were the four major groups and accounted for 87% of the total molluskan aquaculture production (Fig.1). Mollusks are cultured in 76 countries worldwide (FAO, 2014). The top ten countries for molluskan aquaculture production are China (81.4%), followed by Vietnam (2.6%), Korea Republic (2.5%), Japan (2.3%), Chile (1.7%), Spain (1.4%), Thailand (1.4%), USA (1.1%), France (1.1%), and Italy (0.7%) (Fig. 2, FAO, 2012).

The Mollusks are soft bodied animals with or without an external protective shell. They inhabit usually water bodies, marine, estuarine, as well as freshwater; many are also terrestrial, often associated with moist shaded lands. The first Mollusk appeared at the end of the Pre-Cambrian period, approximately 550 million years ago (Sturm et al., 2006). It is the second largest phylum in the invertebrates comprising more than 100,000 species worldwide and of theses about 5000 species are of freshwater (Venkataraman and Wafar, 2005). Mollusks are also one of the larger invertebrate groups in the fresh water habitat of Bangladesh (Baby et al., 2010). If the body of the Molluskan taxa is enclosed by a pair of shells hinged in the middle it can be classified under the Class Bivalvia. It has two shells or valves join together with the help of teeth like structure called hinge and fibrous tissue-ligament. The shells are made up of calcium carbonate. Most of the forms are completely sedentary remaining attached to hard substrata by thread-like byssus of the foot or by one of their shell valves. A few forms burrow into submerged timbers, and commensal and parasitic types are also known. Some marine forms extended to a depth of 4.94 km. Life histories of bivalves pass through larval stages, which undergo remarkable changes before attain adult characteristics. Bivalve mollusks are typically filter feeders of phytoplankton and are primarily free spawning with external fertilization.

There are different species of mollusks that occurred naturally in the freshwater habitat of Bangladesh of which freshwater mussel *Lamellidens marginalis* is the most preferred species used as food by some ethnic groups in Bangladesh. *L. marginalis* is found in the lower and upper Gangetic plains in India and Bangladesh, SriLanka, Myanmar and Terai region of Nepal (Nesemann et al., 2007). They are encountered in greater abundance in waterways located in alluvial soil areas with soft soil substrate harboring green algae. It is said that *L. marginalis* is a typical pond species normally distributed in stagnant to slow flowing habitats like ponds, tanks, lakes and reservoirs at a depth of 0.5 m and beyond (Misra et al., 2000; Misra, 2005). Neutral to slightly alkaline waters are in general conducive for mussel colonization. The shells are used to extract edible lime in Terai region of Nepal. It is used in pearl culture in India (Ramakrishna and Dey, 2007) and Bangladesh. It is also used as medicine.

In the past freshwater bivalve *L. marginalis* was abundantly available in the natural habitats of Bangladesh. Now a day's bivalve population is declining as because of over harvesting, environmental degradation, alteration of water body into crop land, water pollution, lack of proper knowledge on biological aspects of this species. Understanding their biological functions within aquatic ecosystems is vital for successful management. Globally substantial work has been done in different aspects of biology of freshwater bivalves (Narain, 1972; Moorthy et al., 1983; Tippeswamy and Joseph, 1988; Ravera et al., 2007; Satit et al., 2008; Ramesha and Tippeswamy, 2009; Malathi and Tippeswamy, 2011; Suryawanshi and Kulkarni, 2014a, b; Mondol et al., 2016b; Niogee et al., 2019; Nahar et al., 2019). From Bangladesh some information on bioogical aspects of marine mollusks was also provided by Ali & Aziz (1976), Ahmed et al., (1978) and Asaduzzaman et al. (2019). To the best of my knowledge, there is no precise and detailed information particularly on freshwater mussel *Lamellidens marginalis* from Bangladesh. Therefore, the present study was designed to investigate growth indices and reproductive pattern of freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in northwest Bangladesh.

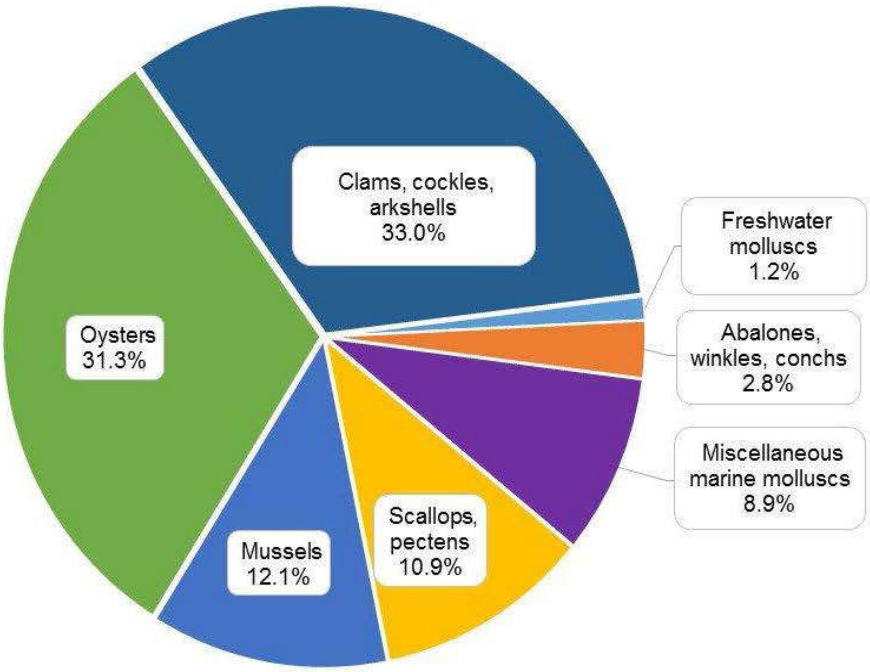

Fig.1. The percentage of aquaculture production (15.2 million metric tonnes) for the major molluskan species groups in 2012 worldwide. (Source: FAO, 2014)

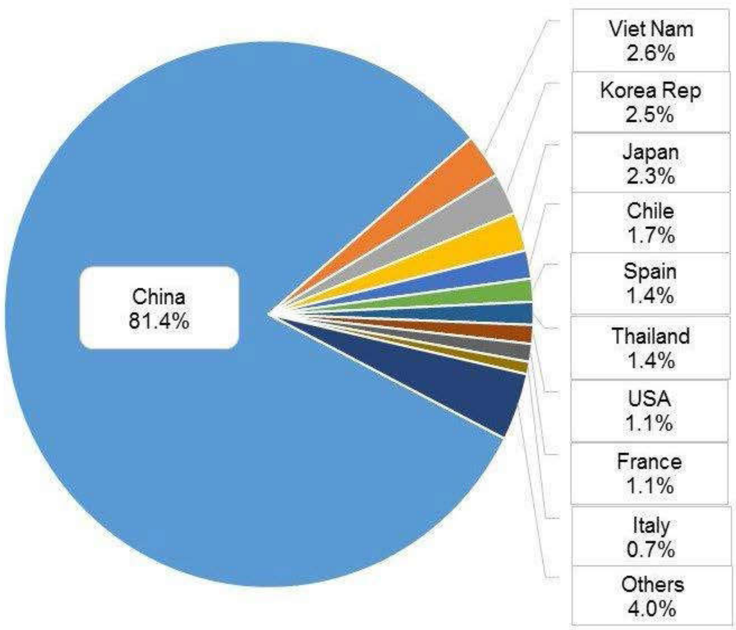

Fig. 2. The percentage of molluskan shellfish aquaculture production in the top ten countries worldwide in 2012. (Source: FAO, 2014)

1.1 Specific objectives

The specific objective of this study were-

- to know the water quality parameters of the study area;
- to know the length-frequency distribution;
- to know length-length and length-weight relationships;
- to know the temporal variation of condition index and meat yield index;
- to know the sexuality, sex ratio, spawning & peak spawning season and reproductive effort; and
- to know the seasonal changes in biochemical composition (water content, carbohydrate, protein and lipid) and its link with somatic growth and reproduction of the *L. marginalis* in northwest Bangladesh.

1.2. Confinement of this thesis

The present thesis is comprised into seven chapters. Chapter I provides general introduction providing the overall information on the bivalves including the ecological, economical importance and morphological features of *Lamellidens marginalis*. Objectives of the present work are also given in this chapter.

Chapter II deals with the review of literature. The data generated in the present investigation would strengthen the existing knowledge on *L. marginalis* from the northern Bangladesh.

Chapter III deals with the water quality parameters of the study site. In this chapter seasonal dynamics of water temperature, transparency, pH, dissolved oxygen, free carbon-dioxide and alkalinity of the study area were studied.

Chapter IV deals with the growth pattern of freshwater bivalve mollusk *L. marginalis* (Lamarck, 1819) (Bivalvia: Unionidae) from the northwest Bangladesh. In this chapter the morphometric and length-weight relationships between shell length-shell breadth and shell length-shell depth, shell length-total weight, shell length-shell weight, shell length-wet weight, and shell length-dry weight variables of *L. marginalis* were studied.

Chapter V deals with the seasonal variability in condition indices of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in the northwest Bangladesh. In this chapter different methods were used to determine the condition index of the bivalve.

Chapter VI deals with the seasonal variation in reproductive pattern (sexuality, sex ratio, gonado-somatic index and reproductive output) of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in the northwest Bangladesh.

Chapter VII deals with the seasonal variation in biochemical composition (water content, protein, carbohydrate and lipid) of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in the northwest Bangladesh.

CHAPTER II

Review of Literature

It is very important to review the earlier studies or research works related to the planned study before conducting the study or experiment. Literature on the biology of freshwater bivalve is scare in Bangladesh. So the research works done in various parts of the world are reviewed below for clear and better understanding of the present study.

1. Water quality parameters

The primary productivity of a water body depends on some common physico-chemical factors. Therefore, it is important to understand the role of various physico-chemical factors that affect the life process of fish and other aquatic biota. Many works have been done on water quality parameters and some works relevant to present study are reviewed below:

Alikunhi (1957) reported that good water for fish cultivation should have a fair amount of dissolved oxygen (5.0 to 7.0 ppm). He also stated that a good pond water for fish cultivation should be concentration of 0.2 to 4.0 ppm of phosphate and 0.06 to 0.1 ppm of nitrate.

Banerjee (1967) stated that ponds become unproductive when dissolve oxygen value ranged from 3.0 to 5.0 ppm. He also reported that 7.0 ppm dissolve oxygen and pH value ranging from 6.5 to 7.5 good for fish culture.

Swingle (1967) reported that good relationship between pH and fish growth and obtained satisfactory results and reported pH 6.5 to 9.0. He also observed that water pH more than 9.5 was unproductive and pH more than 10.0 was lethal for fish.

Dewan (1973) recorded temperature range of 19.0 °C to 35.0 °C, dissolve oxygen value of 3.8 to 10.7 ppm in his study on the ecology of a lake in Mymensingh. He also observed an inverse relationship of dissolve oxygen with temperature and free carbon dioxide (CO_2) and direct relationship with pH and total alkalinity.

Openheimer et al. (1978) measured water quality parameters of three ponds in Dhaka with the use of portable Hack Kit. They reported the mean value of temperature 25.54 to 26.85°C, dissolved oxygen 3.18 to 7.58 ppm and pH 6.9 to 7.6.

Mumtazuddin et al. (1982) studied the physico-chemical characteristics of freshwater ponds in Mymensingh. They observed vertical and fortnightly variations of temperature, pH and dissolve oxygen which were within the acceptable range of fish rearing practices. They found temperature value around 30 °C, DO values of 5.0-10.0 ppm and pH value above 7.0 in four rearing ponds.

Rahman et al. (1982) measured the water temperature and dissolved oxygen of a fish pond. They found temperature of 25.05 to 32.89 °C and 26.05to 32.22°C for surface and bottom temperatures respectively and dissolve oxygen range of 0.40 to 8.60 ppm in four selected ponds.

Azim et al. (1995) recorded the mean values of temperature 26.0°C, transparency 36.2 cm , pH 7.1 , total hardness 50.5 ppm from their experiment conducted in a set of ponds at the field Laboratory of the Faculty of Fisheries, Bangladesh Agricultural University.

Wahab et al. (1995) measured a number of water quality parameters in nine experimental pond at Bangladesh Agricultural University Campus, Mymensingh. They reported that, the water temperature varied between 27.2-32.4 °C, secchi disc depth between 26 and 50 cm. The pH was alawys around 6.0 and dissolved oxygen was always low and varied between 2.2 to 7.5 ppm.

DoF (1996) reported that the suitable range of dissolved oxygen of a water body for fish culture would be 5-8 mg/ l, range of pH 6.5-8.5 and temperature was in the range of 20-30 °C.

Hossain (2007) found the temperature (19.75 to 27.25 °C); transparency (24.75 to 29.50 cm); pH (6.62 to 7.85); dissolved oxygen (3.87 to 5.85 ppm); free Co_2 (5.25 to 7.25 ppm) and alkalinity (81.25 to 147.5 ppm) in earthen fish pond of Bangladesh.

Jasmine et al. (2013) studied the plankton production in relation to water quality parameters in lentic and lotic water bodies during post monsoon season in the northwestern Bangladesh. They reported the water temperature for large ponds 16-33.6°C, transparency 30-38 cm, pH 5.2-7.5, dissolved oxygen 5.0-7.5 ppm, free carbon dioxide 5.9-7.0 ppm and total alkalinity 68-100 ppm.

2. Morphometry and length-weight relationship

Hemchandra and Thippeswamy (2008) described the morphometry, length–weight and condition index of green mussel *Perna viridis* (L.) from St Mary's Island, off Malpe, near Udupi, from December 2004 to December 2005. They calculated regression equations between length and breadth and length and width for the entire study period were L=0.5071+ 0.3921BandL=0.0179+ 0.3225W *r*espectively. The monthly *b* values of length–breadth and length–width relationships varied from 0.3636 (March 2005) to 0.4374 (December 2004) and from 0.3022 (December 2005) to 0.3466 (August 2005) respectively. The data on length–total weight ($W=0.0986L^{2.9495}$), length–wet tissue weight ($W=0.031L^{2.7173}$), length–dry weight ($W=0.005L^{2.9337}$) and length–shell weight ($W=0.0351L^{3.061}$) showed a nonlinear pattern. The coefficient of allometry (*b*) values ranged from 2.7949 (April 2005) to 3.0999 (September 2005) for length–total weight relationship and from 1.5203

(August 2005) to 3.328 (March 2005) for length–dry weight relationship.

Kovitvadhi et al. (2008) reported the freshwater pearl mussel, *Hyriopsis (*Limnoscapha) *myersiana* (Lea, 1856), cultured in the laboratory (0–120 days old), in an earthen pond in acrylic containers (120–270 days old) and in pocket nets with frames (270–360 days old). Measurements were taken of the length–height and the total body weight–size (shell length, shell height and shell width). It was found that the length–height in each period of culture showed a linear relationship and that the shell height increased as the shell length increased, along with the same linear relationship. When statistical analyses were performed, this linear regression line consisted of three distinct lines ($P<0.01$). The general morphometric relationships between the length and height of mussels grown in the laboratory, in acrylic containers and in pocket nets were Log H = -0.147 + 0.828Log L, Log H = -0.211 + 0.909Log L and Log H = -0.264 + 0.951Log L, respectively. The relationship between the total body weight–size in different culture phases was a positive allometric equation and the two curved lines were not the same ($P>0.01$). The general morphometric relationships between the length and the total body weight of the mussels cultured in acrylic containers and pocket nets were Log W = -3.747 + 2.674Log L and Log W = -4.149 + 2.976Log L, respectively. Such measurements are useful in the management of different culture phases of *H.myersiana* (L.).

Laxmilatha (2008) studied the length-weight (L-W) and other biometric relationships of the surf clam *Mactra violacea*, collected from the north coast of Kerala. In *M. violacea*, the L-W relationship between males and females was not found to be significantly different and therefore a combined equation was derived. The growth in length is accompanied by weight increase. However, in all other biometric relationships such as length-width (L - B), length-height (L - H), total weight-meat weight (TW - MW), total weight-shell weight (TW - SW), and shell weight–meat weight (SW - MW)), allometric growth was observed. In the case of L - B, L - H and TW - MW, a positive biometric relationship exists ($b > 1$) while in TW - SW and SW - MW, a negative biometric relationship ($b < 1$) was observed.

Ramesha and Thippeswamy (2009) described the Allometry and condition index of freshwater bivalve *Parreysia corrugata* (Muller) inhabiting the river Kempuhole, tributary of river Nethravathi, in the Western Ghats from April 2005 to May 2006. The length-breadth and length-width relationships for the entire study period were $L = 2.314 + 0.585B$ and $L = 1.907 + 0.333W$ respectively. The b values of length-breadth and length-width relationships varied from 0.5518 (March 2006) to 0.6519 (August 2005) and 0.2986 (July 2005) to 0.3735 (August 2005) respectively. The relationships between length-total weight, length-wet weight, length-shell weight and length-dry weight were $W = 0.0003428L^{2.777}$, $W = 0.00004197L^{2.885}$, $W = 0.0001738L^{2.802}$ and $W = 0.00001127L^{2.832}$ respectively. The monthly equilibrium constant values of length-total weight relationship ranged from 2.606 (July 2005) to 2.945 (August 2005), whereas for length-shell weight relationship, the values ranged from 2.691 (May 2006) to 3.1 (August 2005). The best values fluctuated between 2.781 (June 2005) and 3.58 (July 2005) for length-wet weight, while the values for length-dry weight fluctuated between 2.665 (April 2006) and 3.668 (October 2005). The monthly values of equilibrium constant indicated the relative growth in body weight of *P. corrugata.*

Malathi and Thippeswamy (2011) studied the Allometry and condition index in the freshwater mussel *Parreysia corrugata* (Müller, 1774) from river Malthi in the Western Ghats, India from March 2007 to June 2008. They reported the length-breadth and length-width relationships for the entire study period were $L = 2.0944 + 0.603B$ and $L = 1.4318 + 0.4322W$ respectively. The monthly *b* values of length-breadth relationship varied from 0.5472 (March 2008) to 0.6513 (May 2007) whereas for length-width relationship, the values ranged from 0.3858 (February 2008) to 0.4681 (May 2007). The length -total weight, length-wet weight, length-shell weight and length-dry weight relationships were $W=0.0007L^{2.6660}$, $W= 0.0001L^{2.6684}$, $W=0.0003L^{2.7124}$ and $W=0.0001L^{2.9370}$ respectively. The monthly equilibrium constant values of length-total weight relationship ranged from 2.2712 (June 2008) to 2.8682 (May 2007) whereas the *b* values of length-shell weight relationship varied between 2.3759 (May 2007) and 2.9452 (January 2008). The *b values* fluctuated between 1.1655 (Nov 2007) and 2.9639 (December 2007) for length-wet weight, while the values for length-dry weight ranged from 2.2107 (May 2007) to 3.3155 (April 2008). The monthly values of equilibrium constant indicated the relative growth in body weight.

Obirikorang et al. (2013) studied the length-weight relationship of the threatened freshwater clam, *Galatea paradoxa* (Born 1778) from the Volta Estuary, Ghana over a two-year period, from March 2008 to February 2010, to aid in the development of stock assessment models for the sustainable management of the remaining clam stock. Data reported in their study were collected at monthly intervals and covered varying depths ofthe Estuary ranging from 0.5 to about 10m. Overall, a total of 5276 clams with shell lengths ranging from 3.40 to 89.24mm and total weight from 0.10 to 154.00g were sampled during the study period. The length-weight relationships were highly significant ($p<0.0001$) for all the months and the b-values ranged from 2.023(January 2010) to3.874 (June2009).The calculated b-values indicated that clams exhibited different growth patterns at different periods but overall, the pooled samples of5276 individuals exhibited anisometric growth pattern (b═3.003). The observed monthly growth patterns exhibited by *G. paradoxa* appeared to be largely influenced by the reproductive cycle of the organism. During the periods leading to spawning, the clams generally exhibited positive allometric growth patterns (weight increasing faster than length) which appeared to be strongly linked to the build-up of proteins and carbohydrates in their tissues. Successive negative allometric growth patterns (length increasing faster relative to weight) were, however, observed from March to June 2008 and from December 2009 to February 2010, which are possibly indicative of the loss in tissue weight that occurs as a direct result of the spawning process. It will thus be suitable to institute a close season to coincide with the spawning period of the clams to avoid the harvesting of clams during the spawning period which will enhance future recruitment of the clam stock.

Singh et al. (2013) studied on the allometry of the eared horse mussel *Modiolus auriculatus* from a beach called Byndoor beach along the Karnataka coast, south west coast of India. They calculated regression equations between breadth-length and width-length for the entire study period were $B = 0.6503 + 0.4026\ L$ and $W = -0.1014 + 0.4260\ L$ respectively. The observed values of breadth and width against their respective lengths revealed linear relationships. The monthly *b* values of breadth-length and width-length relationships ranged from 0.3039 (October 2010) to 0.4593 (July 2010) and 0.3579 (January 2011) to 0.4626 (May 2011) respectively. The data on shell weight-length relationship for the entire period ($SW = 0.000058L^{2.9260}$) showed a non-linear pattern. The coeffcient of allometry (*b*) values ranged from 2.3022 (October 2010) to 3.1196 (May 2011) for length-shell weight relationship.

Suryawanshi and Kulkarani (2014a) studied the comparative length - weight relationship of fresh water mussels, *Parreysia corrugata* and *Lamellidens marginalis* from Nanded region, Maharashtra, India. They recorded 4.5 cm to 6.7 cm length and 10.98 g to 46.98 g in weight for *Parreysia corrugata*. And 3.5 cm to 7.3 cm in length and 4.71 g to 36.35 g in weight for *Lamellidens marginalis*. Relationship of length-weight of *Parreysia corrugata* was $W= 0.0972\ L^{3.2631}$ and the length-weight relationship of *Lamellidens marginalis* was $W=0.2008\ L^{2.6084}$. Comparative study of length-weight relationship indicates that value of exponent is more in *Parreysia corrugate* and both the specimen follow the Cube- Law.

Suryawanshi and Kulkarani (2014b) studied the length-weight, length-height, and height-weight relationship of *Parresia corrugata* from Nanded Region, Maharashtra, India. They reported 4.6 to 6.7 cm in length; and height from 2.6 to 5.3 cm; and weight from 10.98 to 46.98 g of the mussel. Relationship between length-weight was $W=0.0002375L^{3.5489}$; length-height was $H=0.4644L^{1.1956}$; and weight-height was $W = 0.1673L^{2.9680}$.

Thejasvi et al. (2014) discussed the morphometry and length-weight relationships of *P. viridis* inhabiting a subtidal rocky shore of Karwar coast of India from May 2009 to May 2010 by collecting the mussel samples from the sampling site during the low tide at monthly intervals. They focused on relationships between length-breadth, length-width and length-weights (length-total weight, length-wet tissue weight, length-shell weight and length-dry tissue weight). Calculated regression equations of length-breadth and length-width relationships for the entire study period were $B = 1.1192 + 0.5124L$ and $W= 0.7504 + 0.3019L$ respectively. The equations of length-total weight, length-wet weight, length-shell weight and length-dry weight relationships were $W = 0.00019L^{2.7745}$, $w = 0.00004L^{2.8040}$, $w =0.00005L^{2.9354}$ and $w = 0.00002L^{2.5765}$ respectively. The relationships between length-breadth and length-width are seen to be of the linear type and that between length-weights follow the non-linear pattern for the entire period of 12 months in *P. viridis* collected from Karwar, Karnatak.

Ramesha and Sophia (2015) studied the allometry and condition index of the freshwater mussel *Parreysia favidens* (Benson, 1862) during April 2005 and May 2006 inhabiting the west flowing river Seeta at Seetanadi in the Western Ghats, India. They reported the length-breadth and length-width relationships for the entire study period were $L = 1.540 + 0.6742B$ and $L = -0.0873 + 0.4561W$ respectively. The length-total weight, length-wet weight, length-shell weight and length-dry weight relationships were $W = 0.0003805L^{3.066}$, $W = 0.0000255L^{3.054}$, $W = 0.0000705L^{3.139}$ and $W = 0.0000042L^{3.110}$ respectively. The values of *b* indicated the relative growth in body weight and superior physiological condition of the mussels.

3. Condition index

Thippeswamy and Joseph (1988) reported the variations in the condition of *Donax incarnatus* (Gmelin) inhabiting Panambur beach sand were followed for one year. The highest condition indices were in March (9.4) and September (8.0~) and the lowest in January (3.5). Seasonal fluctuations in condition were probably related to cycles of gonadal growth and spawning. Based on the data, the best period for commercial exploitation appears to be during March and September.

Hemchandra and Thippeswamy (2008) reported that the monthly mean values of the condition index of green mussel *Perna viridis* (L.) varied from 5.17 (December 2004) to 7.76 (November 2005). The variation in condition index followed the breeding period and seasons. The maximum condition index (22–24) was recoded in May 2005. Based on the data on condition, it is suggested that the ideal period for commercial exploitation of *P. viridis* from the Island is from March to August, when the meat yield is the highest.

Ramesha and Thippeswamy (2009) described the Allometry and condition index of freshwater bivalve *Parreysia corrugata* (Muller, 1774) inhabiting the river Kempuhole, tributary of river Nethravathi, in the Western Ghats from April 2005 to May 2006. The values of condition indices showed significant fluctuations. The highest condition index (15.1) was recorded during April 2005, whereas the lowest (4.4) was in January 2006. The data indicated that the condition of mussel was fairly good from April to August 2005. The best time for commercial exploitation appears to be during this period. The condition index showed decreasing trend from September onwards and reached the lowest value during January and then the condition gradually increased. Seasonal fluctuation of condition in *P. corrugata* was probably, related to reproductive activity.

Malathi and Thippeswamy (2011) studied the Allometry and condition index in the freshwater mussel *Parreysia corrugata* (Müller, 1774) from river Malthi in the Western Ghats, India from March 2007 to June 2008. They reported that the monthly mean data on condition index showed significant fluctuation. The maximum (20.05) condition was noticed during May 2007 and the minimum (2.14) was in November 2007. The condition of mussels was fairly high from March to October and decreased from November to February.

Rupendra (2013) described the condition indices of *Perna viridis* from Mandvi shore in Ratnagiri district of Maharashtra state were collected and lengthwise grouped as small, medium and large. Different methods were used to determine condition index of the mussels using Ci-volume, Ci-shell and Ci-body. The results showed that the mussels from Mandvi shore irrespective of the size grouping almost showed equal values of Ci when any of the method was used, however, calculation of Ci - volume gave higher values for all the groups.

Ramesha and Sophia (2015) studied the condition index of the freshwater mussel *Parreysia favidens* (Benson, 1862). They reported maximum (16.36) condition during August 2005 and the minimum (4.16) in December 2005. The condition of mussels was fairly high from during May to September. The best period for harvest of *P. favidens* in river Seeta could be between May and September.

4. Reproduction

Abraham (1953) found that *Meretrix casta* from Adayar estuary spawns actively from July to August, October to November and March to April. He also suggested that spawning period does not remain constant and show year to year variation in the same environment.

Bachelet (1980) reported in *Macoma balthica* that gonad development started in June, as soon as spawning was completed. Recovery of condition continued throughout the summer months so that mature gametes were again present in late October. He also reported that analysis of data for individual years revealed some variations from one year to another.

Howard et al. (1983) studied the of the gametogenic cycle of the oyster *Ostrea iridescens* using histology in Costa Rica, Central America. Reproductive activity occurred continuously throughout the year, ripeness was detected at the end of the dry season, and spawning started at the beginning of the rainy season when salinity decreased from 32 to 29 PSU. Gonadal development was asynchronous and parallel in both sexes. The sex ratio was approximately 3:1 (312 males, 101 females and 57 indeterminate), and sexes were generally separate, though around 2% hermaphrodites were encountered.

Shpigel (1989) studied the gametogenic cycle of *Ostrea edulis* and *Crassostrea gigas* in the outflow water of fish ponds in Eilat, Israel. Despite relatively high water temperatures (14–28°C) and high salinity (41±2 ppt), both species developed gonads. *O. edulis* spawned from March to May, when water temperature ranged from 18 to 22°C, with a dormant period during the summer months. *C. gigas* spawned between May and August, when water temperature ranged between 20 and 26°C, with a dormant period between August and April.

Shaffee et al. (1991) reported the aspects of gametogenesis of the carpet-shell clam, *Ruditapes decussatus (L.)*, in two lagoons on the Atlantic coast of north Morocco. The clams sampled did not show any sign of hermaphroditism and the percentage of males was 42%. The gonads started developing in mid-winter and were ripe in spring. Successive spawning and reconstitution of gametes look place simultaneously starting from May up to the end of September. Two major spawning periods were noticed: a partial one in May-June and a complete spawning in August-September. From October to December the gonads were at resting stages.

Gist et al. (1997) evaluated the effect of temperature and trophy on zebra mussel reproduction. The annual reproductive cycle of *Dreissena polymorpha* was determined by histological examination of gonads. Populations from Lake Erie, its tributaries, and the Ohio River were compared. Gonadal development of Lake Erie commenced overwinter. Mussels from Lake Erie spawned only once during a season but approximately 30% of mussels from Lake Erie tributaries had an additional spawn. In contrast, complete gonadal regression was not seen in the Ohio River population. Spawning in the river populations was estimated to occur from March through September. Temperature appeared to regulate the onset of major reproductive events, but food must have played a role in determining the frequency of reproduction.

Pouvreau et al. (2000) studied the gametogenic cycle and reproductive effort of the tropical blacklip pearl oyster, *Pinctada margaritifera* (Bivalvia: Pteriidae), cultivated in Takapoto atoll (French Polynesia). They reported that Pearl oysters attained sexual maturity in the end of their first year (height » 40 mm), implying that *P. margaritifera* is a late-maturing species in comparison with other Pteriidae. This species was also confirmed to be a marked protandrous successive hermaphrodite in culture, with 100 % of males at first maturity and 75 % in older pearl oyster (height> 120 mm). The general pattern of gametogenic activity, fairly synchronous in both sexes, was comparable with that of other tropical bivalves: reproduction occurs continuously throughout the year with a maximal activity during the warm season (November–May). No resting period was observed. Quantitative growth data showed that *P. margaritifera* exhibits an annual synchronised polymodal spawning pattern, with two spawning peaks. They showed that, in a fairly stable tropical environment such as the Takapoto lagoon, *P. margaritifera* is a multiple spawner, which uses an opportunistic reproductive strategy, allowing investment, all year around, of any surplus energy into gamete production. Surplus energy is ensured by the high pumping rates developed by this non-symbiotic bivalve to succeed in low seston conditions.

Chung et al. (2001) investigated the gonadal development and gametogenic cycle of the equilateral venus, Gomphina collected in the coastal waters of Gangneung, Gangwon-do, Korea by means of histological method. Sex of the clam was dioecious. The gonad index (GI) was reached the maximum in June (3.87), and the minimum in August (0.66). The condition index (CI) was reached the maximum in May (0.051), and the minimum in January (0.028). The gametogenic cycle of the clam could be divided into five stages inactive (September to November), early active (November to March), late active (April to May), ripe (June and July) and spent/degenerative stage (July to August). Sex ratio (male/female) was 1:0.9.

Juhel et al. (2003) investigated gametogenic cycle of three size classes of the freshwater mussel *Dreissena polymorpha* by histological examination at Lough Derg Co. Limerick, Ireland. They observed gonads and gamete development in mussels in all the months and in all the sizes of mussel. Only one period of successful reproduction existed. Variations in the sex ratio were observed with a shift from female dominance in September to equal proportions of male and female in October. Gamete development in males appeared to be more dependent on external factors, such as water temperature.

Cek and Sereflisan (2006) observed the seasonal reproductive cycle, gonad structure and sex ratio of the freshwater mussel *U. terminalis*. A total of 695 individuals were collected in monthly samples from October to September in Gölbaşı Lake, Turkey. Calculation of the GSI and histological examination of the gonads showed that gametogenesis began in winter, and spawning occurred in summer and early autumn. Five stages of oogenesis and five stages of spermatogenesis were identified. Spermatogenesis followed a pattern similar to that of typical oogenesis. Although sex ratios of *U. terminalis* were not significantly different from the expected 1:1 ratio ($P>0.05$), slightly female-biased sex ratios were recorded.

Mohite and Mohite (2009) studied gonadal development in the shortneck clam *Paphia malabarica* in relation to hydrographic parameters in Kalbadevi estuary. They reported an extended spawning period from September to January and maturation observed from April and the clams reached the ripe stage during May-June. Cytolysis in the gametes was noticed by the end of January. Only one spawning peak was observed followed by the regression of gonads. Minimum and maximum condition indices were observed in February and September, respectively. From February, most individuals were either in spent or in a short gonadal resting stage. Gonadal development initiated from March onwards. Active gametogenesis started from April to July. The sudden drop in salinity and temperature in September appears to act as stimuli for the initiation of spawning.

Misra et al. (2010) revealed that the freshwater bivalve *Lamellidens marginalis* is a prolific breeder. The degree and number of spawning phase differ significantly from one population to another. For development of a simple and practical approach of induced spawning the study was attempted with selected physical treatments viz., heat, cold, alternate heat and cold shocks, and chemical treatments viz., aqueous solution of H2O2, NaOH, Tris Buffer and NH4OH. The physical treatments were found to be better in terms of spawning, higher larval survival and longer duration of larval viability compared to the chemical stimulus.

Uddin et al (2012) investigated the reproductive effort of the Manila clam *Ruditapes philippinarum* during different gametogenic stages with combining histology and immune assay techniques (ELISA). At the Begmiri tidal flat in Incheon Bay off the west coast of Korea, clams commenced gametogenesis in February and the first spawning female was observed in July. Clams continued spawn up to October. The GSI increased rapidly from April (4.3) to May (16.8), peaked in July (20.6), then dropped dramatically from August (14.0) to September (5.5), indicating that clams have a major spawning pulse during August and September.

Gaikwad and Kamble (2013) analysed the annual reproductive cycle of *L. marginalis* and *Parreysia corrugata* from Panchaganga River Maharashatra, Indiaduring June, 2010 to May, 2011. They reported 1:1 sex ratio for both of the species and continuous gonadal development of the bivalves.

Mondol et al. (2016b) studied length-weight relationship, condition index and sex ratio of freshwater mussel *Lamellidens corrianus* (Lea, 1834) from a freshwater lake, northwest Bangladesh and reported the overall male to female sex ratio was 1: 0.92 and did not differ significantly from the expected 1: 1 ratio.

Asaduzzaman et al. (2019) investigated the reproductive biology and ecology of the green mussel (*Perna viridis*) by interlinking among ecological factors, ingested gut plankton, gonad fatty acid profile, and reproductive traits. Mussels were collected throughout the year from the coastal region of the Bay of Bengal of Bangladesh, and the histological analysis of 242 mussels revealed five stages of gametogenesis with an annual spawning season from January to April. The gonadosomatic index showed a strong correlation with the water quality parameters, ingested gut plankton groups, and gonadal fatty acids, and also displayed prominent effects of seasonality. High salinity, nutrients, chlorophyll *a*, and plankton abundance in the water column were positively correlated with gonad development and the spawning of mussels.

Niogee et al. (2019) investigated the ovarian cycle of freshwater pearl mussel, *Lamellidens marginalis* (Lamarck, 1819) that was collected monthly from a rearing pond in Bangladesh. The overall male-female sex ratio, 1:1.08 did not deviate significantly from the 1:1 ratio as expected (χ^2-test). *Lamellidens marginalis* females exhibited a prolonged spawning period from February to November when water temperature remains above 22 °C. Four distinct peaks were noted during October, February, June and August when most of the females were ripe and ready for spawning. Condition indices (CIs) ranged from 0.42 ± 0.23 (September) to 0.85 ± 0.25 (October) by mean (± SD) exhibiting five peaks over 12-month study period.

5. Biochemical composition

Seasonal variations in biochemical composition of bivalves have been evaluated for their nutritive potentials (Gabbott and Bayne, 1973; Ruiz et al., 1992; Dridi et al., 2007).

Patil (2011) studied on the protein changes in different tissues of freshwater bivalve *Parreysia cylindrica* and observed (28.65 to 68.91mg/100 mg of dry tissue).Significant change in overall protein content in mantle, foot, gills, digestive glands and whole body tissue of *Parreysia cylindrica* was noticed due to acute and chronic exposure to pesticide indoxa carb. The depletion was maximum in digestive glands than in mantle, gills, foot and whole body tissue.

Jadhav and Gulave (2012) stated that variation in the protein content in soft body tissues of *Lamellidens marginalis* were collected from Jayakwadi dam, at Paithan near Aurangabad. As changes in the environmental conditions, it showed an effect on protein contents in the tissues like, mantle, hepatopancreas, gonad and foot. Protein content found maximum in gonads throughout all the three seasons, whereas mantle shows minimum values of protein. There are great fluctuations in the values of protein during different seasons.

Shetty et al. (2013) studied on the seasonal changes in the biochemical composition of freshwater bivalves, *Parreysia* sp. Protein was found maximum during summer season (60.8%) and was found minimum during winter season (40.5%) of dry tissue weight. There is great fluctuation in the values of Carbohydrate (glycogen) present in the body tissues during different seasons. During summer season, maximum carbohydrate (40%) was found, whereas during monsoon and winter, minimum carbohydrate (14.12%) was found. Similarly, lipid was found maximum (8.2%) during summer season and was minimum (6.8%) during winter season. This shows the mobilization of biochemical constituents in body tissues during different seasons. The bivalve mollusks shows maximum variation of biochemical constituents as it undergoes different stages like development, maturation and spawning during different seasons.

Pandey et al. (2014) studied the biochemical properties of *Lamellidens marginalis* from Harike wet lands area, Ferozpur district, Punjab. They collected and cultivated the specimens for six months (June to November, Shell length 4.3-9.0cm) in pond environment and studied for variations in the biochemical composition. The nutritional contents of the whole body tissues of *L. marginalis* was found to be, moisture (82.54 to 85.58%), proteins (4.74% to 8.11%), lipid (2.43% to 2.95%), ash (2.30% to 2.65%) and glycogen content (3.72% to 4.10%) of wet tissue weight. They reported that protein and lipid contents were significantly higher in large adult mussels compared to small young mussels where as glycogen content was significantly higher in small mussels compared to large adult mussels. The biochemical composition of *L. marginalis* meat having high nutrients (protein, lipid and glycogen up to 8.11%, 2.95% and 4.10% respectively) justifies the edible quality of these mussels by humans and also as a substitute of fish meal in fish feed and poultry feed.

Periyasamy et al. (2014) studied on the marine mollusks have been given more importance, because they have both ecological and economically importance to mankind. Bivalve mollusks comprise major marine fishery resources; they have rich in biochemical compounds. The proximate composition of protein, carbohydrate, lipid, amino acids and fatty acids were studied. The results of proximate composition in *D. incarnatus* showed the percentage of protein was high 23.51%, followed by the carbohydrate 10.23% and lipid 1.34 %. The total essential amino acids were found to be as 58.21 % and nonessential amino acids were 35.4 % in body tissue.

Ranajit et al. (2014) studied on the seasonal variation of carbohydrate, protein and lipid of common fresh water gastropod *Bellamya bengalensis* in nine selected study sites of Medinipur. For biochemical analysis, shell of the gastropods were removed, flesh were dried with filter paper to avoid the outer water content. The study revealed that protein was much higher than carbohydrate & lipid (mg %) in all nine study sites in all seasons. Protein (17.20 ± 1.78) carbohydrate (13.12 ± 0.74) and lipid (4.83 ± 0.017) were the maximum during Pre-monsoon period than to Monsoon & Post Monsoon period. Protein, carbohydrate, lipid were the maximum during Pre-monsoon period than monsoon & post-monsoon period.

Sable and Vedpathak (2017) stated that variations in organic constituents in different soft body parts of *Lamellidens marginalis* found in Godavari River near Aurangabad. As environmental condition changes, it shows an effect on protein content in the tissues like mantle, hepatopancreas, foot and gonad. Protein was found maximum in gonads viz. 11.270 to 11.320 mg/100mg during summer, 8.810 to 8.954 mg/100mg during monsoon and 6.754 to 7.002 mg/100mg during winter season.

CHAPTER III

Seasonal variation of environmental parameters in the study area, the northwest Bangladesh

Abstract

The aim of this study was to investigate the seasonal variation of water quality parameters of the study site. During this study water temperature of the study site was found to vary from 18.5 to 34.5 ^{0}C. The highest temperature was recorded in August and the lowest temperature was recorded in January. The transparency was ranged from 19.8 cm to 31.2 cm. Lower values of transparency were during March to July indicates a better food availability of the study area during this period. During the study pH ranged from 6.6 to 7.8. Dissolved oxygen of the study area showed some seasonal fluctuations and ranged from 4.8 to 7.0 mg/l. Free carbon dioxide ranged from 4.3 to 6.8 mg/l and total alkalinity ranged from 65 to 95 mg/l.

Key words: Temperature, transparency, pH, DO, free $Co_{2,}$ total alkalinity.

1. Introduction

The water quality parameters of water are of great importance for fish culture and fisheries management. It plays the most important role in governing the production of planktonic organisms or primary production in fish ponds (Banerjee, 1967). Quality of aquatic environments in turn depends on a greater extent on the environmental characteristics of a particular geographical region and it is an important scientific tool to understand the pond dynamics as well as to maximize the fish production (Knud-Hansen et al., 1998). Climate change has a major influence on water quality (Boyd, 1990), as aquatic animals are more dependent on water quality parameters for growth and development (Nikolsky, 1963). The overall productivity of a water body can easily be deduced from its primary productivity, which forms the backbone of the aquatic food chain (Ahmed and Singh, 1989). Scientific management of the water bodies are closely related to the acquisition of knowledge of the environmental factors which affect the aquatic community as well as the fish production (Rounsefell and Everhart, 1962). The present study was designed to investigate the seasonal variation of water quality parameters of the study site in Rajshahi, northwest Bangladesh.

2. Materials and Methods

2.1 Study area and duration

The study was conducted in the freshwater lake situated in the Rajshahi University Campus, the northern Bangladesh. The geographical location of the study site is 24º 22´ 21.56´´ N and 88º 38´ 11.16´´E (Fig. 3). Total area of the lake is about 2.5 ha and the average depth is about 3.0 meter. There is a continuous water source in the lake with inlet and outlet system. The management of this lake is controlled by the Department of Fisheries, University of Rajshahi. The water quality parameters of the study area were monitored monthly during January, 2014 to July, 2015.

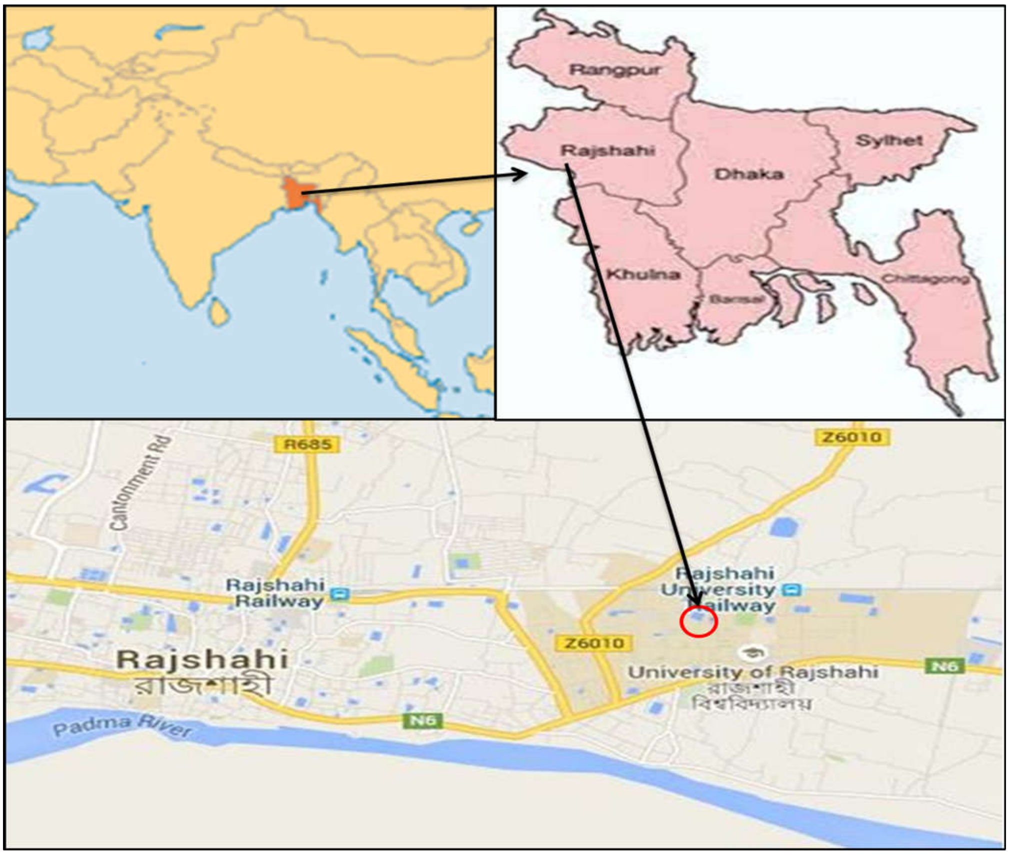

Fig. 3. Map showing the study area at Rajshahi, the northwest Bangladesh (red circle indicating the study site).

2.2 Water quality parameters

The water quality parameters such as temperature, transparency, dissolved oxygen (DO), carbon dioxide (CO_2), pH and alkalinity of the study area were monitored monthly using the following methods-

2.2.1 Water temperature

Water temperature was recorded on the spot with the help of a digital thermometer dipping at the depth of 20 to 30 cm below the surface water and expressed in (^0C).

2.2.2 Transparency

Transparency was recorded by Secchi disk. The value of transparency was expressed in cm.

2.2.3 Hydrogen ion concentration (pH)

The negative logarithm of hydrogen ion concentration of the study area was recorded by pH meter.

2.2.4 Dissolved oxygen

Dissolved oxygen of the pond water was recorded with the help of a water testing kit (HACH kit, FF-2, USA). The concentration of dissolved oxygen was expressed in milligram per liter (ppm) of water.

2.2.5 Free carbon dioxide

Free carbon dioxide (CO_2) of the pond water was recorded with the help of a water testing kit (HACH kit, FF-2, USA). The concentration of free carbon dioxide (CO_2) was expressed in milligram per liter (ppm) of water.

2.2.6 Total alkalinity

Total alkalinity was measured by a water testing kit (HACH kit, FF-2, USA). The value of total alkalinity was expressed in milligram per liter (ppm) of water.

3. Results

The water quality parameters measured during the study period were within the optimum range and suitable for fish as well as bivalve growth. During the study the water temperature (^{0}C), water transparency, pH, dissolve oxygen (ppm), free carbon dioxide (ppm), and total alkalinity (ppm) showed monthly variations (Fig. 4-9).

3.1. Water temperature

In the present study the water temperature ranged from 17.4 to 34.5°C. At the beginning of the study in January, 2014 the water temperature of the study site was 18.5°C, increased dramatically at reaching the highest 34.5°C in August. Water temperature then decreased dramatically at reaching the lowest level 17.4°C in the month of January, 2015 and then again increased again at reaching 33.2 °C in July, 2015 (Fig. 4).

3.2. Transparency

During the study the transparency ranged from 19.8 cm to 31.2 cm. Transparency showed several monthly fluctuations, with lower values during March to July (Fig. 5).

3.3. Hydrogen ion concentration (pH)

In the present study the pH was ranged from 6.6 to 7.8 and there were very little monthly fluctuations with alkaline pH in most of the months (Fig. 6).

3.4. Dissolved oxygen

Dissolved oxygen of the study area showed some seasonal fluctuations and it was ranged from 4.8 to 7.0 ppm (Fig. 7). Except in the month of January, 2014 the DO level of the study area was more than 5.0 ppm.

3.5 .Free carbon dioxide

During the study the free carbon dioxide ranged from 4.3 to 6.8 ppm and showed several monthly fluctuations with lower value during March to August and higher value during September and October (Fig. 8).

3.6 Total alkalinity

Total alkalinity ranged from 65 to 95 ppm in the study area during this study period

(Fig. 9). At the start of the study the alkalinity was 78.0 ppm, it gradually increased to 95.0 mg/l in the month of June, 2014. From June, 2014 the alkalinity gradually declined at reaching the lowest level (65.0 ppm) in the month of January, 2015 and then again increased at reaching the second highest level (92.0 ppm) in the month of June, 2015.

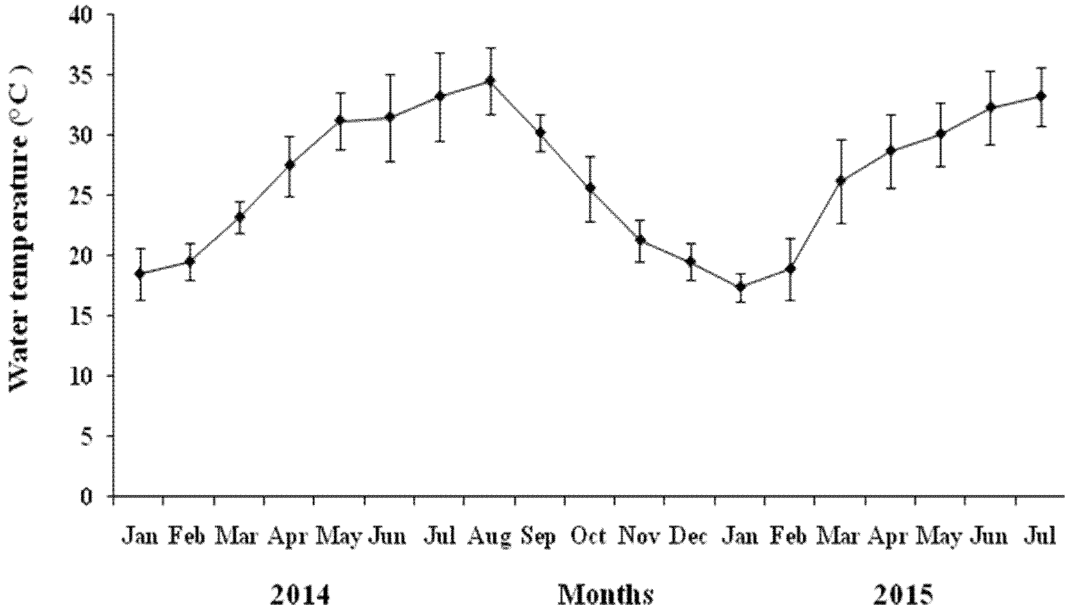

Fig. 4. Monthly variations (mean±SE) of water temperature (°C) in the study area during January, 2014 to July, 2015. Vertical bars represent standard error.

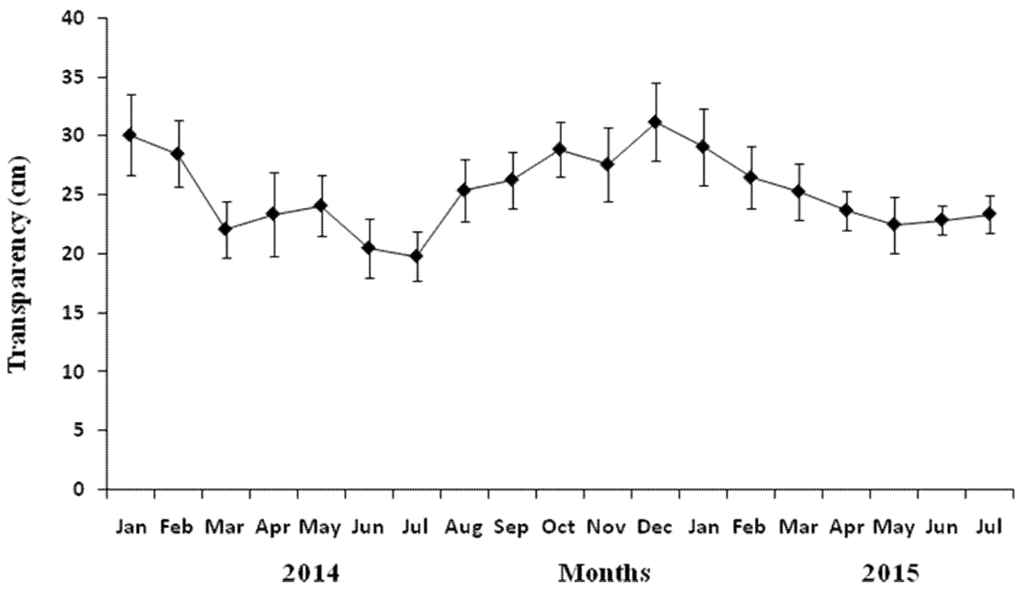

Fig.5. Monthly variations (mean±SE) of transparency (cm) in the study area during January, 2014 to July, 2015. Vertical bars represent standard error.

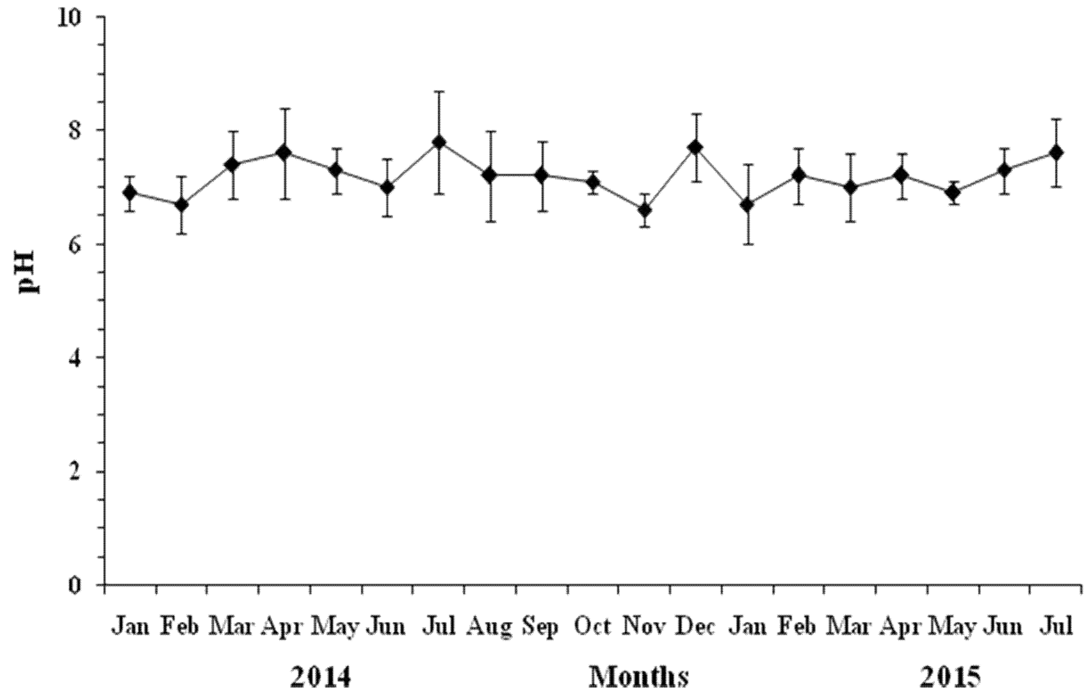

Fig.6. Monthly variations (mean±SE) of hydrogen ion concentration (pH) in the study area January, 2014 to July, 2015. Vertical bars represent standard error.

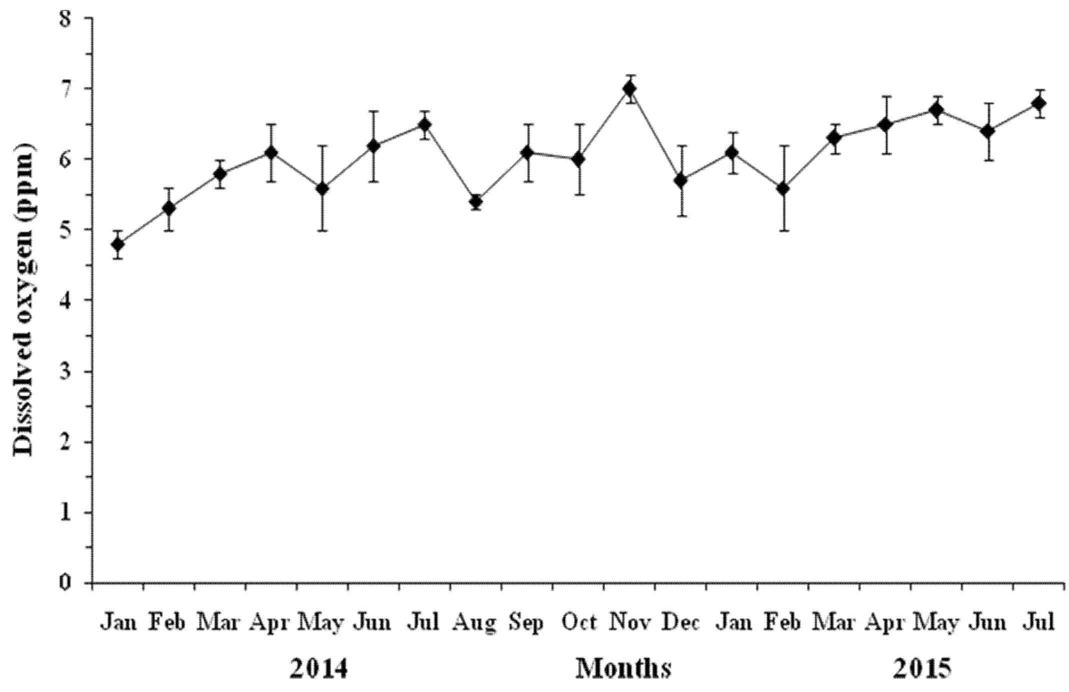

Fig.7. Monthly variations (mean±SE) of dissolved oxygen (ppm) in the study area during January, 2014 to July, 2015. Vertical bars represent standard error.

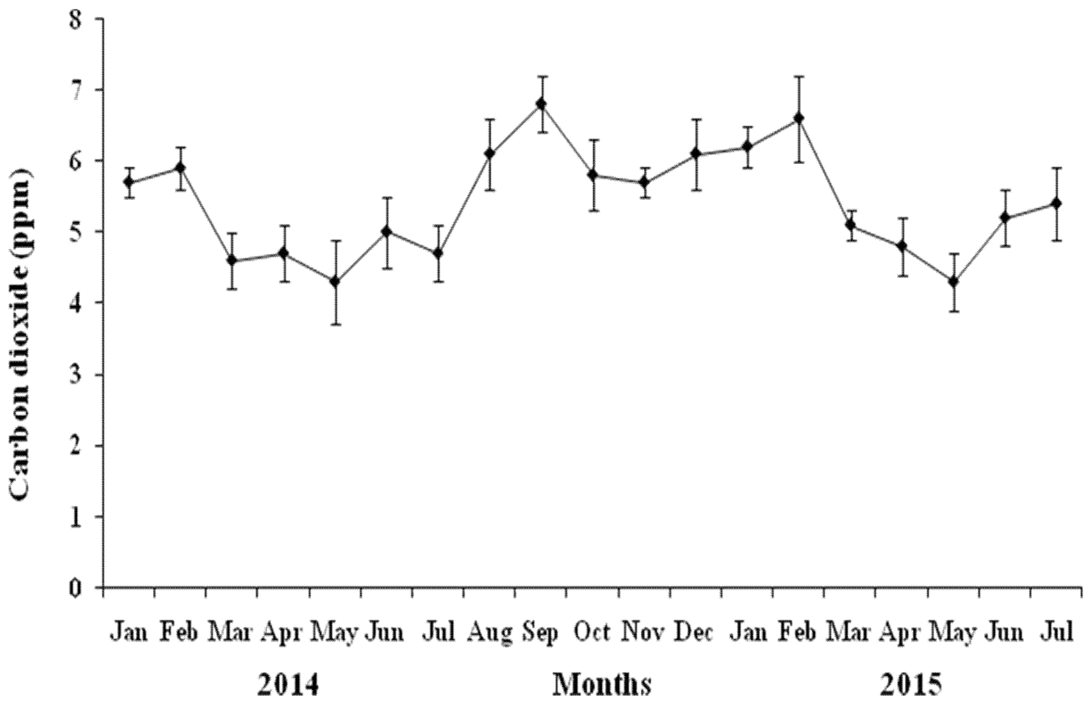

Fig. 8. Monthly variations (mean±SE) of free carbon dioxide (ppm) in the study area during January, 2014 to July, 2015. Vertical bars represent standard error.

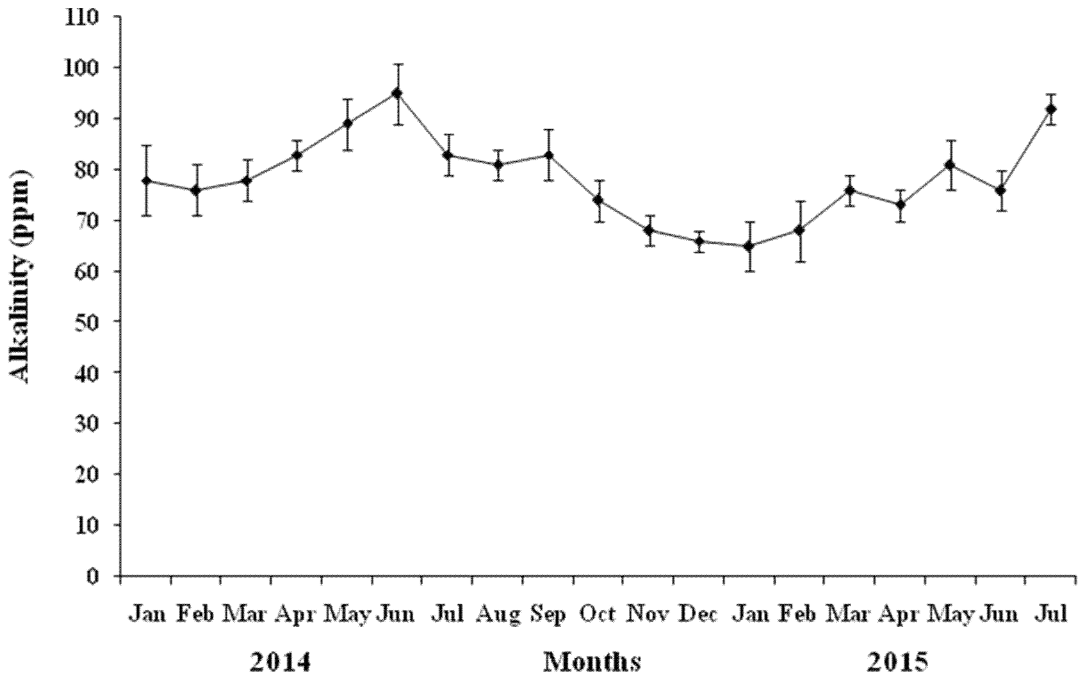

Fig. 9. Monthly variations (mean±SE) of total alkalinity (ppm) in the study area during January, 2014 to July, 2015. Vertical bars represent standard error.

4. Discussion

4.1 Water temperature (°C)

Growth, reproduction and other biological activities of aquatic animals are influenced by water temperature. Temperature has a powerful effect on growth, both through its influence on rates of biochemical processes and hence metabolism, and through associated factors such as feed intake (Brett, 1979). In 2014, the water temperature of the study site was found to vary from 18.5 to 34.5 ^{0}C. The highest temperature was recorded in the month August, 2014 and the lowest temperature was recorded in the month of January. Jasmine et al. (2013) reported 16-33.6 °C water temperature in the lentic ecosystem of Northwest Bangladesh, which are in agreement with findings of the present study. Dewan et al. (1991) recorded temperature 23.2 to 34.0 °C from the experimental ponds situated in Bangladesh Agricultural University Campus, Mymensingh.

4.2 Water transparency (cm)

It also expresses as the level of primary productivity of a water body. Secchi disc reading indicates the productivity of the water body. Secchi disc reading and productivity are inversely related, higher the secchi disc reading lower the productivity and lower the secchi disc reading higher the productivity of the water body. Secchi disc reading ranged from 20 to 30 cm indicates the waterbody productive (Boyd, 1990). During the study period the water transparency varied from 19.8 to 28.8 cm, indicates a good productive condition of the study site.

4.3 Hydrogen ion concentration (pH)

Hydrogen ion concentration (pH) is considered as an important chemical factor in fish culture. pH indicates the acidity and alkalinity condition of a water body. It is also called the productivity index of water body. The circum neutral pH or slightly alkaline pH is most suitable for fish culture. An acidic pH of water reduces the growth rate, metabolic rate and other physiological activities of fishes (Swingle, 1957). During the study period, the pH was ranged from 6.6 to 7.7, indicating a better condition of the study site. From the same geographic area Jasmine et al. (2013) also reported the pH 5.2-7.5, which are more or less similar with the result of the present study. DoF (1996) stated that the range of pH water body suitable for fish culture would be 6.5 to 8.5. Swingle (1967) found a good relationship between pH of pond water and fish culture and he stated that pH 6.5 to 9.0 is suitable for aquatic life. pH value in alkaline condition in water was supposed to be helpful for proper growth and development of fishes and aquatic organisms (Nikolsky, 1963; Jhingran, 1991).

4.4 Dissolved oxygen (ppm)

Dissolved oxygen is the most important chemical factor for all aquatic organisms. The slight fluctuation in DO value might be due to alteration in the rate of photosynthesis in the pond and due to the rate of DO consumption by the fish species through respiration. During the study period the dissolved oxygen varied from 4.8 to 7.0 mg/l. Alikunhi (1957) considered 5.0 to 7.0 ppm of dissolved oxygen content of water to be fair or good in respect of productivity and water having dissolved oxygen below 5 mg/l to be unproductive. Swingle (1967) stated that the concentration of dissolved oxygen below 5 ppm were undesirable in fishponds. DoF (1996) reported that the range of a suitable DO for water body for fish culture would be 5-8 ppm. Jasmine et al. (2013) reported 5.0-7.5 ppm DO from the lentic ecosystem of Northwest Bangladesh.

4.5. Free carbon dioxide (ppm)

The concentration of free carbon dioxide was found directly related to the amount and nature of biological activities in the water. The carbon dioxide, pH and alkalinity are thus directly related with each other (Michael, 1969). During the present study the values of free Carbon dioxide were fluctuated between 4.3 to 6.8 mg/l. Suitable range of free CO_2 is not more than 12 mg/l (DoF, 2015). According to Boyd (1990) the suitable range of free CO_2 for fish culture is less than 10.0 ppm. Therefore, it can be concluded that the amount of carbon dioxide in the study site was within the production range.

4.6. Total alkalinity (ppm)

During the study period the value of alkalinity ranged from 65 to 95 mg/l. According to Alikunhi (1957) total alkalinity more than 100 ppm should be present in highly productive water bodies. According to Boyd (1990) total alkalinity should be more than 20 ppm in fertilized ponds. Rath (2000) stated that calcareous water with alkalinity more than 50 ppm is most productive. He also described the range of alkalinity 0-20 ppm as low productive, 20-40 ppm as medium productive and 40-90 ppm as high productive. Therefore, it might be concluded that this range was within the suitable range for bivalve growth. Jasmine et al. (2013) has reported 68-100 ppm total alkalinity from the lentic ecosystem of Northwest Bangladesh, which are in agreement with the findings of the present study.

CHAPTER IV

Growth pattern of the freshwater bivalve mollusk *Lamellidens marginalis* (Lamarck, 1819) (Bivalvia: Unionidae) from the northwest Bangladesh

Abstract

Growth, length-length and length-weight relationships of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) inhabiting the freshwater habitat in Rajshahi, northwest Bangladesh were studied during January, 2014 and July, 2015. Monthly sampling was carried out and a total of 620 bivalves were collected for the study. The length-breadth and length-width relationships for the entire study period were Br=0.179 + 0.475L and Wi=0.196+ 0.3163L respectively. The monthly *b* values of length-breadth relationship varied from 0.3466 (October 2014) to 0.8132 (July 214) whereas for length-width relationship the values ranged from 0.2495 (February 2014) to 0.3982 (August 2014). The length-body weight, length-wet tissue weight, length-dry tissue weight, length-shell wet weight, length-dry shell weight relationships were $W=0.1241L^{2.9066}$, $W=0.0524L^{2.7377}$, $W=0.0116L^{2.7849}$, $W= 0.0533L^{2.9178}$ and $W=0.0391L^{3.0386}$ respectively. The monthly equilibrium constant values of length-weight relationships were 2.3373 (March, 2015) to 3.5011 (June, 2015) for length-body weight, between 1.8185 (July, 2014) and 3.6224 (May, 2014) for length-tissue wet weight, between 2.1091 (March, 2015) and 3.7399 (June, 2014) for length-shell weight.

Keywords: Freshwater mussel, *Lamellidens marginalis*, Growth Pattern, Morphometry, Allometry.

1. Introduction

Length–weight and other biometric relationships are important to the understanding of various aspects of a species *viz*., growth, ecology and physiology. These also allow life history and morphological comparisons between species or between populations of a species from different habitats/regions. Length-weight relationships are also useful in estimations of weight from length, conversions of growth-in-length to growth-in weight for predictions of weight at particular age and further use in stock assessment models (Pauly, 1983).

The morphometric relationships between length and weight can be used to assess the well-being of individuals and to determine possible differences between separate unit stocks of the same species (King, 1995). In addition, length-length relationships are also important in fisheries management for comparative growth studies. Allometry is most reliable and widely used method to study the growth pattern of bivalve. The allometric principle of animal morphology has long been recognised since Huxley and Tessier (1936) who proposed the concept of allometry. Allometry is the study of relationship between two measurable variables, or in its most general sense, allometry is the study of size and its consequences (Reiss, 1989). The allometric relationships are generally explained by shell growth along the three dimensional axis. The study of shell dimensions in mollusks is aimed at ascertaining the interrelationships in growth of body characters. The influence of proximate or mechanistic factors will cause differences in the intercept and slope of the allometry. In allometric length-weight relationship, the variation in equilibrium constant represents the growth in weight than that of length. Relatively large variation in meat content occurs in bivalve mollusks depending upon the variation in physiological condition and environmental variables (Wibur and Owen, 1964). The biology of freshwater mussels *viz*., *Parreysia* *corrugata* (Nagabhushanam and Lomte, 1971; Lomte and Jadhav, 1980) *Parreysia favidens* (Ramesha and Sophia, 2015) and *Lamellidens* spp. (Narain, 1972; Nagabhushanam and Lohgaonker, 1978; Moorthy et al., 1983) inhabiting the Indian freshwater habitats has been reported previously. However, information on growth pattern of freshwater mussels *L. marginalis* from Bangladesh is lacking. Even though *L. marginalis* is listed as least concern by IUCN (Budha and Daniel, 2010), they are potentially susceptible to a number of threats mainly fishing, in the present study locality. Freshwater mussels have important relationship with fish especially for reproduction. Other threats *viz.,* habitat destruction, poor water quality, siltation and agricultural run-off may act directly on the mussel population or have an indirect impact through decline in the host fish species that are required to complete the mussel's life cycle. Therefore, a reduction in the diversity or abundance of freshwater mussels can indicate a negative change in the ecosystem. Hence, the present investigation was undertaken to study the growth, length-weight and length-length relationship of *L. marginalis* inhabiting the freshwater ecosystem, in Rajshahi, the northwest Bangladesh.

2. Materials and methods

2.1 Study area and duration

The study was conducted in the freshwater lake situated at Rajshahi University Campus, the northern Bangladesh. The geographical location of the study site is 24º 22′ 21.56′′ N and 88º 38′ 11.16′′E (Fig. 3). Total area of the lake is about 2.5 ha and the average depth is about 3.0 meter. There is a continuous water source in the lake with inlet and outlet system. The management of this lake is controlled by the Department of Fisheries, University of Rajshahi. The fisheries diversity of this lake is rich and the freshwater mussel *Lamellidens marginalis* is abundantly available in this lake. The tribal people (Swantal community) in Rajshahi region occasionally harvest the mussel from this lake for their consumption. The present study was conducted in this study site during January, 2014 to July, 2015.

2.2 *Test animal: Lamellidens marginalis*

The bivalve as the name implies, possesses two valves (shells) lying on the right and left sides of the body. Bivalve's symmetry is a characteristic feature. Among the exploited freshwater molluskan resources, bivalves contribute to bulk of the catch. Among the freshwater bivalve, *Lamellidens marginalis* is an emerging resources for commercial exploitation. Hence, the *L. marginalis* was selected as a candidate species for the present work (Fig. 10).

Fig.10. Photographs of *Lamellidens marginalis* collected from the study site.

2.3 Sampling and biometric measurements

Monthly 30-50 specimens were collected by hand picking from the sampling site during January, 2014 to July, 2015. A total of 620 individuals of *L. marginalis* were collected during the study period and subjected to morphometric measurements. Shell length (maximum antero-posterior distance), breadth (maximum distance from hinge to ventral margin) and width or depth or thicknesses (maximum distance between outer edges of two valves) of individual organisms were measured accurately to 0.01 mm using Vernier calipers (Fig. 11). Total weight of individual mussels was taken. The mussels were dissected to remove the meat which was then blotted and weighed individually. The individual weight of shell was also determined (Fig.11). Both the meat and shell were dried at constant temperature of 60°C for 2 days and weighed accurately to 0.01 g using an electronic balance. Allometric examination was done for morphometric, length-breadth, length-width and length-weight (length-total weight, length-wet tissue weight, length-dry tissue weight, length-shell weight and length-shell dry weight) relationships (Pauly, 1983).

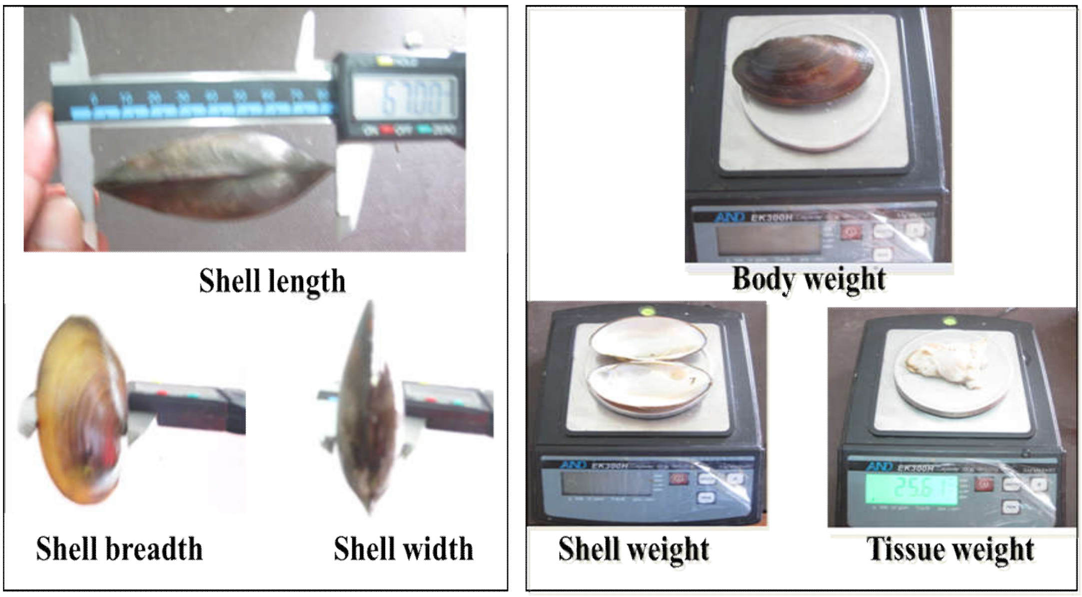

Fig.11. Lengths and weights measurements of *L. marginalis* collected from the study area.

2.4 Statistical analysis

Dimensional relationships was estimated using the linear regression equation (Y = a + bX) and the length-weight relationship by a non-linear regression equation ($W = aL^b$) in which, *a* (intercept) and *b* (slope / equilibrium constant) are constants. A *t*-test (H0, b = 3) was performed with a confidence level of 95% to confirm if the values of b obtained by linear regression were significantly different from the isometric value, expressed by the following equation *t*s = (b - 3) / Sb, where *t*s = *t* -test value, b = slope, Sb = standard error of the slope (b). Subsequently, comparison between the obtained value of *t* - test and the tabled critical value of *t* - test, allowed the determination of the statistical significance of the b value. To know the temporal variation of growth parameters a one way ANOVA with DUNCAN POSTHOC ranking was used. All the statistical analysis were performed on SPSS version 20 software at $P<0.05$ level.

3. Results

3.1 Length-frequency distribution

A total of 620 (male and female) specimens of *L. marginalis* were collected during this study. During the study the morphometric measurements, length-weight relationships were studied for combined sex. Frequency distribution (both length and body weight basis) for the whole collected specimens of *L. marginalis* illustrated in the Fig. 12 using histogram. Frequency distribution data showed that different size group and age group individuals are present in the study site, indicates a continuous recruitment of the *L. marginalis* population in the study area. Table 1 represents the overall descriptive statistics of the shell length, shell breadth, shell width, body weight, tissue wet weight, shell wet weight, tissue dry weight and shell dry weight of

L. marginalis. During the study minimum shell length was 3.65 cm, maximum shell length was 10.55 cm and mean shell length was 7.33 cm. On the other hand minimum body weight was 3.60 g, maximum body weight was 124.05 g and the mean body weight was 46.14 g. A one way ANOVA indicated a significant monthly variation of all length and weight measurements ($P<0.05$). Monthly variations of shell length, shell breadth, shell width, body weight, tissue wet weight, tissue dry weight, shell wet weight and shell dry weight are shown in Fig. 13-20.

3.2 Morphometric relationship

The data on morphometric relationships (length-breadth and length-width) for the entire period is shown in Fig. 21-22. This study data revealed that the length-length variables are linearly related. Further the data depicted that long individuals are wide (more height) and high (more thickness) and inversely the short individuals are narrow (less height) and low (less thickness).The calculated values of length-breadth and length-width relationships for the entire study period were $B = 0.179 + 0.475L$ and $W = 0.196 + 0.3163L$ respectively (Fig. 21-22). The monthly *b* values of length-breadth relationship varied from 0.3466 (October 2014) to 0.8132 (July 214) whereas for length-width relationship, the values ranged from 0.2495 (February 2014) to 0.3982 (August 2014) (Fig. 23-24). The linear relationships of length-breadth and length-width of *L. lamellidens* during the study period showed that the short individuals were of less height/thickness and inversely long individuals were wide/high.

3.3.3 Length-weight relationship

A non-linear relationship was noticed in all the measured length-weight relationships in all sizes. The nonlinear relationships of length-body weight, length-wet tissue weight, length-dry tissue weight, length-shell weight, length-dry shell weight were W

= $0.1241L^{2.8282}$, W = $0.0524L^{2.5905}$, W =$0.0116L^{2.6296}$, W = $0.0533L^{2.8769}$ and W = $0.0391L^{2.9979}$respectively for the entire study period (Table 2, Figure 25-29). All the length-weight relationships were highly significant ($P<0.05$). The monthly equilibrium constant values of length-weight relationships were 1.8037 (July, 2014) and 3.5011 (June, 2014) for length-total weight, between 1.8185 (July, 2014) and 3.6224 (May, 2014) for length-wet tissue weight, between 1.6247 (May 2014) and 3.7295 (July 2014) for length-dry tissue weight, between 2.1091(March, 2015) and 3.7399 (June, 2014) for length-shell weight and between 2.2617 (April, 2015) and 3.8662 (June, 2014) for length-shell dry weight (Fig. 30-34).

Table 1. Descriptive statistics of shell length, shell breadth, shell width, body weight, tissue wet weight, tissue dry weight, shell wet weight and shell dry weight of *L. marginalis* during the study period.

Measurements	***n***	**Minimum**	**Maximum**	**Mean (±SE)**
Shell length (cm)		3.65	10.55	7.33±0.06
Shell breadth (cm)		1.38	5.98	3.65±0.03
Shell width (cm)		1.21	3.64	2.52±0.03
Body weight (g)	620	4.70	124.05	46.30±0.99
Tissue wet weight (g)		1.30	46.43	13.66±0.30
Tissue dry weight (g)		0.33	11.21	3.33±0.07
Shell wet weight (g)		1.55	51.25	20.86±0.46
Shell dry weight (g)		1.00	50.10	19.67±0.45

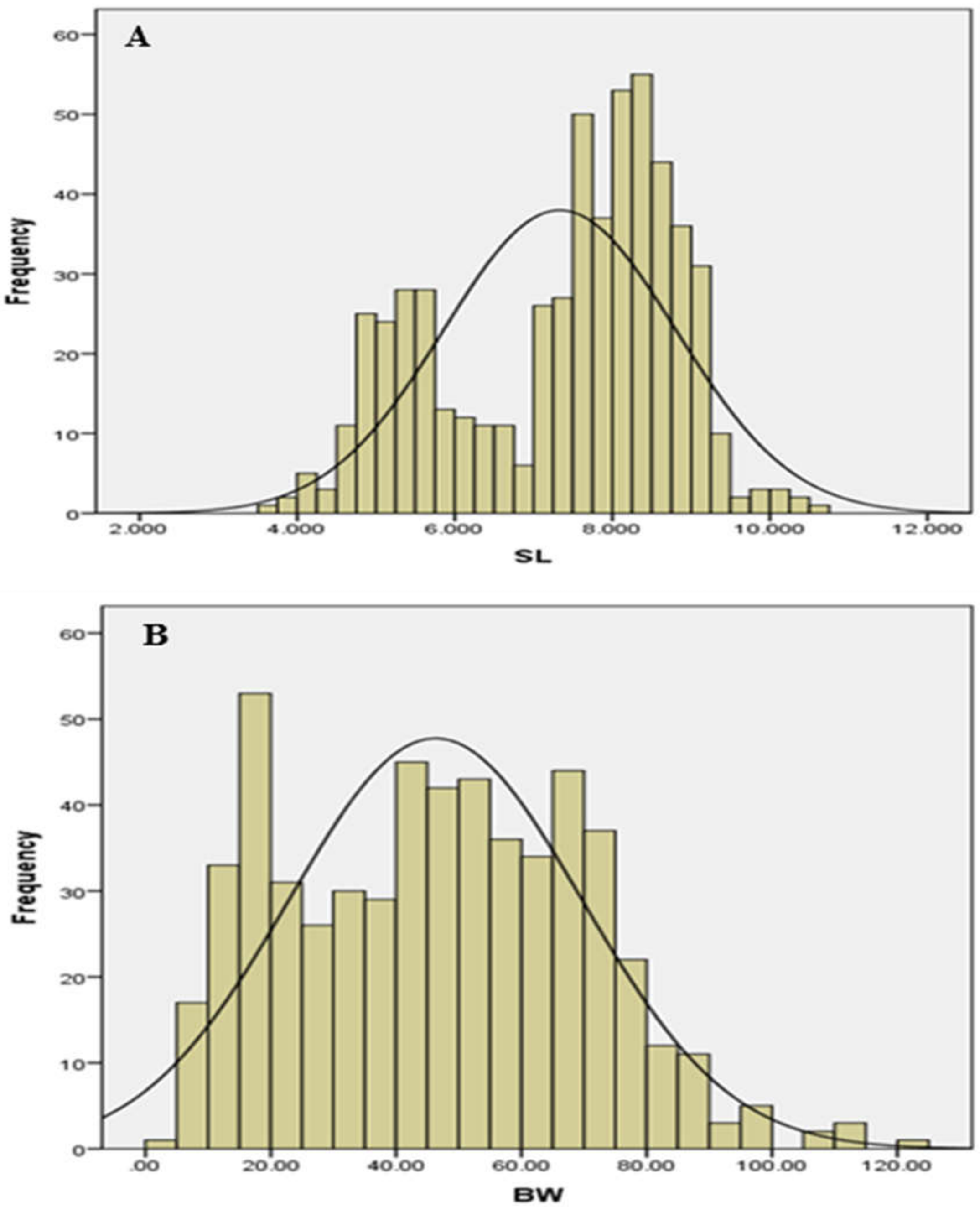

Fig. 12. Frequency distribution of length (A) and body weight (B) for combined sex of *L. marginalis* during the study period.

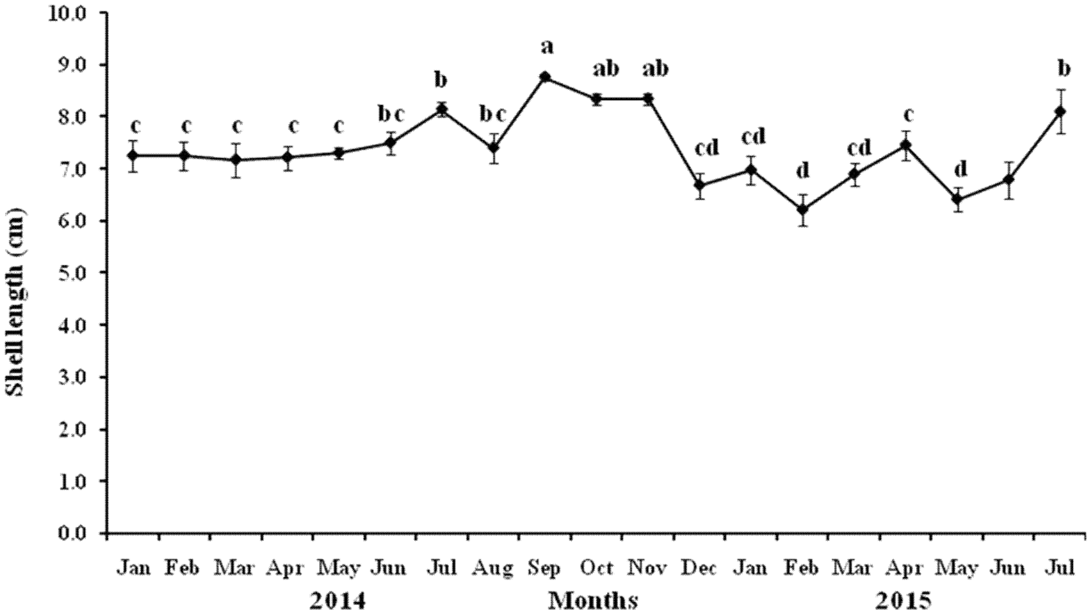

Fig. 13. Monthly variation of shell length (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

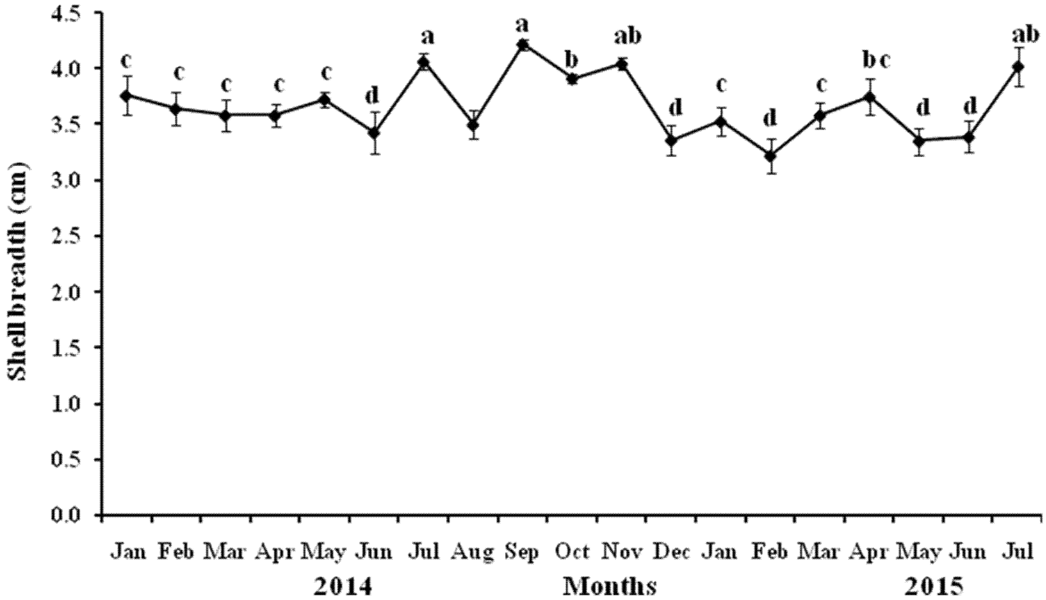

Fig. 14. Monthly variation of shell breadth (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

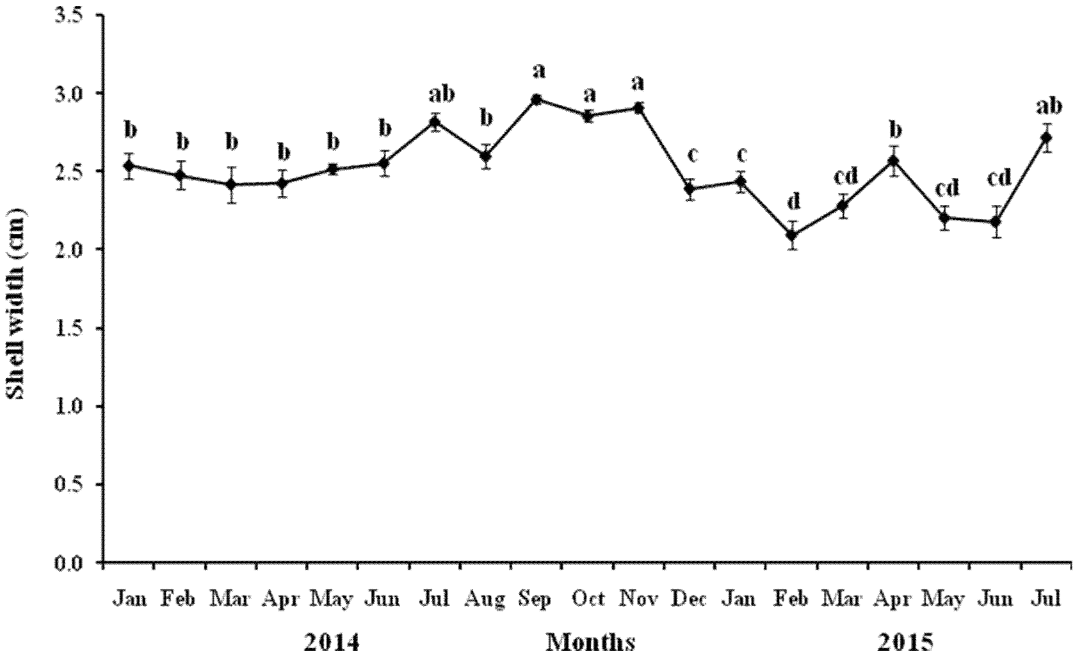

Fig. 15. Monthly variation of shell width (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

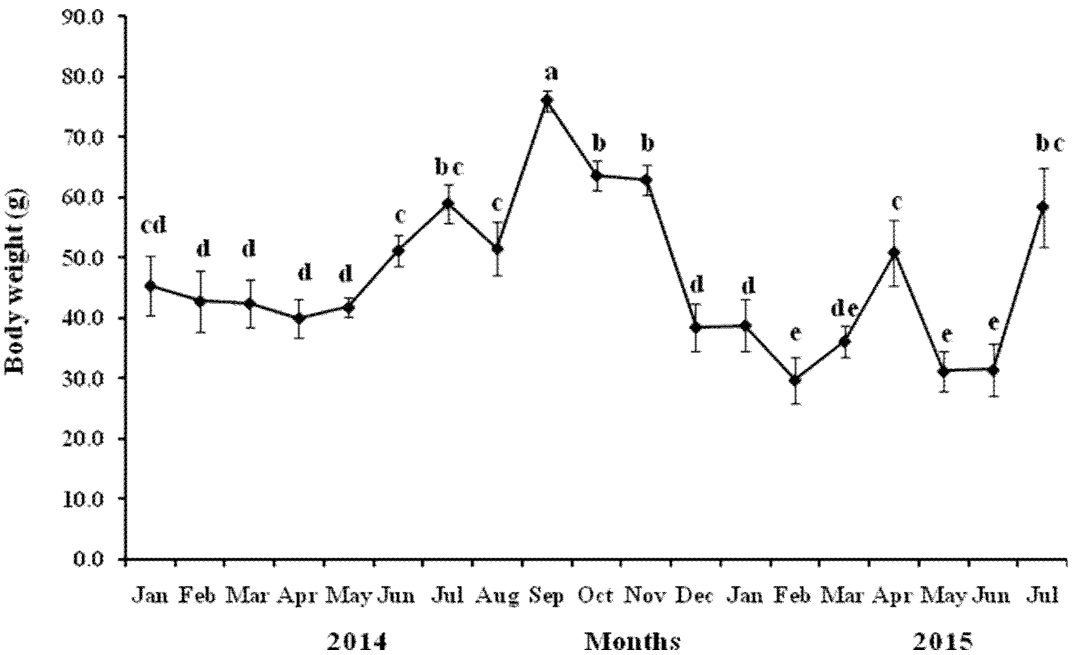

Fig.16. Monthly variation of body weight (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

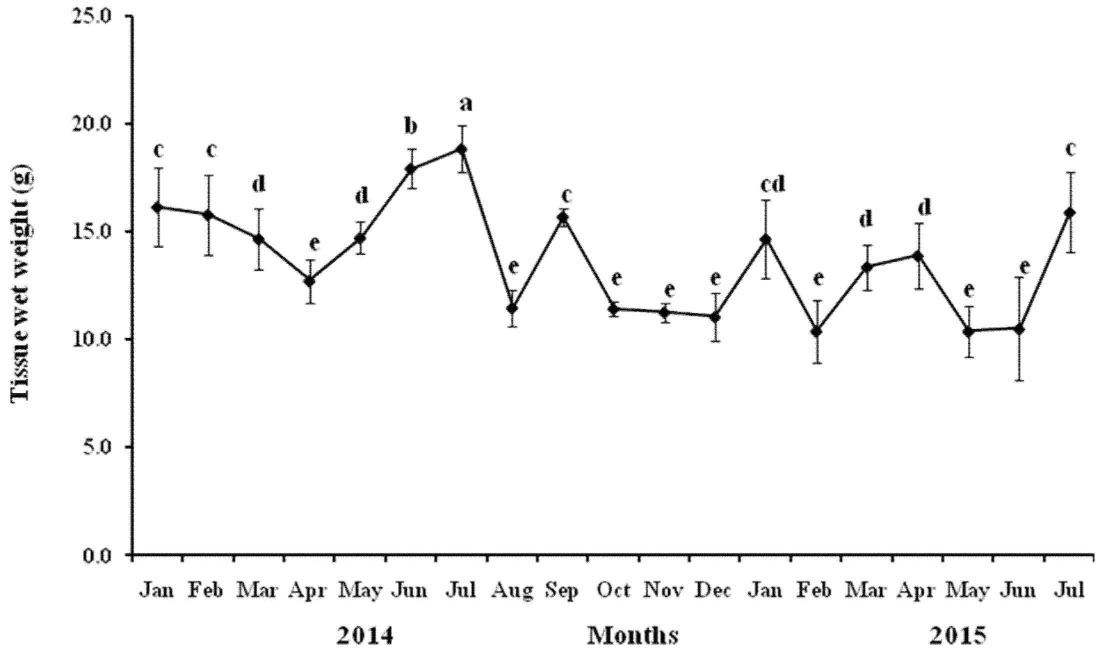

Fig.17. Monthly variation of tissue wet weight (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

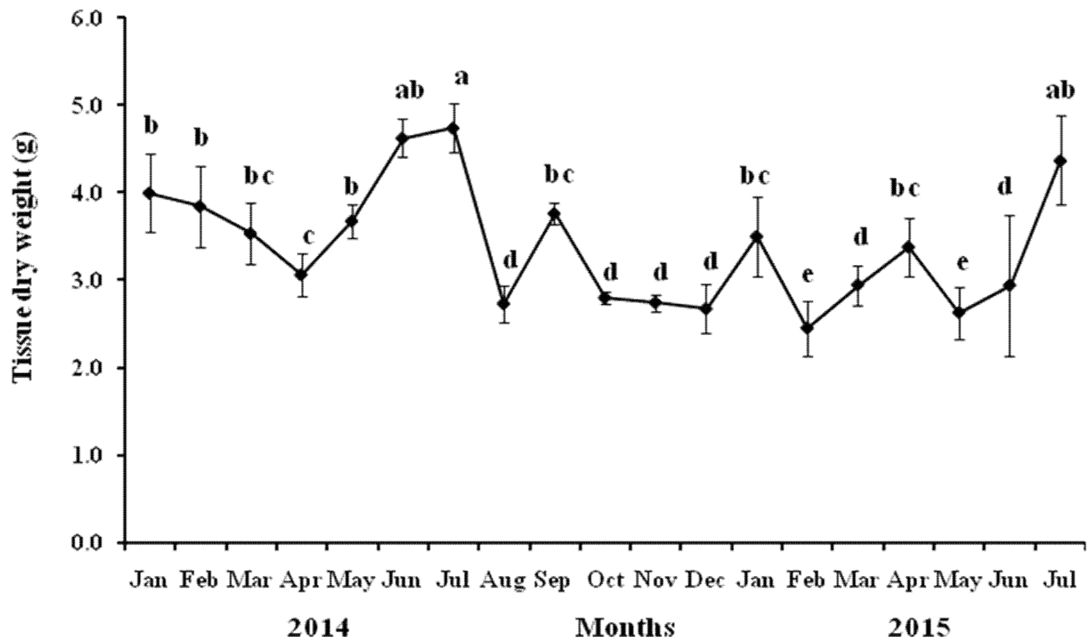

Fig.18. Monthly variation of tissue dry weight (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

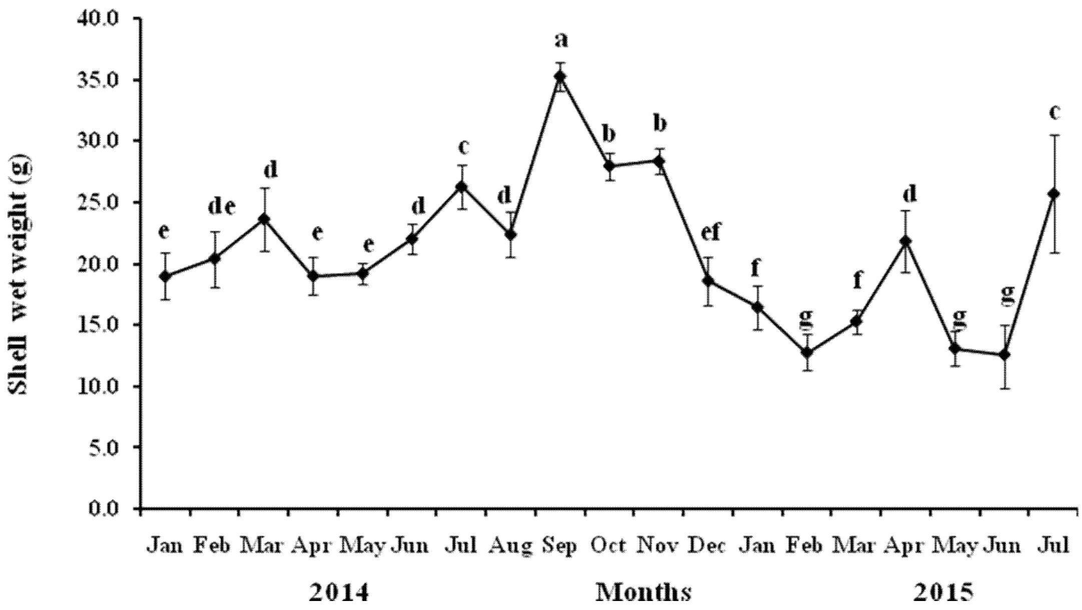

Fig.19. Monthly variation of shell wet weight (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

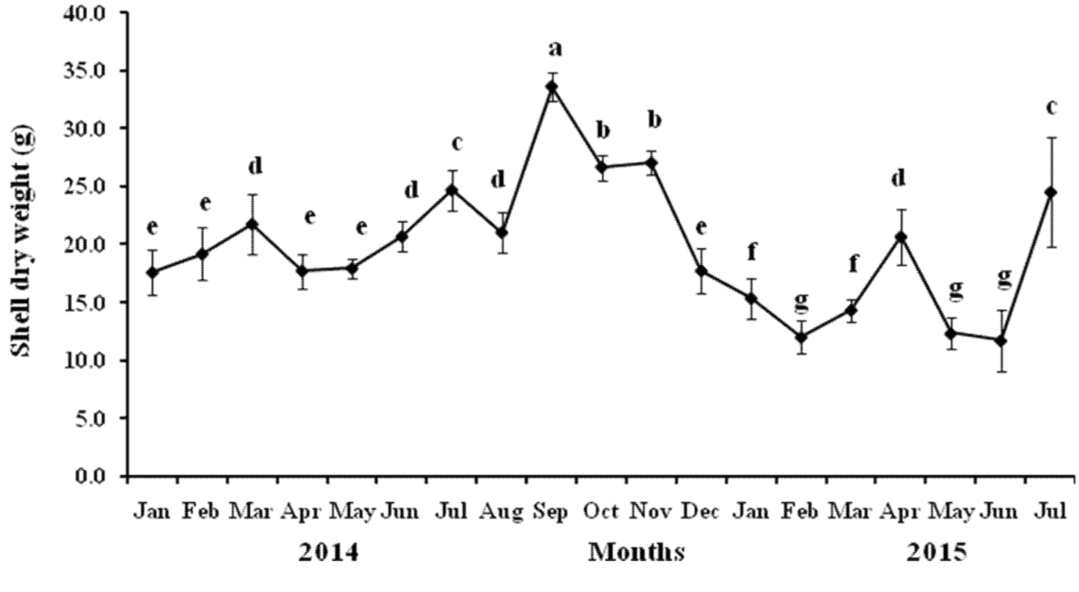

Fig. 20. Monthly variation of shell dry weight (mean±SE) of *L. marginalis* during the study period. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

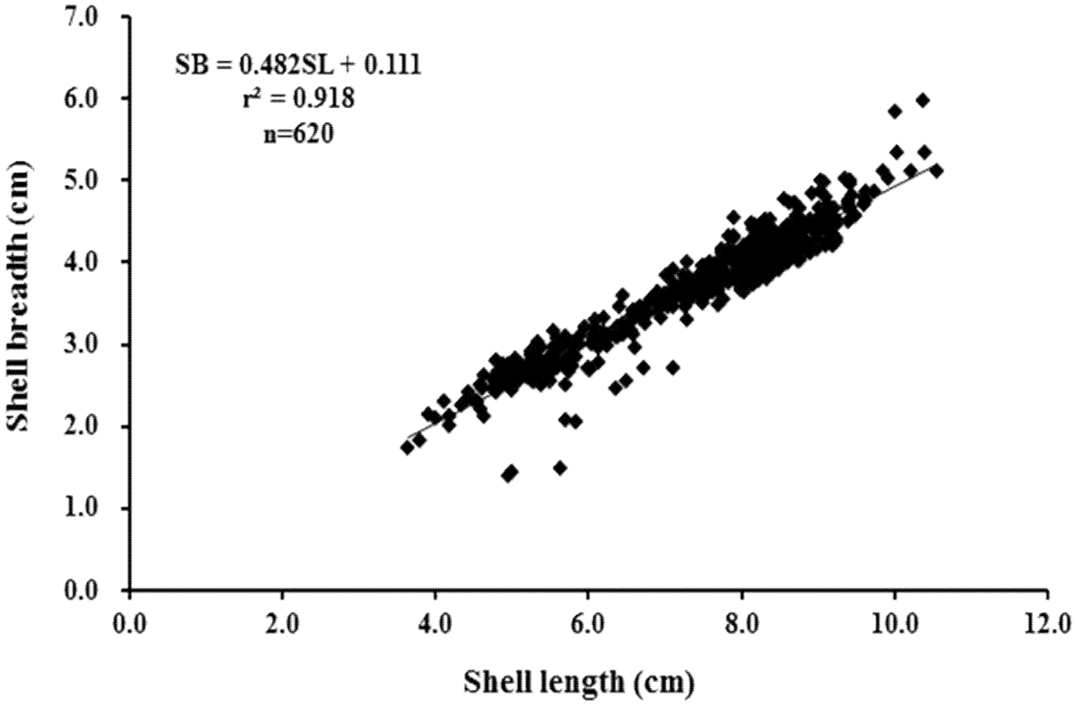

Fig. 21. Bivariate scatter diagram of length-breadth relationships of *L. marginalis* during the study period.

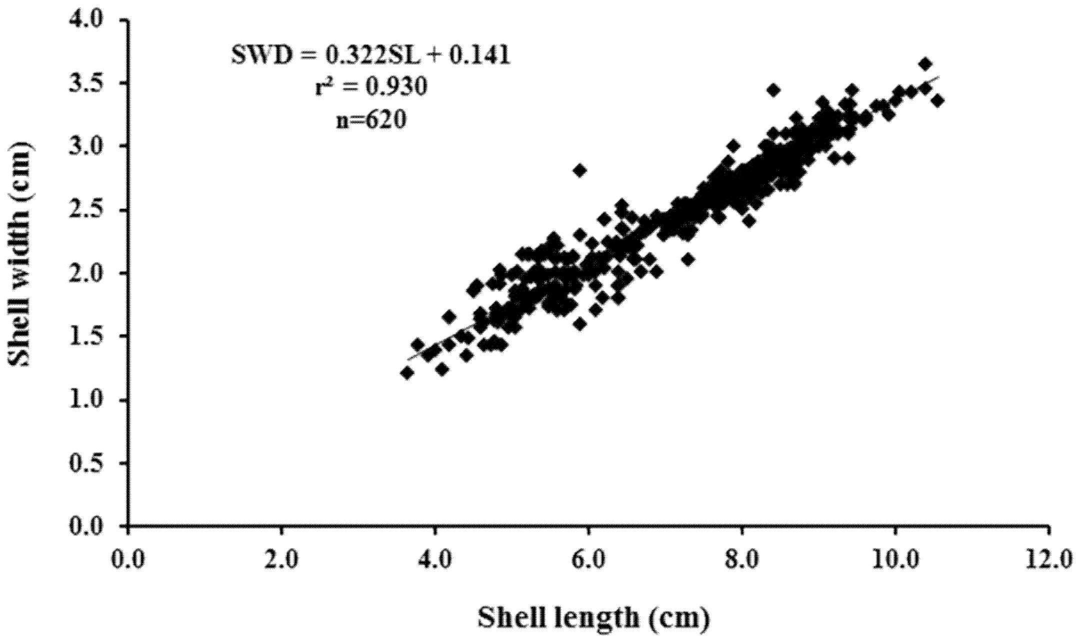

Fig. 22. Bivariate scatter diagram of length-width relationships of *L. marginalis* during the study period.

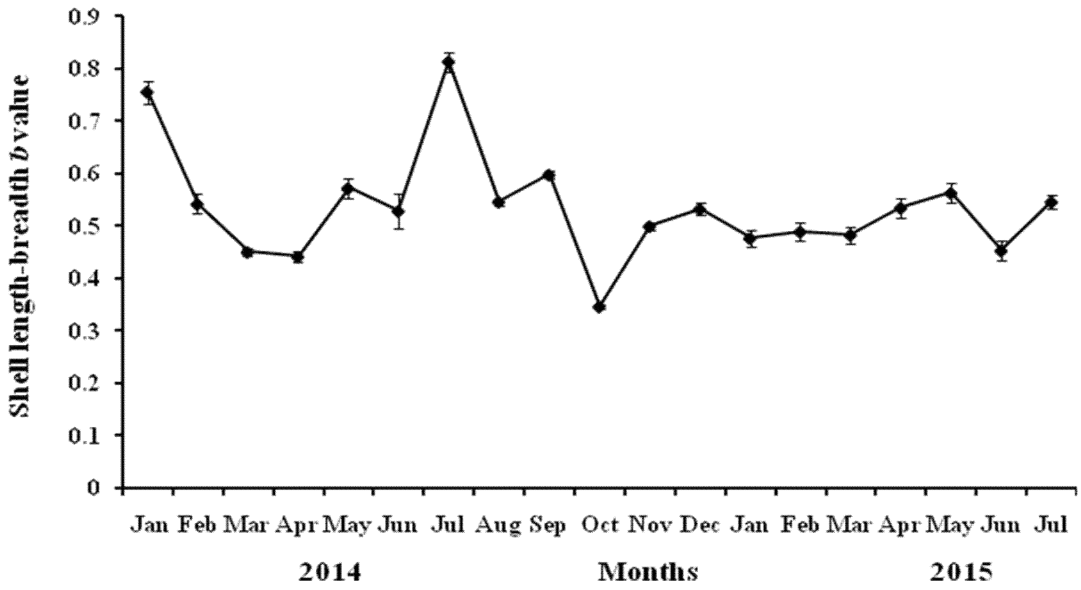

Fig. 23. Monthly variability in the *b* values of length-breadth relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

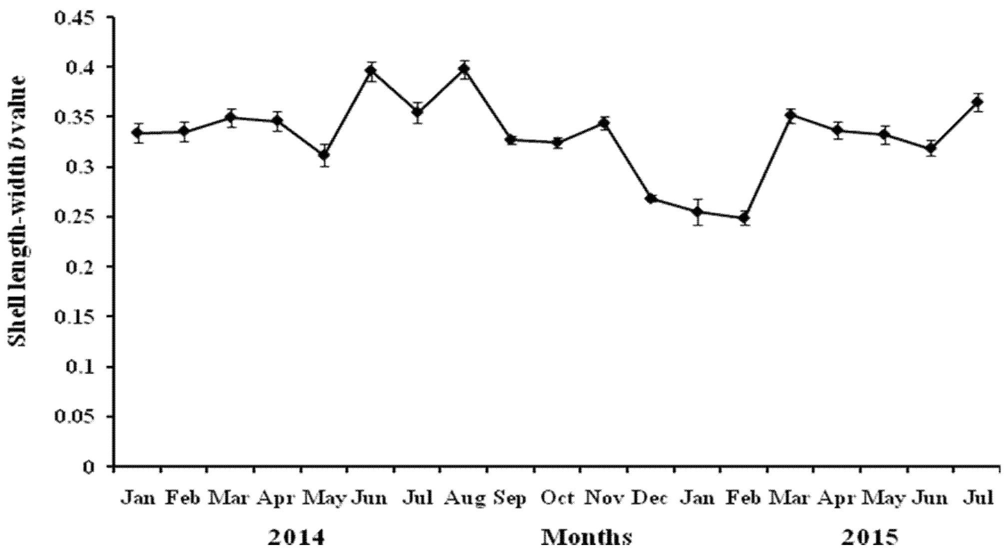

Fig. 24. Monthly variability in the *b* values of length-width relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

Table 3. Descriptive statistics and estimated regression parameter on shell length and body weight of *L. marginalis* during the study period. (I, Isometric; A+, Positive allometric and A-, Negative allometric growth).

Months	**n**	**Shell length**			**Body weight**			**Regression parameter**			**Growth type**
		Mean	**Min**	**Max**	**Mean**	**Min**	**Max**	***a***	***b***	***r2***	
Jan,14	30	7.26	4.78	10.39	45.35	9.32	114.65	0.089	3.0566	0.9644	I
Feb,14	30	7.25	4.42	9.85	42.76	7.33	112.4	0.072	3.1481	0.9586	A+
Mar,14	30	7.17	3.92	9.16	42.44	4.7	71.12	0.1649	2.715	0.9646	A-
Apr,14	40	7.22	4.62	10.22	39.98	11.41	105.12	0.2675	2.414	0.9837	A-
May,14	40	7.31	5.11	8.32	41.77	16.67	62.52	0.2711	2.5228	0.9554	A-
Jun,14	30	7.5	4.96	9.06	51.19	26.14	74.12	0.0368	3.5011	0.9765	A+
Jul, 14	30	8.15	6.52	9.42	58.96	26.85	90.12	0.3229	1.8037	0.9634	A-
Aug,14	30	7.41	4.2	8.91	51.46	5.92	77.55	0.520	2.2743	0.9544	A-
Sep,14	30	8.76	8.05	9.75	76.07	60.12	95.85	0.3533	2.4721	0.9668	A-
Oct,14	30	8.34	7.28	9.25	63.58	39.27	89.76	0.1175	2.9598	0.9354	I
Nov,14	30	8.35	7.27	9.25	62.88	39.27	89.76	0.0705	3.1928	0.9549	A+
Dec,14	30	6.68	4.2	9.92	38.48	5.92	83.54	0.0944	3.1254	0.9607	A+
Jan,15	30	6.98	4.85	10.55	38.76	9.32	110.9	0.1079	2.987	0.9639	I
Feb,15	30	6.21	3.65	9.1	29.73	5.86	72.51	0.1937	2.6895	0.956	A-
Mar,15	40	6.9	4.12	9.1	36.15	9.9	64.28	0.3698	2.3373	0.9561	A-
Apr, 15	50	7.46	5.3	10.4	50.88	12.82	124.05	0.082	3.1095	0.9531	A+
May,15	30	6.42	4.6	9.02	31.21	9.55	78.54	0.1265	2.9254	0.9523	I
Jun, 15	30	6.95	5.50	9.60	31.42	14.7	84.64	0.082	3.055	0.917	I
Jul, 15	30	8.10	6.45	9.60	58.42	23.90	110.65	0.027	3.628	0.947	A+

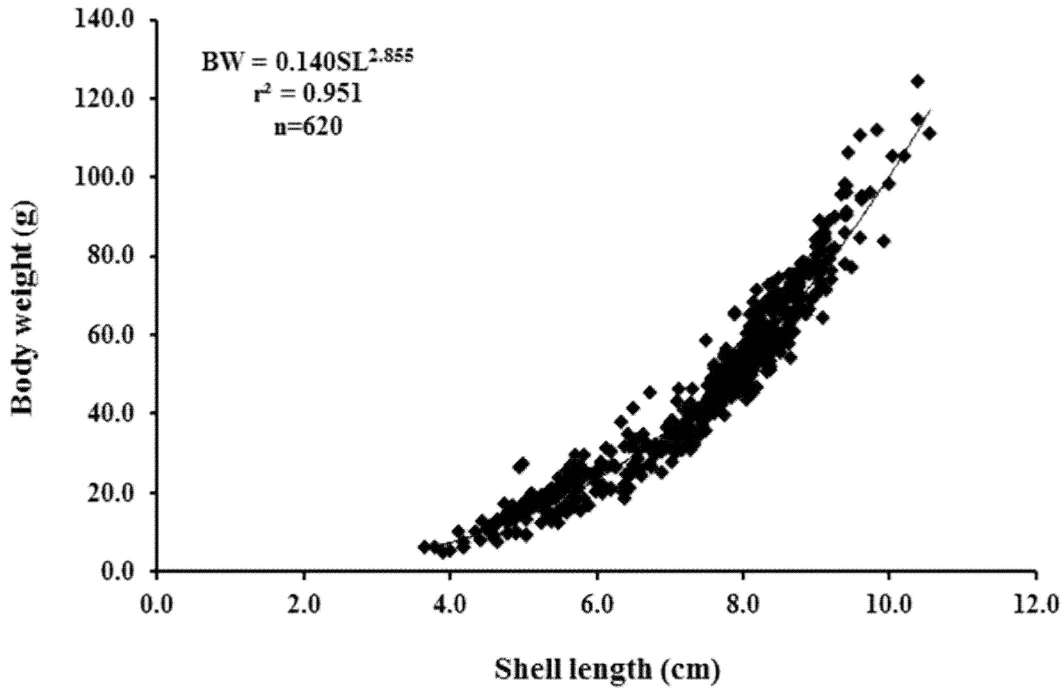

Fig. 25. Bivariate scatter diagram of length-body weight relationships of *L. marginalis* during the study period.

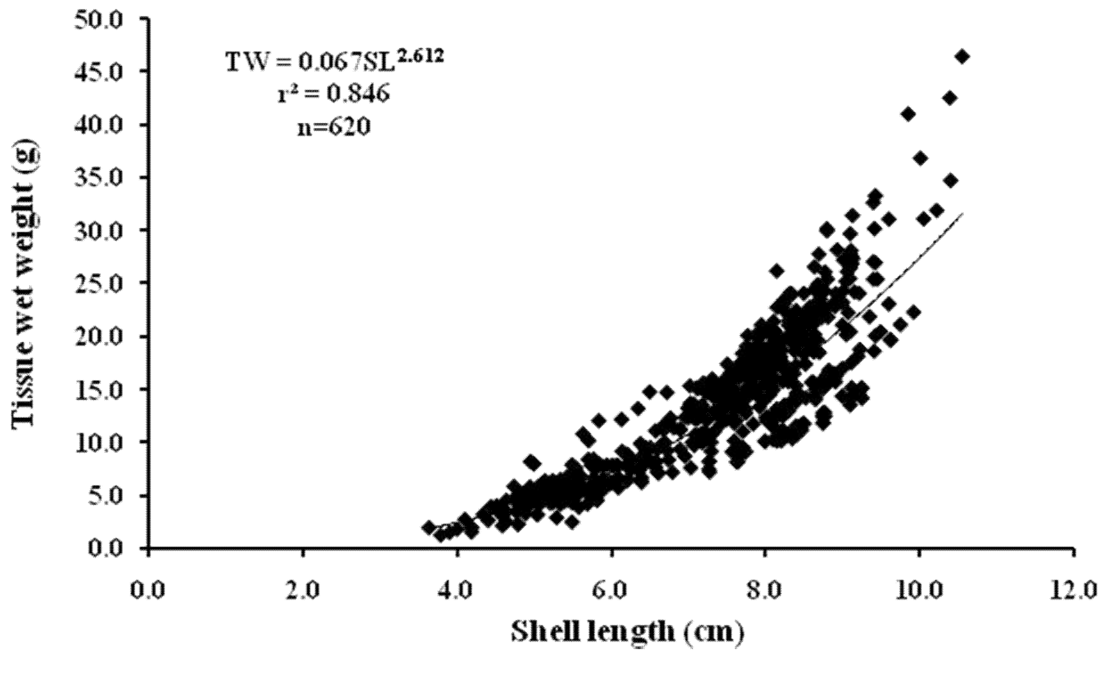

Fig. 26. Bivariate scatter diagram of length-tissue wet weight relationships of *L. marginalis* during the study period.

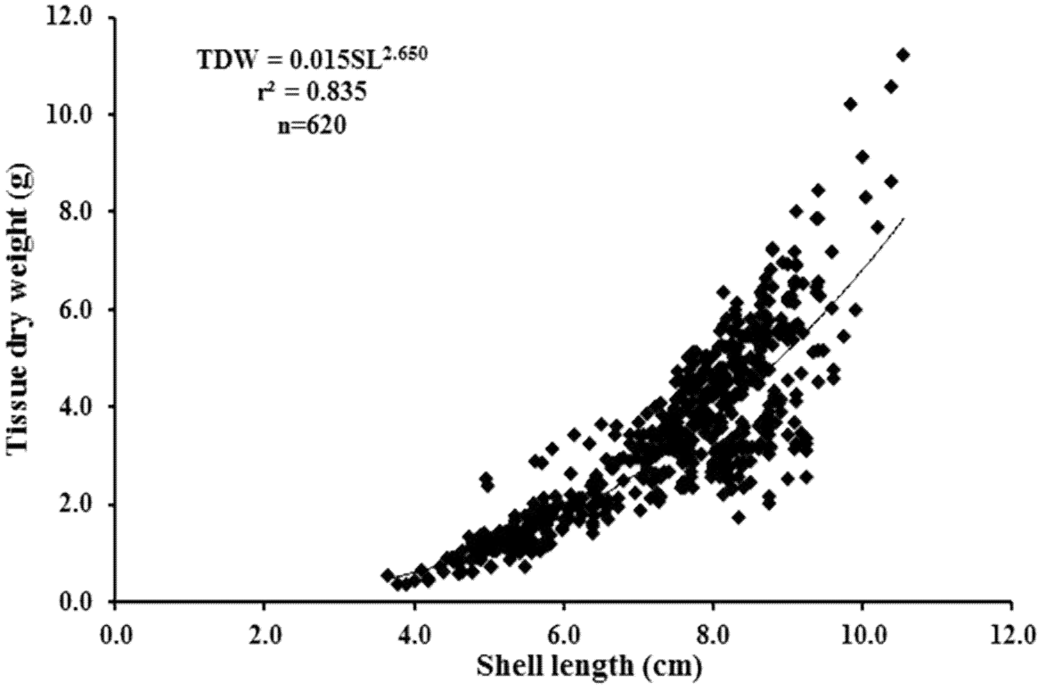

Fig. 27. Bivariate scatter diagram of length- tissue dry weight relationships of *L. marginalis* during the study period.

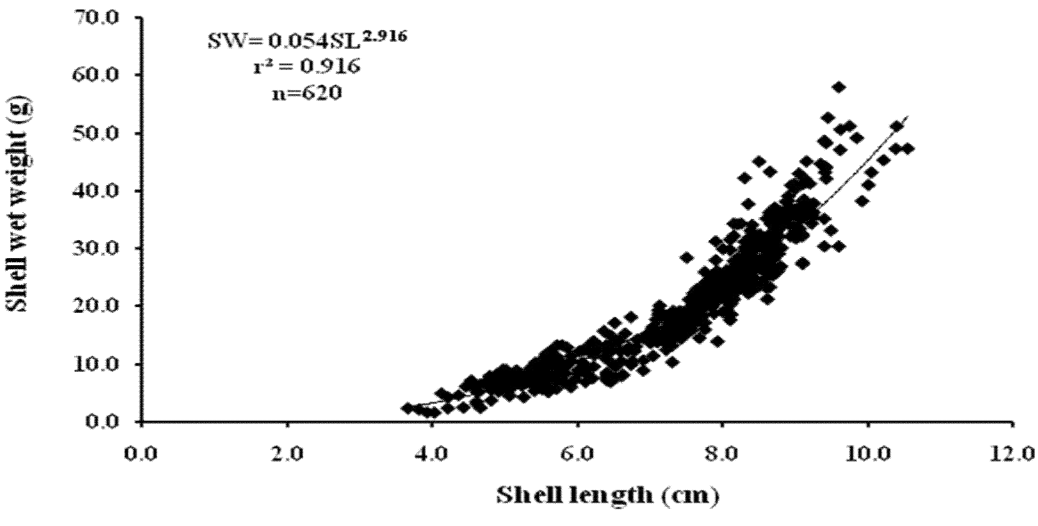

Fig. 28. Bivariate scatter diagram of length-shell wet weight relationships of *L. marginalis* during the study period.

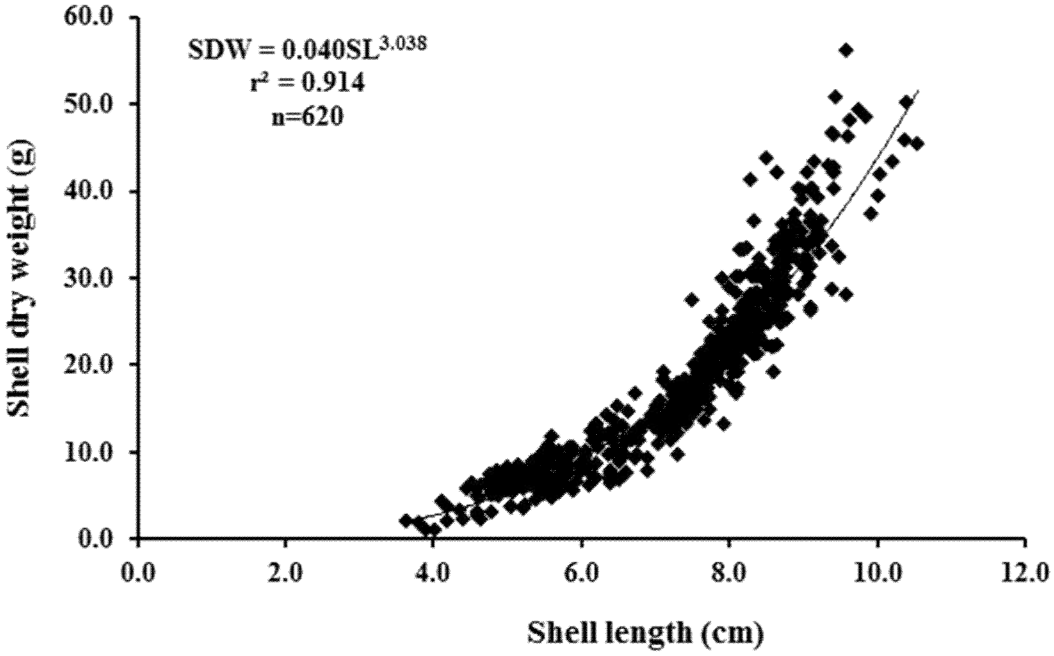

Fig. 29. Bivariate scatter diagram of length-shell dry weight relationships of *L. marginalis* during the study period.

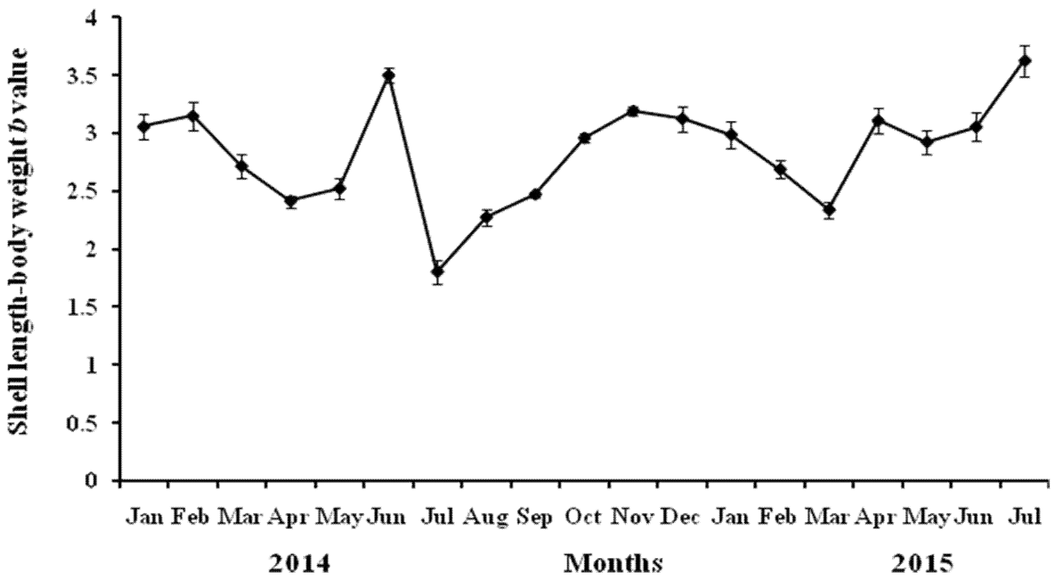

Fig. 30. Monthly variability in the *b* values of length-body weight relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

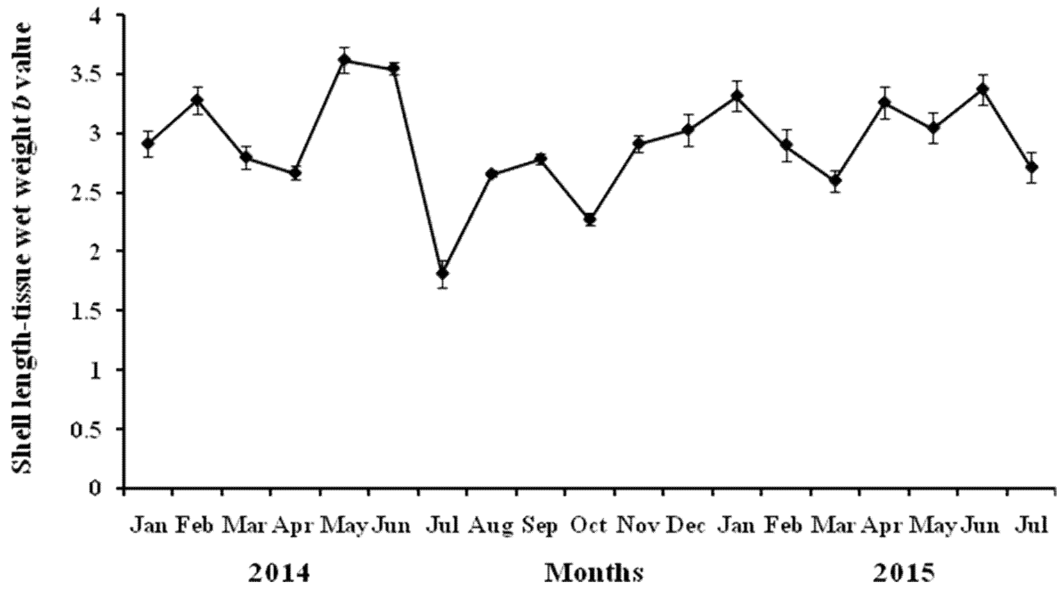

Fig. 31. Monthly variability in the *b* values of length-tissue wet weight relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

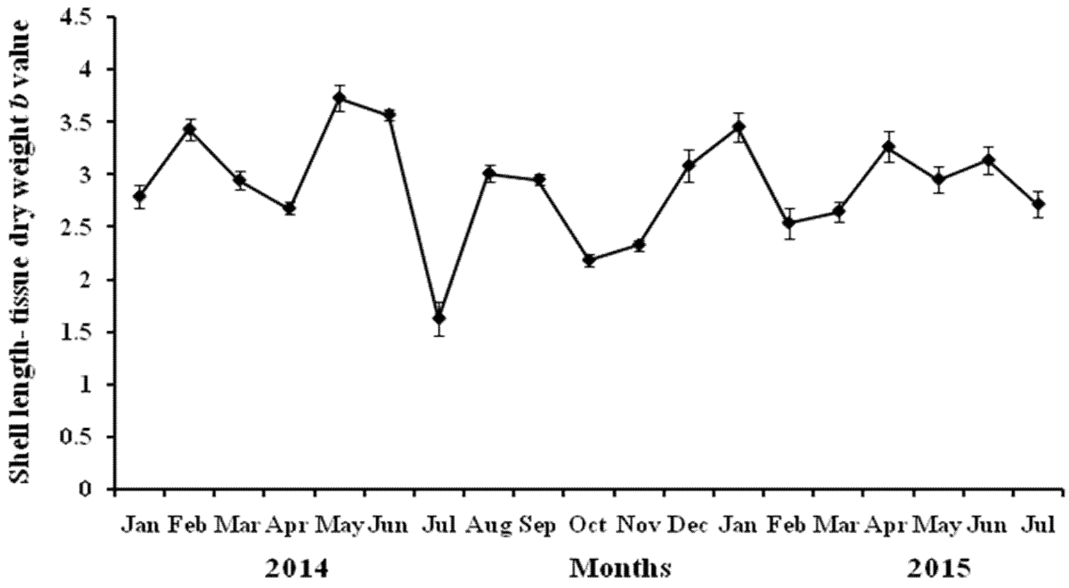

Fig. 32. Monthly variability in the *b* values of length-tissue dry weight relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

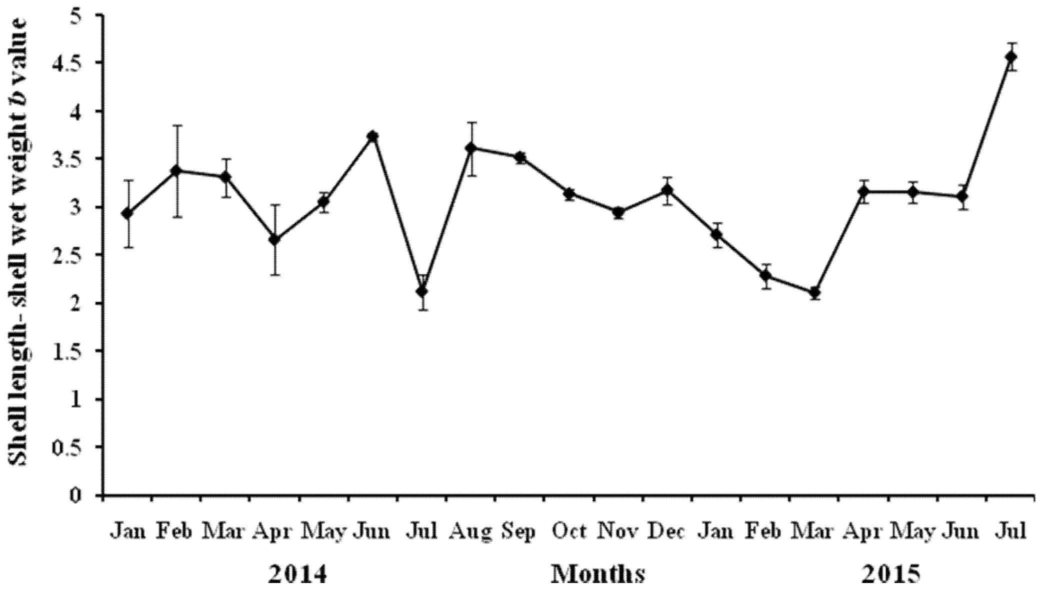

Fig. 33. Monthly variability in the *b* values of length- shell wet weight relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

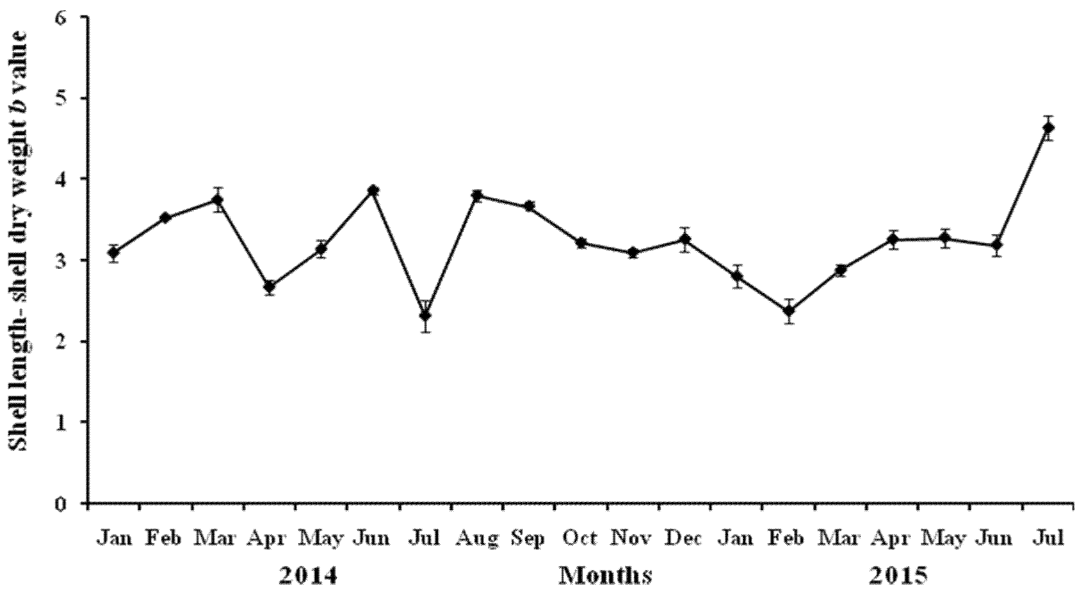

Fig. 34. Monthly variability in the *b* values of length-shell dry weight relationships of *L. marginalis* during the study period. Vertical bars represent standard error.

4. Discussion

4.1 Morphometric relationships

The calculated values of length-breadth and length-width relationships for the entire study period were B = 0.179 + 0.475L and W =0.196+ 0.3163L respectively. The monthly *b* values of length-breadth relationship varied from 0.3466 (October 2014) to 0.8132 (July 214) whereas for length-width relationship, the values ranged from 0.2495 (February 2014) to 0.3982 (August 2014). The linear relationships of length-breadth and length-width of *L. marginalis* during the study period showed that the short individuals were of less height/thickness and inversely long individuals were wide/high. The estimation of rates of aquatic secondary production in a given ecosystem is possible with the knowledge of size and shape, differential body growth and population ecology of organisms (Wilber and Owen, 1964). The data on morphometric relationship in the present study revealed that the length, breadth and width were influenced by variation in size. However, some individuals of same length showed different breadth and width, and these differences constituted shape variation as reported in other organisms (Jolicoeur and Worimann, 1960; Ramesha and Thippeswamy, 2009).

A variety of environmental factors are known to influence shell form in bivalves. The size of shell is more affected than their shape by fluctuation of ambient environmental parameters (Wilbur and Owen, 1964; Seed, 1968). Thus shape, rather than size generally provides more accurate knowledge on the dimensional relationships. During the present study period, the length of the mussels ranged between 3.65 cm and 10.55 cm. The average length was 7.33 cm, while the breadth and width of the mussels varied from 1.38 to 5.98 cm and 1.21 to 3.64 cm, respectively. The average breadth was 3.64 cm whereas the average width was 2.51 cm. The monthly *b* values of length-breadth and length-width showed fluctuation, which may be due to the fact that, the shape is controlled by genetics of the organism and size by ambient environment as stated by Seed (1968). The *b* values of morphometric relationships are usually compared between dimensional growth of related species or same species in various geographical regions. In the present study, the *b* values of length-breadth and length-width relationships were 0.482 and 0.3203, respectively. The *b* values of length-breadth and length-width relationships of the freshwater mussel *P. corrugata* from river Kempuhole, India were 0.585 and 0.333 respectively (Ramesha and Thippeswamy, 2009). The *b* values of length-breadth relationship of *Amblema plicata* at different sampling sites varied between 0.696 and 0.758 in the river Mississippi, USA (Hart, 1999). The morphometric relationships of *L. marginalis* were in good agreement with the above mentioned studies.

4.2 Length-weight relationships

During this study all the length-weight relationships represented a nonlinear pattern. The nonlinear relationships of length-body weight, length-wet tissue weight, length-dry tissue weight, length-shell weight, length-dry shell weight were $W = 0.1241L^{2.8282}$, $W = 0.0524L^{2.5905}$, $W = 0.0116L^{2.6296}$, $W = 0.0533L^{2.8769}$ and $W = 0.0391L^{2.9979}$ respectively for the entire study period. All the length-weight measurements showed a significant nonlinear relationships ($P<0.05$). The monthly equilibrium constant values of length-weight relationships were 1.8037 (July, 2014) and 3.5011 (June, 2014) for length-body weight, between 1.8185 (July, 2014) and 3.6224 (May, 2014) for length- tissue wet weight, between 1.6247 (May 2014) and 3.7295 (July 2014) for length-dry tissue weight, between 2.1091(March, 2015) and 3.7399 (June, 2014) for length-shell wet weight and between 2.2617 (April, 2015) and 3.8662 (June, 2014) for length-shell dry weight. The *b* values of length- body weight relationships showed some monthly fluctuations. During January and February, 2014 b value was slightly higher than 3, decreased from February to April and then rapidly increased at reaching the highest value 3.5011 in the month of June. From June to July a sharp decline of b value was observed and then the b value again increased from August to onward at reaching the value more than 3 during the winter in 2015. The b value of length-tissue weights and length- shell weights relationships represented almost same pattern during this study.

The length-weight relationship provides a mathematical expression to the relationship between two variables, length and weight, and also provides a measure of deviation between two variables indicating the state or general wellbeing of an organism. Length and weight of organisms have been highly correlated with life-history measures in cross taxonomic comparisons (Peter, 1983). The bivalve mollusks are known to show variations in quality of meat depending on their environmental and physiological conditions as well as seasons. The skewed relationship in the present study indicated that short individuals were light and long individuals were heavy. Such observation in other organisms has been reported by Ramesha and Thippeswamy (2009) and Bhattacharya and Banik (2012) Normally, as the age increases, the weight also increase; however, some individuals of the same age showed different weight and these differences could be due to reproductive strategies and environmental variables (Haag and Station, 2003; Ravera et al., 2007).

The length-weight relationship is an important measure for identifying the condition of bivalves in all stages of its life cycle. It helps to compare the condition of the bivalves in the context of pollution within the species or between the species. In fisheries research, length–weight relationships are important for the estimation of weight, where only length data are available and as an index of the condition of the animal (King, 1995). In the present study the *b* value of length-body weight, length-tissue wet weight, length- tissue dry weight, length-shell wet weight, length-dry shell weight relationship were 2.8550, 2.6120, 2.6500, 2.9160 and 3.038 respectively. Malathi and Thippeswamy (2011) studied the Allometry and condition index in the freshwater mussel *Parreysia corrugata* (Müller, 1774) from river Malthi in the Western Ghats, India from March 2007 to June 2008 and reported the b value for length- total weight, length-wet weight, length-shell weight and length-dry weight relationships were 2.6660, 2.6684, 2.7124 and 2.9370 respectively, which are similar with the findings of the present study. Ramesha and Sophia (2015) studied the allometry and condition index of the freshwater mussel *Parreysia favidens* (Benson, 1862) during April 2005 and May 2006 inhabiting the west flowing river Seeta at Seetanadi in the Western Ghats, India. They reported the b value for length-total weight, length-wet weight, length-shell weight and length-dry weight relationships were 3.066, 3.054, 3.139 and 3.110 respectively and are higher than the respective b values of the present study. Suryawanshi and Kulkarani (2014a) studied the comparative length - weight relationship of fresh water mussels, *Parreysia corrugata* and *Lamellidens marginalis* from Nanded region, Maharashtra, India. They reported the b value 3.2631 for *Parreysia corrugata* which is higher than present studyand 2.6084 for *Lamellidens marginalis* which is similar to the present study. Lowe-McConnell (1987) stated that many aspects could be accountable for the changes in fish growth such as variations in the habitat, fish activities, food availability and seasonal growth rates. The bivalves displayed marked seasonal variations in weight and biochemical content of the soft tissue (Williams and McMahon, 1989). In bivalves, the gonadal growth and maturation proceeds with bulkiness of soft body and consequent high body weights. Such sudden shift in the *b* values indicates the onset of maturation and gonadal growth in bivalves.

CHAPTER V

Seasonal changes in condition index and meat yield of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) from the northwest Bangladesh

Abstract

Condition index is an important parameter, shows the physiological status of bivalves and its seasonal variation linked with the reproductive activity as well as the seasonal energy storage of bivalves. Variations in condition indices of *Lamellidens marginalis* inhabiting in the freshwater ecosystem were determined for a period of 19 months (January 2014 to July 2015). Four different methods were used to determine condition index of the mussel. All the methods showed significant monthly variation of the condition index ($P<0.05$). In case of condition index-1 (CI-1) the highest value was recorded in June and lowest in November. A rapid increase of condition index was observed during April and June and then a sharply declined during June and August, this seasonal change of condition index could be linked with gonadal ripeness and spawning of the mussel. Meat yield index also showed significant monthly fluctuation ($P<0.05$). The higher condition index and meat yield index of the mussel during winter and summer indicated a better period for commercial exploitation of the mussel from the natural ground.

Key words: *Lamellidens marginalis,* condition index, meat yield index, seasonality.

1. Introduction

Relatively large variations in meat content occur in bivalve mollusks depending upon their physiological condition and variations in environmental parameters. In most bivalves, gonad development and growth prior to onset of spawning result in fattening and increased meat yield. Since such variations in meat yield are of importance in timing of harvesting and utilization of commercial species of bivalve, a study on the meat content and its temporal variations forms an important aspect of bivalve biology. Seasonal cycles in condition have been studied for different bivalve from Indian subcontinent as species of *Crassostrea* (Joseph and Madhyastha, 1986), species of *P. viridis* (L.) (Hemchandra and Thippeswamy, 2008), species of *Meretrix* (Durve and George, 1973), species of *Donax (*Ansell et al.,1973; Nagbhushanam and Talikhedkar, 1977a, 1977b and Balasubramanian et al., 1979), *Donax incarnatus* (Thippeswamy and Joseph, 1988) and freshwater mussel *Parreysia corrugata* (Müller, 1774) (Malathi and Thippeswamy, 2011). Due to lack of detail information about condition index of the mussels from Bangladesh, the present study was designed to estimate the condition indices and meat yield of *L. marginalis* inhabiting in the freshwater ecosystem in the northwest Bangladesh.

2. Material and methods

2.1 Study area and duration

The study was conducted in the freshwater lake situated in Rajshahi, the northern Bangladesh. The geographical location of the study site is 24º 22´ 21.56´´ N and 88º 38´ 11.16´´E (Fig. 3). The study was conducted during January, 2014 to July, 2015.

2.2 Sampling and biometric measurements

Monthly 30 individuals of *L. marginalis* were collected during the study period and subjected to morphometric measurements. Shell length of individual organisms was measured accurately to 0.01 mm using Vernier calipers. Total weight of individual mussels was taken using electric balance. The mussels were dissected to remove the meat which was then blotted and weighed individually. The individual weight of shell was also determined. Both the meat and shell were dried at constant temperature of 60°C for 2 days and weighed accurately to 0.01 g using an electronic balance. Condition indices (CIs) and meat yield index (MYI) were measured by following methods.

a. CI-1=Dry tissue weight (g)/ Dry shell weight (g) x 100 (Lucas and Beninger, 1985);

b. CI-2= Dry tissue weight (g) x 100/ Total weight (dry shell+ dry tissue) g (Barkati and Ahmed, 1994);

c. CI-3 = Dry meat weight (g) x 100/ (shell weight + tissue weight) g (Crosbay and Gale, 1990);

d. CI-4= Dry meat weight x 100 /(Body weight-Shell weight) g (Crosbay and Gale, 1990);

e. MYI= Weight tissue weight (g) x 100/ Body weight (g)

2.3 Statistical analysis

To know the temporal variation of the condition indices and meat yield index was tested using one way ANOVA with DUNCAN POSTHOC ranking. All the statistical analysis were performed on SPSS version 20 software at $P<0.05$.

3. Results

Monthly mean values of condition index (CI-1) varied from 10.44 (November) to 23.35 (June) (Fig. 35). At the start of this study in the month of January the CI was 21.48. CI gradually decreased from January to April (2014) and then sharply increased at reaching the highest annual peak 23.35 in the month of June. CI rapidly declined from June to onward at reaching the annual lowest value (10.44) in the month of November. CI again rapidly increased during December, 2014 and January, 2015. Monthly fluctuation of CI during January to May 2015 was similar as January to May 2014. During this study two peaks were distinct in January and June. Peaks were followed by periods of sudden decline which might be related with the spawning of the mussel. All of the CIs showed similar monthly fluctuation during this study (Fig. 36-38). During this study meat yield index was ranged from 18.0 to 36.23 (Fig. 39). Meat yield index also showed similar monthly fluctuation as condition indices. MYI was 34.99 at the start of the study, slowly decreased from January-April, 2014 and then slowly increased up to June. MYI sharply declined during July to onwards at reaching the lowest value 10.44 in November. Buildup of MYI took during December, 2014 and January, 2015. During this study two peak of MYI during winter and summer, indicates the better period for harvesting of the mussel from the natural ground.

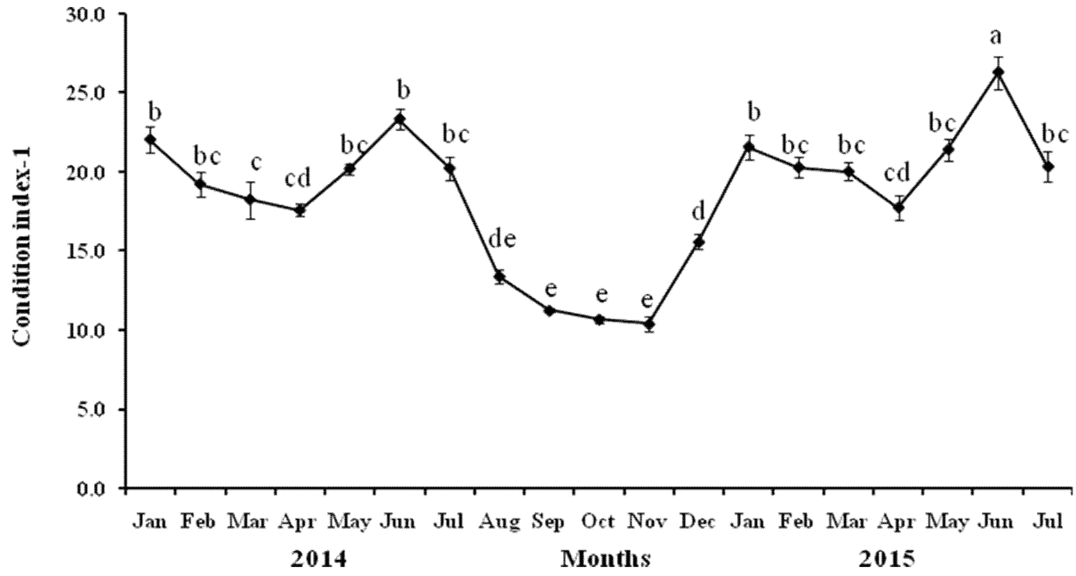

Fig. 35. Monthly variation of condition index (CI-1) (mean±SE) of *L. marginalis* (Lamarck, 1819) during the study from the northwest Bangladesh. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

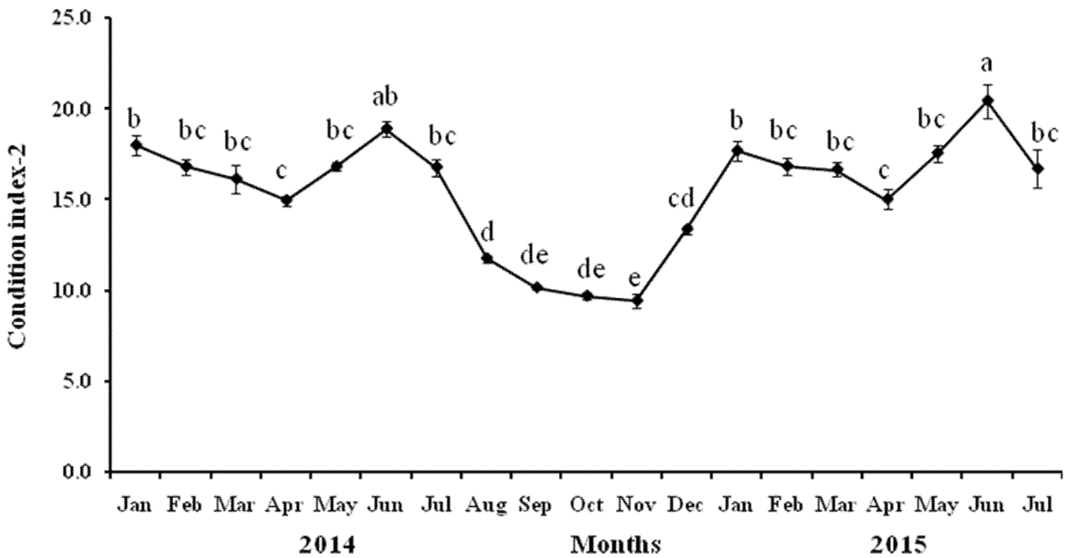

Fig. 36. Monthly variation of condition index (CI-2) (mean±SE) of *L. marginalis* (Lamarck, 1819) during the study from the northwest Bangladesh. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

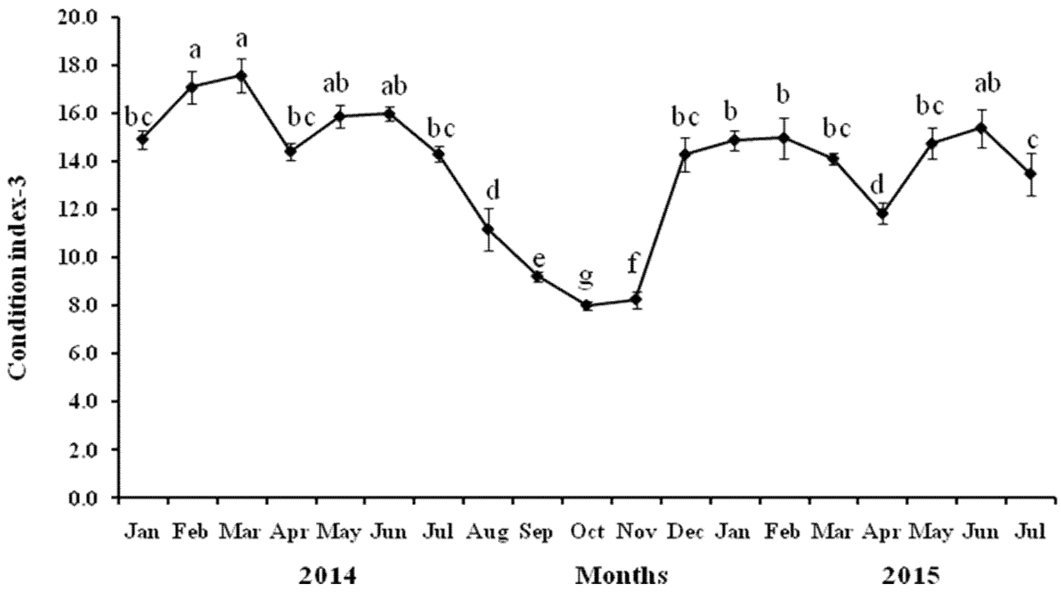

Fig. 37. Monthly variation of condition index (CI-3) (mean±SE) of *L. marginalis* (Lamarck, 1819) during the study from the northwest Bangladesh. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

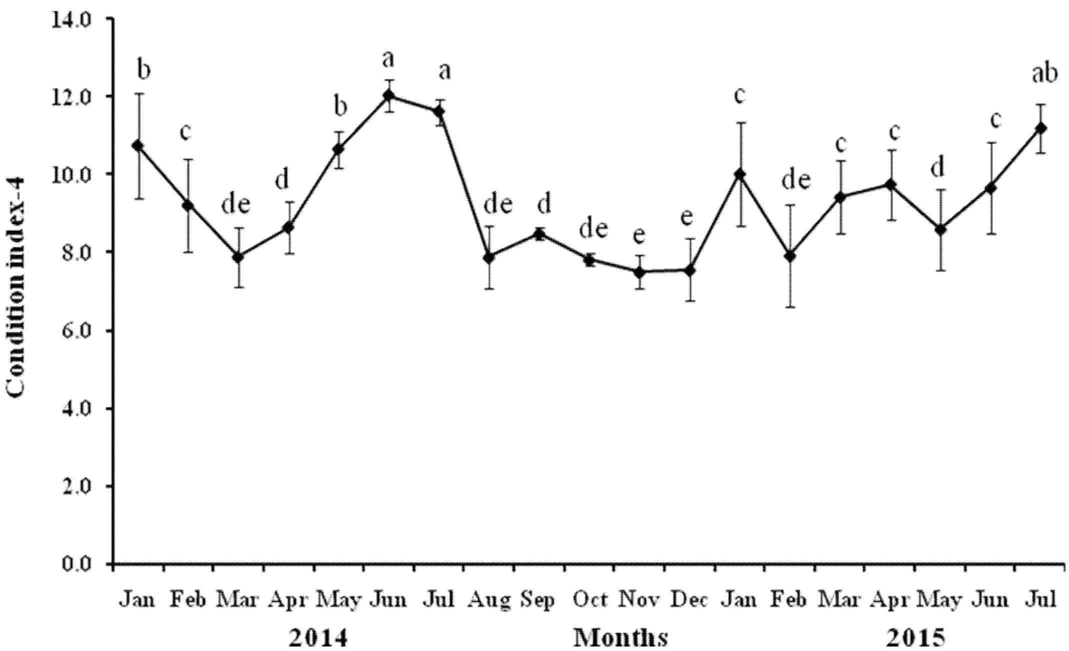

Fig. 38. Monthly variation of condition index (CI-4) (mean±SE) of *L. marginalis* (Lamarck, 1819) during the study from the northwest Bangladesh. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

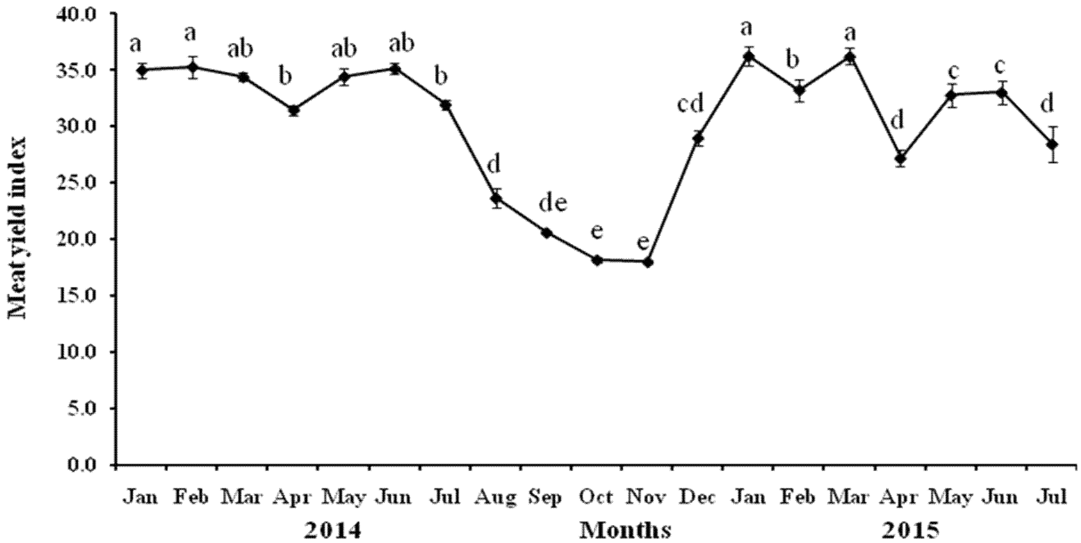

Fig. 39. Monthly variation of meat yield index (MYI) (mean±SE) of *L. marginalis* (Lamarck, 1819) during the study from the northwest Bangladesh. Vertical bars represent standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

4. Discussion

The data on monthly mean values of condition index of *L. marginalis* revealed primary peak during January 2014 and secondary peak during June 2014 (Fig. 35). After the primary peak, the condition index slowly decreased during January to April, 2014 and then rapidly increased during May and June. CI sharply declined from June to onward at reaching the lowest value in November, 2014. Build up of CI occurred during December and January. An examination of the temporal variation in the condition indices revealed obvious annual cycle of *L. marginalis.* The differences in the condition index observed in different months may be due to factors, such as environmental condition, food availability and the gonadal maturity (Jhingran, 1952; Bashirullah, 1975). Accumulation of gametes in follicles and resultant bulkiness of the gonad result in increased condition while release of gametes from the follicles and corresponding shrinking of gonadal mass result in lowering condition. Seasonal variations in the tissue mass or condition index of many bivalves follow a closely related pattern. Such pattern have been found useful for describing the period when harvesting could give maximum meat yield (Nair and Nair, 1987), for ecological purposes in elucidating the spawning period (Etim, 1990), and for bio-monitoring of environmental stress (Marcus et al., 1989). The ideal period for commercial exploitation of *L. marginalis* in the freshwater ecosystem of northwest Bangladesh could be during winter and summer months when the mussels are in good condition with more meat yield.

CHAPTER VI

Seasonal variation in reproductive pattern of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in the northwest Bangladesh

Abstract

The present study investigated seasonal variation in reproductive pattern of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in the northwest Bangladesh using gonadal index and histology. During the study sex of the mussel was differentiable in all the months. Out of 620 specimens collected 45.97% were female and 54.03% male. The overall male to female sex ratio was 1: 0.82, which was statistically different from the expected sex ratio of 1:1 (X^2 = 5.90, P < 0.05). Histological observation revealed five gonad development stages as early developing, late developing, ripe, spawning, spent both for male and female mussels. During this study sexually undifferentiated and hermaphroditic individual was not observed. Patterns of gonadal development, maturation and spawning were more or less similar for males and females. The mussel *L. marginalis* spawned throughout the year with a spawning peak during July and August when most of the mussels (up to 70%) participated in spawning. In the present study the 6-7 cm size group mussels were more potential group in respect to reproductive performance. Seasonal changes of gonadal index also clearly related with maturity stages: GI increased progressively with the development of maturity and reach a maximal value (9.41± 1.23) in June indicates the maximum ripeness of the gonad of the mussel. The GI value sharply declined from July to afterwards at reaching a minimal value of 4.83±0.96 in the month of September, indicates the spawning of the mussel population during this time.

Key words: *Lamellidens marginalis,* sexuality, spawning season, GI, GSI

1. Introduction

The sustainability of aquatic animals facing challenges due climate change. In natural ecosystem, mollusk comprises second largest group of invertebrate and regarded as most suitable phyla, to tolerate such a great threat of freshwater instability. Freshwater mollusk is economically important group of animal and widely used as food because of their high proteinacious and less amount of fat content. Although, mollusk having such high commercial importance, they receive very little attention. Among freshwater mollusk, bivalves are well classified. The developmental strategies of freshwater Uninoids showed variation from marine bivalves.

Reproductive activity of bivalves includes the initiation of gametogenesis, gonad maturation, spawning and egg fertilization and successive larval development. Freshwater mussels have a unique life cycle, a modified female reproductive system for brooding larvae which are adapted to parasitic development on fish (Dillon, 2004). The male releases spermatozoa into the water that are captured by the female using the incurrent siphon. Internal fertilization takes place in marsupia (modified suprabranchial chambers) where the embryos develop into glochidia (mature larvae). Glochidia discharged from the suprabranchial canal through the excurrent siphon and attach to a suitable host fish for the development of larvae to juvenile. The juvenile mussels ultimately drop from the host fish to the bottom of the waterbody and they become adult in the sediment (Dillon, 2004).

The reproductive processes of bivalves are internally influenced by genetic factors and externally by the environmental parameters, like water temperature, salinity and food availability (Gosling, 2003). The food availability is the main factor which determines the seasonal energy storage and gametogenic development of bivalves (Bayne, 1976; Arakawa, 1990; Kang et al., 2000). Numerous studies have been carried out in relation to the reproductive cycle and biochemical composition of bivalves and suggested that reproductive activity of bivalves are closely related to the seasonal cycle of biochemical composition (Whyte et al., 1990; Ruiz et al., 1992; Dridi et al., 2007). In general, prior to gametogenesis, reserves are accumulated in the form of glycogen, lipids, and protein, when food is available. The specific importance of these substrates and the timing of consumption in relation to gametogenesis vary between species as well as among populations of the same species (Bayne, 1976; Barber and Blake, 1981).

Worldwide, there are many research reports on reproductive cycles of marine bivalves (Kang et al., 2003; Park and Choi, 2004; Yang et al., 2011; Mondol et al. 2012; Mondol et al., 2015; Mondol et al., 2016a) but only a few studies have been conducted on reproductive cycles of freshwater bivalves (Lomte and Nagabhushanam, 1969; Nagabhushanam and Lohgankar, 1977a; Gaiawad and Kamble, 2013; Mondol et al., 2016b). Still to date there is no comprehensive research on reproductive biology of *Lamellidens marginalis* from Bangladesh. Therefore, the present study was undertaken in the freshwater habitat of northwest Bangladesh to investigate the annual gametogenic cycle of *L. marginalis* for better management and conservation of this species.

2. Materials and Methods

2.1 Sampling and biometry

Reproduction of freshwater mussel *Lamellidens marginalis* was studied in the freshwater lake situated in Rajshahi, the northern Bangladesh. The geographical location of the study site is 24º 22´ 21.56´´ N and 88º 38´ 11.16´´E (Fig. 3). Monthly collected specimens ranging from 4.0-10.0 cm in length were used for the reproductive assessment of the bivalve during January, 2014 and July, 2015. A systematic sampling scheme was conducted monthly and 30-50 individuals were randomly collected by hand picking. The collected specimens were taken in the laboratory of Fisheries Department, University of Rajshahi and analyzed. For all specimens shell length and body weight was taken prior to sexing, dissection, weighing and histology.. After collection the shell length and total weight of individual mussels were taken. The mussels were then dissected to remove the meat which was then blotted and weighed individually. Sex was then determined by rapid microscopic observation of fresh gonad smears. Sex ratio (expressed as the number of female per males, M: F) was determined. The gonad (gonad+digestive gland) was then separated from the soft tissue and weighted. Gonad, soft tissue and shell were then dried at constant temperature of 60°C for 2 days and weighed accurately to 0.01 g using an electronic balance.

2.2 Histology

To investigate the spawning season and gonadal maturity, histology was performed for a period of one year during January and December, 2014. Monthly 20 *L. marginalis* specimens were used for histology. After being opened the shell soft tissue was removed and a cross section (about 5 mm thickness) of the soft body of was cut through the middle of the body which contains the gonad, digestive gland, mantle and muscular foot tissue. The sectioned tissue was then fixed in Davidson's fixative for 24 hours. The samples were then dehydrated in graded alcohol series, cleared in xylene and embedded in para□n. The trimmed para□n blocks were sectioned at 6 µm thickness and stained with Harris' haematoxylin followed by eosin Y and mounted on a slide. The sex and gametogenic stage were determined from the histological preparations. The gonad developmental stages of *L. marginalis* were categorized into six stages using a modified version of the maturity scale accordingly by Juhel et al. (2003) as: (1) early developing; (2) late developing; (3) ripe; (4) partially spawning; (5) spent; and (6 undi□erentiated. The description and criteria of each gametogenic stage of males and females are summarized in Table 3 and Fig. 40-41.

Table 3. Description and criteria for gonad development stages in the *L. marginalis.*

Maturity stages	Gonadal development	
	Male	Female
Undifferentiated	Gonad predominantly composed of connective tissue; sex not distingushible.	Gonad predominantly composed of connective tissue; sex not distinguishable.
Early developing	Spermatogenesis occurs in the acini; testes lobes are lined by thick germinal epithelium; spermatocytes present; no spermatids or spermatozoa; the volume of testis is small compared to visceral mass.	Numerous oogonia and pre-vitellogenic oocytes lining the edges of the acini giving a granular appearance; no free oocytes present in the lumen.
Late developing	Spermatocytes develop into spermatids; spermatids move toward the centre of the lumen; small number of spermatozoa occupied the lumina in the acini and germinal epithelium at the periphery.	Free oocytes present in the lumen accounting for less than half of the oocytes present in the follicle while the remaining oocytes attached to the wall.
Ripe	Acini filled with mature spermatozoa with their tails pointing towards the centre of the lumen; spermatozoa bands close the follicle wall in very ripe specimens.	Ovary uniform in colour; acini filled with free oocytes in the lumen. Due to high abundance, oocytes acquire a polygonal configuration.

Spawning	Decrease in number of spermatozoa in the centre of acini with their flagella pointing towards the lumen. Zones evidencing ongoing gamete emptying are present.	Follicle walls breaking down; partially empty acini with variable amounts of post-vitellogenic oocytes; some follicles empty having released their gametes.
Spent	Small number of undischarged spermatozoa are degenerated; many haemocytes present in the tubules; connective tissue present.	Follicles appear broken, scattered and relatively empty; only residual oocytes found in follicle undergoing resorption;

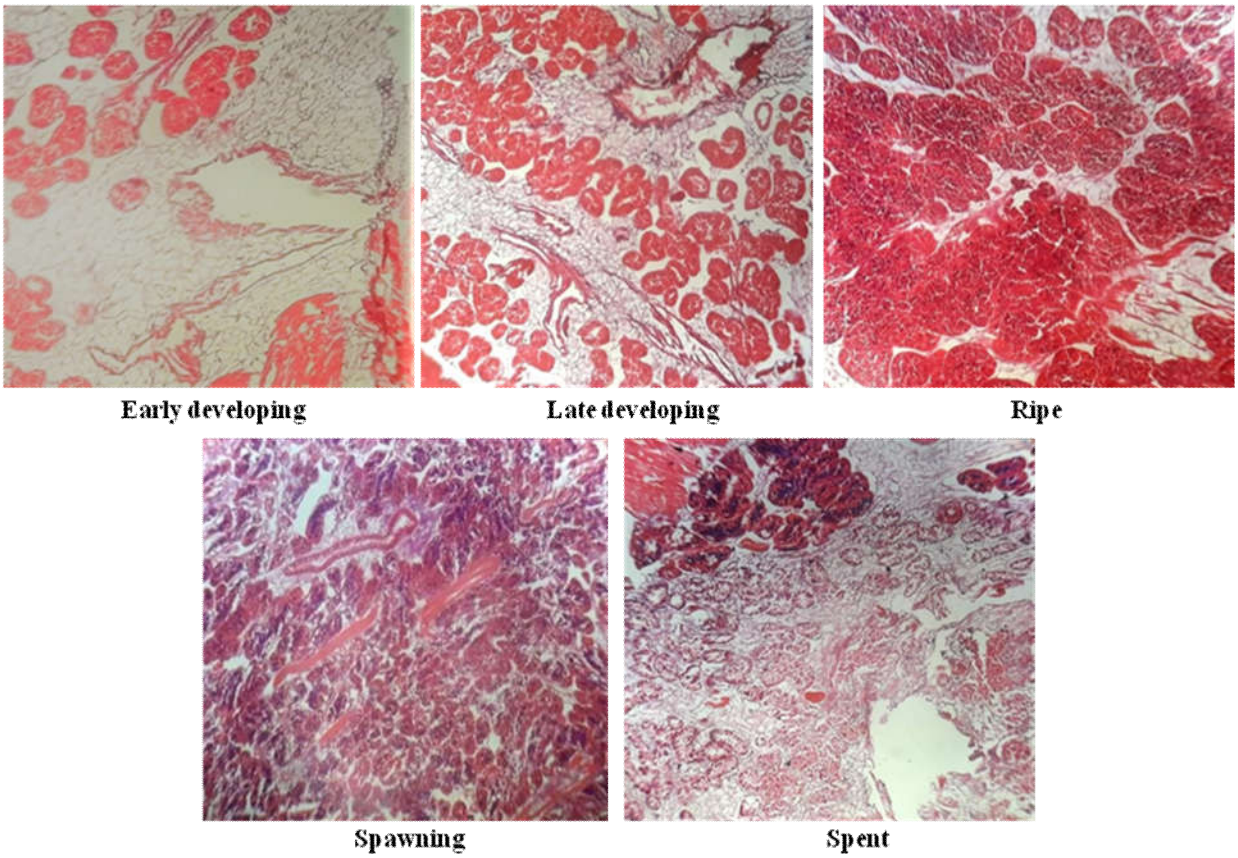

Fig. 40. Gonad development stages of male *L. marginalis* during the study period.

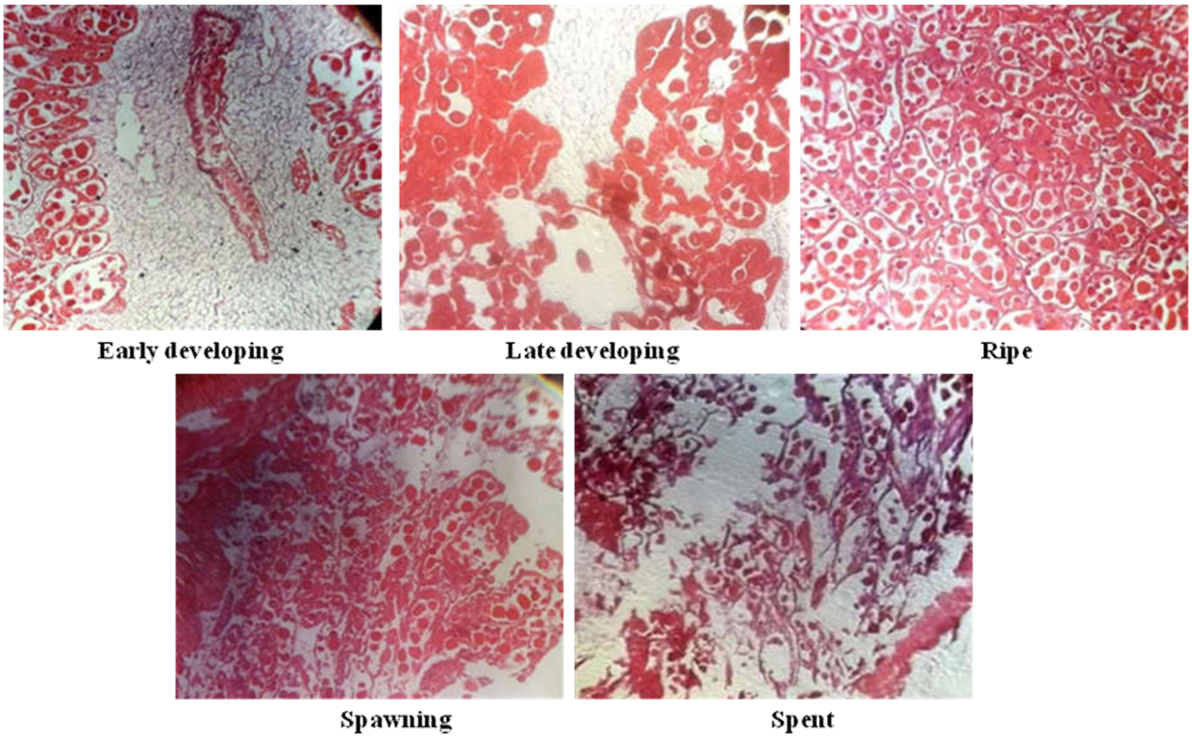

Fig. 41. Gonad development stages of female *L. marginalis* during the study period.

2.3 Reproductive effort

A simple and quantitative indicator of the reproductive state is the gonad index (GI) (Aideed et al. 2014). This index is useful to determine potential spawning period (minimal GI-values). In this study, GI was computed for each individual, by using the following equation:

GI = Dry weight (gonad +digestive gland) (g) / Dry weight of shell (g) x100

In some works, flesh weight is used in place of shell weight, but the calculation of the gonad index on this basis may create problems since flesh weight is known to vary periodically, with higher amplitude than shell weight. Furthermore, preliminary calculations showed that GI based on shell weight was the best discriminant index concerning reproductive activity. Gonad index variation was also useful to analyze synchronicity of gametogenesis.

To know the reproductive effort of the bivalve gonad somatic index was also calculated using the following equation-

GSI = Wet weight (gonad +digestive gland) (g) / Whole body weight (g) x100

2.4 Statistical analysis

All statistical analyses were performed using SPSS (version 20.0). Statistically significant deviations from the expected sex ratio of (M: F) 1:1 were assessed by Chi-square (χ^2) analysis with one degree of freedom. Monthly significant difference in gonad index (GI) and GSI of the mussel was tested one way analysis of variance (ANOVA) with DUNCAN *post hoc* ranking. Statistical significance was set at the alpha < 0.05 level.

3. Results

3.1 Sexuality and sex ratio

Microscopic observations of gonads revealed that sex was differentiable in all the months (Table 4 Fig. 42). During the study out of 620 mussels examined microscopically, of which 285 were female (45.97%) and 335 were males (54.03%). The overall male to female sex ratio was 1: 0.82, which was statistically different from the expected sex ratio of 1:1 indicating the prevalence of males in the study area during the study period (χ^2= 5.90, P < 0.05).

In the month of January and February the sex was male dominated. During March and April, 2014 the male and female individuals were equal in number after that a female dominance sex ratio was observed during May and June, 2014 (Table 4). During July and August again a 1:1 sex ratio was in evidenced. From September to December again a male dominance sex ratio was observed. In 2015, a similar trend in sex ratio was observed as 2014 (Table 4).

More precisely, sex-ratio showed a clear relationship with the size of individuals:

individuals < 7.0 cm majority were males. Above 7.0 cm the proportion of females increased progressively and reached approximately 50% in oldest individuals having the shell length >8.0 cm. In this respect, *L. marginalis* is a protandric successive species with a highly dominant male phase (Fig. 43).

Table 4. Monthly analyzed specimens and sex ratio of the *L. marginalis* during the study period.

Month	Male (A)	Female (B)	Total	Ratio	Ratio	*Chi-square* (χ^2) value	M: F Ratio	Significance
2014, Jan	17	13	30	1.31	0.76	0.53	1: 0.76	NS
Feb	16	14	30	1.14	0.88	0.13	1: 0.88	NS
Mar	15	15	30	1.00	1.00	0.00	1: 1	NS
Apr	20	20	40	1.00	1.00	0.00	1: 1	NS
May	16	24	40	0.67	1.50	1.60	1: 0.67	NS
Jun	11	19	30	0.58	1.73	2.13	1: 0.58	NS
Jul	15	15	30	1.00	1.00	0.00	1: 1	NS
Aug	15	15	30	1.00	1.00	0.00	1: 1	NS
Sep	19	11	30	1.73	0.58	2.13	1: 0.58	NS
Oct	20	10	30	2.00	0.50	3.33	1: 0.5	NS
Nov	21	9	30	2.33	0.43	4.80	1: 0.43	*
Dec	22	8	30	2.75	0.36	6.53	1: 0.38	**
2015, Jan	24	6	30	4.00	0.25	10.80	1: 0.25	**
Feb	17	13	30	1.31	0.76	0.53	1: 0.76	NS
Mar	16	24	40	0.67	1.50	1.60	1: 1.14	NS
Apr	25	25	50	1.00	1.00	0.00	1:1	NS
May	16	14	30	1.14	0.88	0.13	1: 0.88	NS
Jun	12	18	30	0.67	1.50	1.20	1: 1.5	NS
Jul	18	12	30	1.50	0.67	1.20	1: 0.67	NS
Total	335	285	620	1.18	0.85	4.03	1: 0.82	*

NS, not significant; * significant at 5% level ($X^2_{t\,1,0.05}$=3.84 and 1% level $\chi^2_{t\,1,0.01}$=6.63)

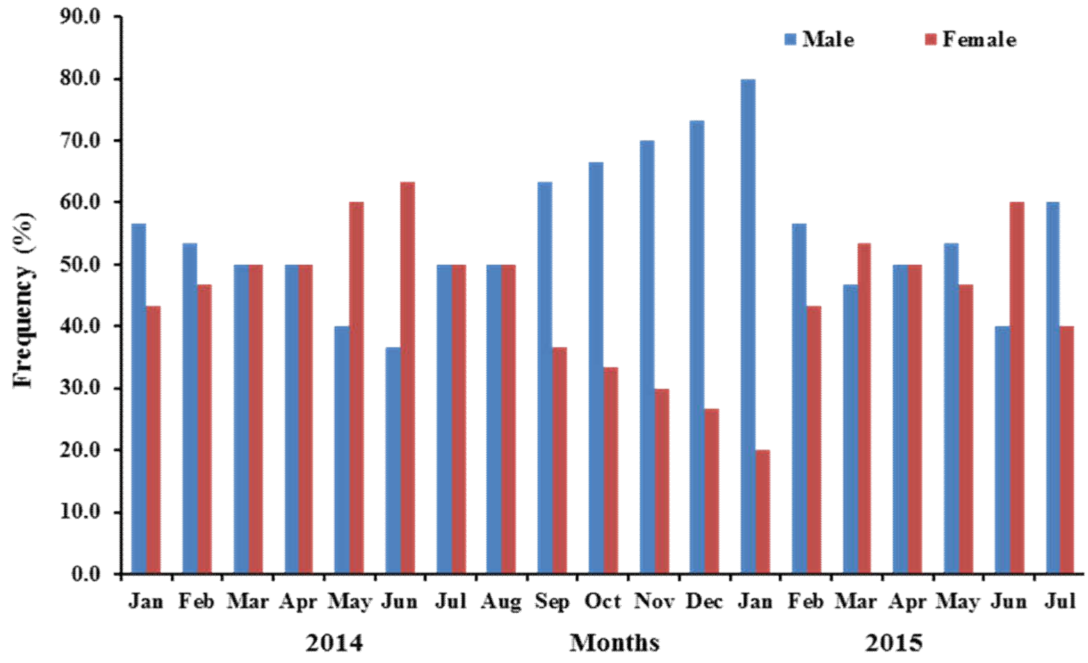

Fig. 42. Monthly frequency (%) distribution of male and female of *L. marginalis* during the study period.

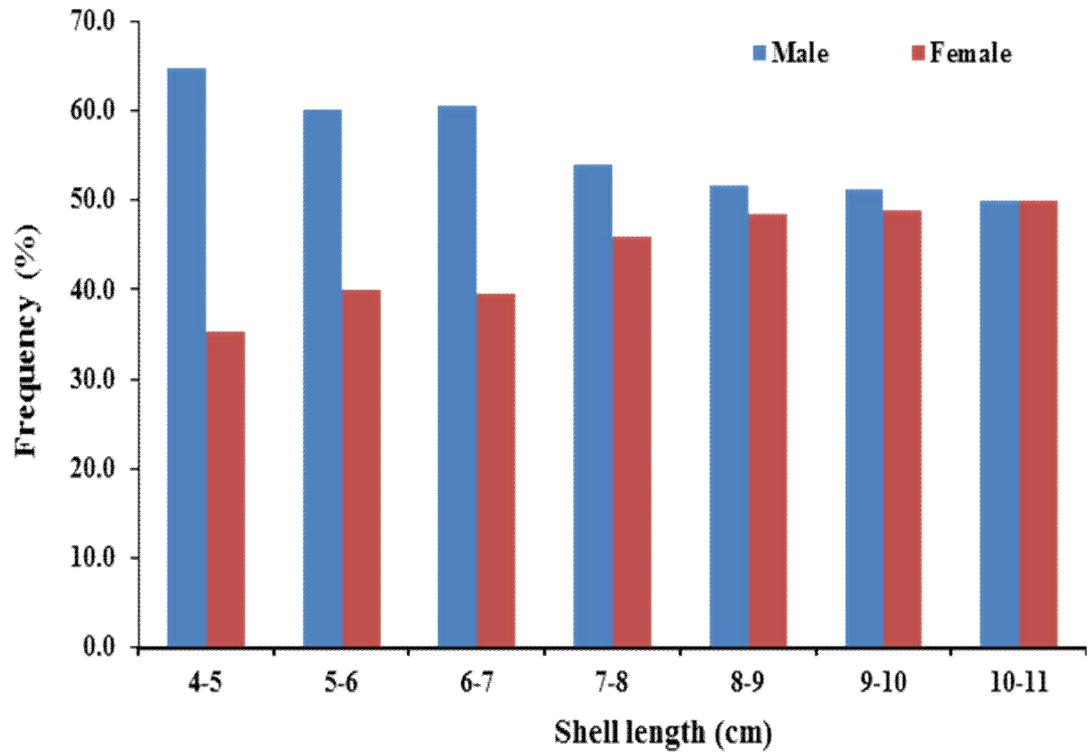

Fig. 43. Length basis frequency (%) distribution of male and female of the studied specimens during the study period.

3.2 Gametogenic cycle

From the histological preparation I observed 5 gonad development stages as early developing, late developing, ripe, spawning, spent both for male and female mussels (Fig. 44-45). During this study there was no sexually undifferentiated and hermaphroditic individual was observed. Patterns of gonadal development, maturation and spawning were more or less similar for males and females throughout the year (Fig. 44-45). Spermatogenesis was initiated during January, February, April, September, November and December when early developing males were observed in the histological preparations (Fig. 44). In the month of January 20%, in February 5%, in April 5%, in September 5 %, in November 20% and in December 30% male gonads were in early developing stage (Fig. 44). Similarly, oogenesis was also initiated during January, February, March, April, October, November and December (Fig. 45). In January 10 %, in February 15%, in March 10 %, in April 5 %, in October 5%, in November 15% and in December 25% females were in early developing stage (Fig. 45). Late developing males were observed during the whole of the year except in the month of July and September (Fig. 44). Similarly, late developing females were also observed whole of the year except in the months of July and September (Fig 45). Ripe male and female were observed throughout the sampling period. Both males and females exhibited similar spawning pattern. Peak spawning was observed during July (when 70% male and 65% female spawned) and August (when 45% male and 45% female spawned). Spent males occurred throughout the study period except January, February, May and June (Fig. 44), whereas spent females were not visible during February, May and December (Fig. 45).

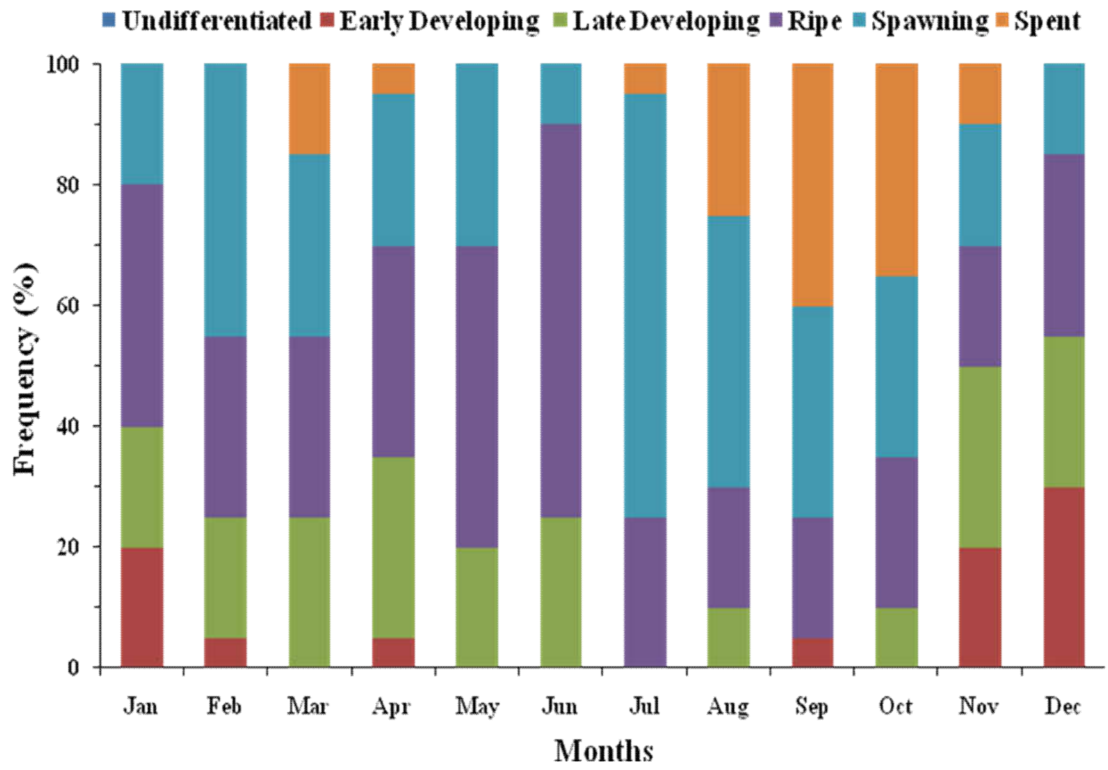

Fig. 44. Monthly percentage distribution of male *L. marginalis* at different gametogenic stages during January and December, 2014.

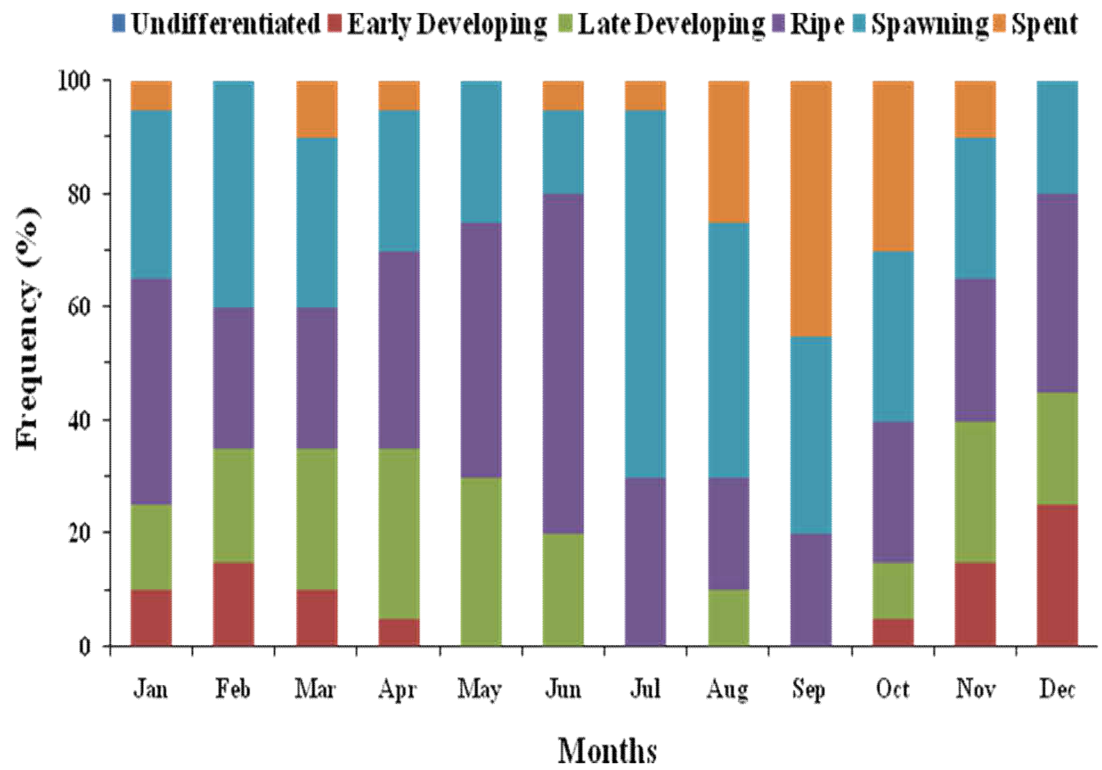

Fig. 45. Monthly percentage distribution of female *L. marginalis* at different gametogenic stages during January and December, 2014.

3.3 Reproductive effort

To quantify in a more precise way the reproductive activity, variations in dry weight of gonad, gonad index (GI) and gonad somatic index (GSI) were analyzed (Fig. 46-49). Since these variations were not significantly different between males and females, data were pooled. During the study seven significant decreases were observed in dry weight of gonad for of the mussel and represented 13-46% weight loss of the gonad, in February, 2014 (19.62%), in April 2014 (13.38%), in August 2014 (24.47%), in October 2014 (18.70%), in November 2014 (18.67%), in February 2015 (35.0%) and in May 2015 (46.0%) (Fig. 46). These variations were presumably due to gamete emissions or digestive gland resorption. To determine the origin of these losses, GI and GSI monthly variations were analyzed as GI clearly related with maturity stages: GI increased progressively with the development of maturity to reach a maximal value (9.41± 1.23) in June indicates the maximum ripeness of the gonad of the mussel (Fig. 47). The GI value sharply declined from July to afterwards at reaching a minimal value of 4.83±0.96 in the month of September, indicates the peak spawning of the mussel population during this time (Fig. 47). The data represented in the Fig. 48 reveals that the 6-7 cm size groups are more potential group in respect to reproductive performance, they represented 8.78% GI. Taking into account the relationship between GI and maturity stages, increases in GI could indicate developing gametogenesis, whereas obvious GI decreases should indicate principal spawning. GSI result also indicates same picture as GI (Fig. 49). In this respect, if some small irregularities in curves were not significant, and were presumably due to sampling variability and/or minor spawning or gamete resorption, the major and statistically significant variations, supporting those observed in gonad weight.

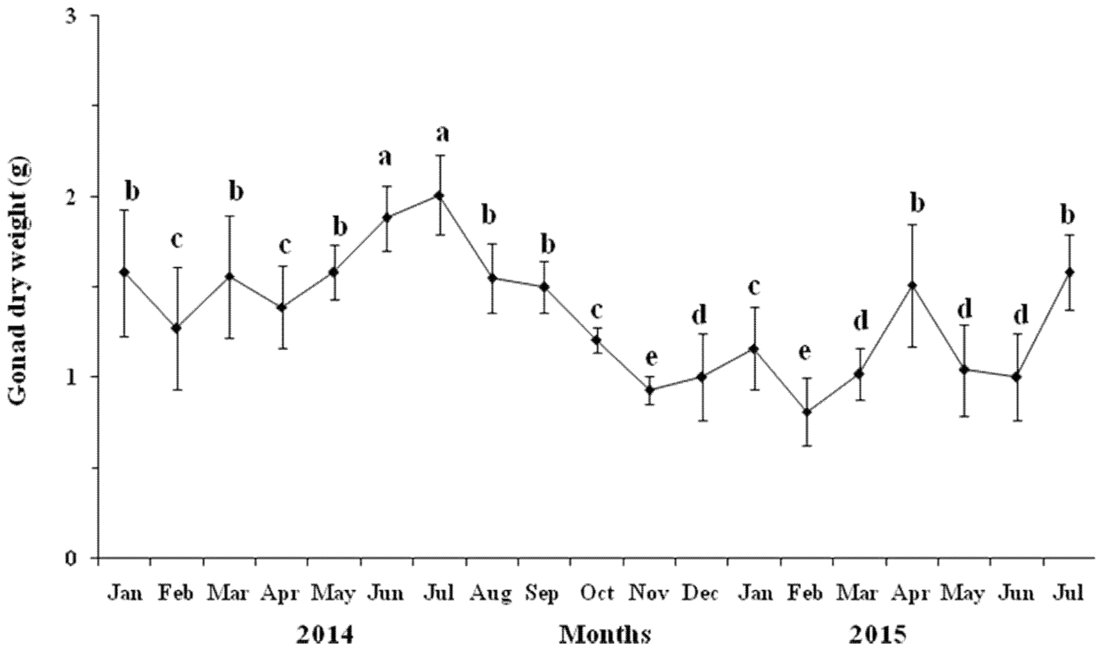

Fig. 46. Monthly changes in gonad dry weight (g) of the studied specimens during the study period. Vertical bar represents standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

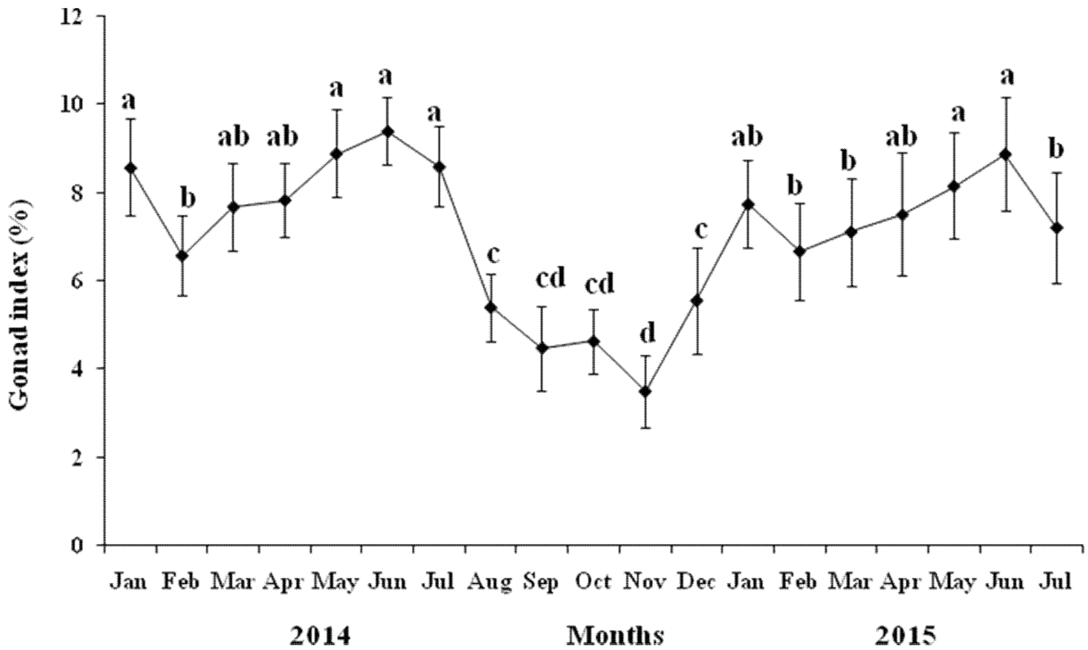

Fig. 47. Monthly changes in gonad index (GI %) of the studied specimens during the study period. Vertical bar represents standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

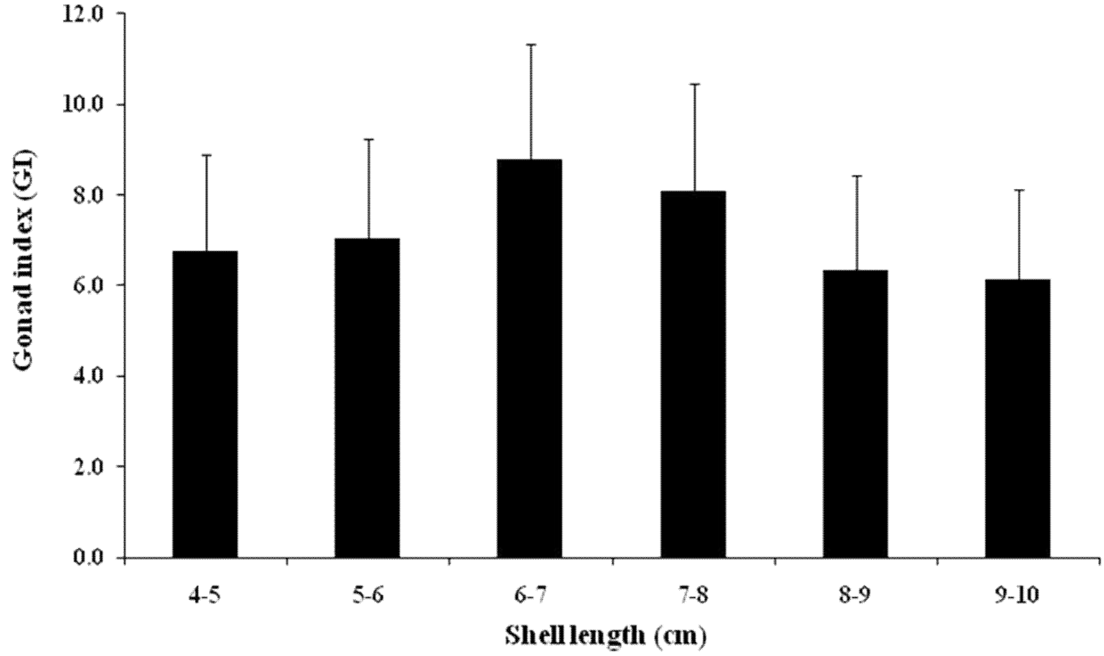

Fig. 48. Gonad index (GI) of the studied specimens in length basis during the study period. Vertical bar represents standard error.

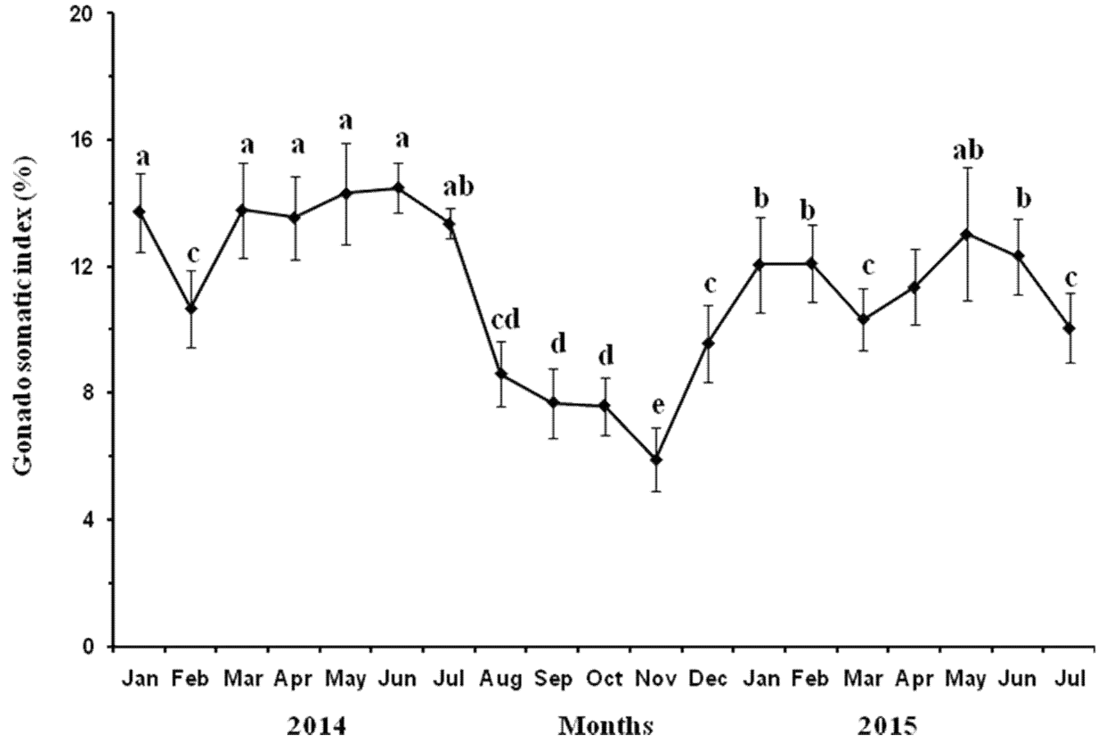

Fig. 49. Monthly changes in gonado somatic index, GSI (%) of the studied specimens during the study period. Vertical bar represents standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

4. Discussion

4.1 Sexuality and sex ratio

In this study, where mussels were sampled over a 19-month period, gonads and gamete development were observed in mussels in all months, and in all sizes of mussel. The sex ratio of male and female of the mussel fluctuated monthly and the overall sex ratio was 1: 0.82 (M: F) which was significantly different from the expected 1:1 (M: F). Similar to the present study Mondol et al. (2016b) studied length-weight relationship, condition index and sex ratio of freshwater mussel *Lamellidens corrianus* (Lea, 1834) from the freshwater ecosystem, northwest Bangladesh and reported the overall male to female sex ratio was 1: 0.92 and di d not differ significantly from the expected 1: 1 ratio. Gaikwad and Kamble (2013) analysed the annual reproductive cycle of *L. marginalis* and *Parreysia corrugata* from Panchaganga River Maharashatra, India during June, 2010 to May, 2011. They reported 1:1 sex ratio for both of the species of the bivalves. Niogee et al. (2019) investigated the ovarian cycle of freshwater pearl mussel, *Lamellidens marginalis* (Lamarck, 1819) that was collected monthly from a rearing pond in Bangladesh and reported the overall male-female sex ratio 1:1.08 did not deviate significantly from the 1:1 ratio as expected (χ^2-test). Cek et al. (2006) observed the seasonal reproductive cycle, gonad structure and sex ratio of the freshwater mussel *U. terminalis* from Gölbaşı Lake, Turkey and reported slightly female-biased sex ratios and were not significantly different from the expected 1:1 ratio (P>0.05).

The majority of young *L. marginalis* were males in the earlier age, as is generally the case in pearl oysters (Tranter, 1958). Afterwards, the sex ratio of *L. marginalis* tends increasingly towards femaleness as the population becomes older, and this phenomenon can only be explained satisfactorily on the basis of extensive protandric sex change. This finding supports those of Tranter (1958) who considered *P. margaritifera* to be a protandric consecutive hermaphrodite. Tranter (1958) also observed rare protogynic changes in this species and assumed that several sexual phases occur in the life of an individual. These sexual changes are known to be potentially related to food availability. As a general case, good conditions will favour femaleness, whereas bad conditions, or stress, retard it.

4.2. G***ametogenic cycle and reproductive effort***

Bivalves are known to undergo an annual reproductive cycle that involves gametogenesis followed by either a single or several spawning events, which are in turn followed by a period of gonad reconstitution. The number of spawning events and duration of the spawning period can vary greatly with respect to species, geographic area and environmental conditions (Gosling, 2003). From the data presented in this study it is noticeable that *L. marginalis* represented a continuous breeding pattern throughout the year, with no 'inactive' period. In that respect, *L. marginalis* can be classified into the 'continuous spawner' bivalve group. Gaikwad and Kamble (2013) studied the gametogenic phenology in freshwater Molluskan species: *L. marginalis* and *Parreysia corrugata* from Indian waters also reported a continuous breeding for both of the species. Continuous breeding is also reported in the genus *Pinctada* (Gervis and Sims, 1992). Concerning histological development, such continuous reproductive competence is explained by the fact that: 1) follicles in a same gonad are not generally at the same stage; 2) gametogenesis is rapid and always active (Joseph and Madhyastha, 1984; Lefort and Clavier, 1994); and 3) spawning is generally incomplete. In this study, peak spawning events occurred during the warm months (July to August) and this is in agreement with other studies that have investigated reproduction in tropical bivalves (Lefort and Clavier, 1994; Grant and

Creese, 1995). This reproductive strategy differs from those observed in temperate areas (Lubet and Mann, 1987), where most marine bivalves have an annual reproductive cycle with well defined periods of storage, gametogenesis, spawning and inactivity controlled by the variation of several environmental factors. In the present study sudden decrease in gonad dry weight, GI, GSI and condition index during July to August also confirm the peak spawning time of the *L. marginalis* in the freshwater ecosystem of northwest Bangladesh.

Surplus energy is allocated between somatic and reproductive compartments. The fraction of total growth allocated to reproduction is called the reproductive effort (Lucas, 1992; Navarro and Iglesias, 1995). In temperate species, surplus energy is first stored and is afterwards transferred to reproduction at the most favorable time for larval development and recruitment success. In tropical species, this allocation is presumably continuous based on the comments above. In the present study the GSI of the *L. marginalis* was ranged from 5.59 to 14.49%, indicates that the bivalve produced up to 14.49% of their body mass as eggs (Fig. 49).

4.3. *Gametogenesis and environmental consequences*

The gametogenesis in the bivalves varies with geographic locations (Ventilla, 1984), and is mainly influenced by both water temperature and food availability (Ruiz et al., 1992). In the present study, the bivalve *L. marginalis* showed gametogenic activity in whole of the year. In association with increasing water temperature the bivalve matured rapidly and became ripe during May and June. Peak spawning event occurred during July and August, when the water temperature was 32.3-34.5°C. Several studies also reported a similar seasonal cycle of gametogenesis of bivalves in different parts of the world. Gaikwad and Kamble (2013) studied the gametogenic phenology in freshwater molluskan species: *L. marginalis* and *Parreysia corrugata* from Indian waters and reported that average water temperature between 20-25°C was most suitable for spawning activity of the bivalves. They also reported the spawning during June to September when the dissolved oxygen level was suitable and carbon dioxide level was very less. Kang et al. (2003) investigated the annual reproductive cycle of adult oysters from Gosung Bay, off the southern coast of Korea. The adult oysters in Gosung Bay, initiated gametogenesis in March at 10-12°C and after rapid gonad maturation spawning occurred from May to October at 20-27°C. Most oysters became reproductively inactive in November when water temperature dropped below 17°C.

In conclusion, the present study reports the annual gametogenic cycle of freshwater mussels (*L. marginalis*) collected from a freshwater lake of northwest Bangladesh using histology and biometry. The mussel exhibited similar gametogenic patterns for both sexes with a continuous breeding throughout the year and a peak spawning pulse during July and August.

CHAPTER VII

Seasonal variation in biochemical composition of the freshwater mussel *Lamellidens marginalis* (Lamarck, 1819) in the northwest Bangladesh

Abstract

The aim of this study was to investigate the seasonal variation of water content, protein, carbohydrate, and lipid contents in freshwater bivalves *Lamellidens marginalis.* The bivalves were studied for a period of 19 months from January 2014 to July 2015 from the freshwater ecosystem of northern Bangladesh. During the study the water content ranged from 74.66 to 81.88%, carbohydrate content ranged from 19.83%-21.25%, protein content ranged from 14.04-38.88% and lipid content ranged from 5.3%- 12.06%. The carbohydrate and protein level showed clear seasonal fluctuation and inversely related to each other. The lipid level showed several monthly fluctuations. Higher carbohydrate observed during January and March; indicate higher energy accumulation of the mussel during this period. A higher protein level observed during June and July could be linked with the ripe gamete of the mussel and lower level of protein during August and October could be linked with the spawning release of gamete of the mussel

Keywords: *Lamellidens marginalis*, carbohydrate, protein, lipid, water content.

1. Introduction

The bivalves are ecologically important because of their widespread distribution and biological filtration activity (Lewandowski and Stanczykowska, 1975; Kasprzak, 1986) and also economically, used as food and in the production of freshwater pearls (Subba Rao and Dey, 1989). Bivalve mollusks are potential sources of valuable proteins, carbohydrates and minerals. The biochemical analysis is also known as percentage composition of some fundamental elements like water, protein, lipids, carbohydrate and minerals for human diet (Ramakrishnan and Venkat rao, 1995). The high protein content foods availability is the biggest problem in some developing countries. The knowledge about nutrition of edible living organisms is tremendously significant since the nutritive value is reflected in its biochemical analysis (Nagabhushanam and Mane, 1978). The edible species of bivalve mollusks are tasteful and it will get more importance next to fish and prawn.

The biochemical composition of mollusks is influenced by its size, growth and reproductive status. Bivalves play an important role in the ecosystem equilibrium and constitute an important economic end point. The bivalves have not been the subject of intense studies despite the presence of rich diversity of edible and commercial species in Bangladesh. Cyclical changes in biochemical composition of animal tissue are mainly studied to assess the nutritive status of an organism. This information may, however, be used in supplementing other studies like assessment of the course of the reproductive cycle. Marine bivalves indicated that seasonal cycle of energy storage and biochemical cycles are closely related to reproductive activity (Ruiz et al., 1992). According to Gabbott and Bayne (1973), seasonal metabolic activities in mollusks result from complex interactions among food availability, environmental conditions, growth and gametogenic cycle.

Of all the components, changes in carbohydrates play an important role in the seasonal variation of the chemical composition. In general, the water content of the tissue of bivalves usually gives an indication of the time of spawning. Variation in dry tissue weight of mollusks is always associated with biochemical components. Seasonal changes in the biochemical constituent are the characteristics of the seasonal activities of bivalves.

In general, energy is stored prior to gametogenesis, when food is abundant, in the form of carbohydrate, lipid and protein. The particular importance of these substrates, where they are stored and the timing of their use varies among species, as well as among populations of the same species (Sastry, 1979, Ruiz et al., 1992; Ngo et al., 2006; Dridi et al., 2007). Bivalves generally store carbohydrates in large amounts during their growing season and use them over the rest of the year (Beukema, 1997); although proteins may be an energy reserve in some bivalve species (Galap et al. 1997). Lipids have been reported to function most importantly as energy storage substances and physical properties of biological membranes (Spector and Yorek, 1985). Lipid accumulates in the developing gonads and depletes during spawning.

Still to date there is no comprehensive research work on biochemical composition of bivalves from Bangladesh. Therefore, the present investigation was undertaken to know the seasonal changes in biochemical composition of bivalves from the freshwater ecosystem of northern Bangladesh.

2. Material and methods

2.1 Sample collection and preservation

The study was conducted in the freshwater lake situated in Rajshahi, the northern Bangladesh. The geographical location of the study site is 24º 22′ 21.56′′ N and 88º 38′ 11.16′′E (Fig. 3). The study was conducted during January, 2014 to July, 2015.

Monthly 30 specimens were collected by hand picking from the sampling site during January, 2014 to July 2015. After collection the sample, soft tissue was separated from the shell and dried at 60ºC for 48hrs and stored in refrigerator (-20 ºC) until further use for biochemical analysis.

2.2 *Biochemical analysis*

Dried meat of *L. marginalis* was pulverized using mortar and pestle and analyzed for its biochemical constituents. All the analysis was carried out and results expressed on an average basis.

2.2.1 Water contents

Water content (WC) of *L. marginalis* was determined by calculating the difference between the wet weight of the tissue and its weight after drying to constant weight at 60ºC and the results were expressed in percentage. The formula used for calculation of water content was,

WC= ((Wet tissue weight (g)-dry tissue weight (g))/Wet tissue weight (g)*100

2.2.2 Carbohydrate contents

Total carbohydrate content was estimated by the Phenol Sulfuric acid method (Taylor, 1995) using dextrose as a standard. Phenol and sulfuric acid were added to digested sample. The solution turns to a yellow orange colour as a result of the interaction between the carbohydrate and the phenol. The absorbance was measured using spectrophotometer at 490 nm, which was proportional to the carbohydrate concentration. The concentration was expressed as percentage of carbohydrate on dry weight basis.

2.2.3 Protein content

Protein content was estimated following the method of Lowry et al. (1951). To a 10mg of sample 1 ml of 1N NaOH was added for protein extraction in water bath at 37°Cfor 30 minutes. Thereafter, it was cooled at room temperature and neutralized with 1 ml of 1N HCL. The extracted sample was centrifuged at 2000 rpm for 10 minutes, and an aliquot of the sample (1 ml) was further diluted with distilled water (1/9 v/v). From the diluted sample, 0.5 ml was taken and made up to 1 ml with 0.1N NaOH. To this, 5 ml of mixed reagent (alkaline copper reagent) and 0.5 ml of FC reagent was added. After 30 minutes, optical density was read at 660 nm using spectrophotometer. The concentration was expressed as percentage of protein on dry weight basis.

2.2.4 Lipid content

Lipid was estimated by the method of Bligh and Dyer (1959). 50 mg of dried tissue sample was mixed well with 15 ml of chloroform-methanol mixture (1/2 v/v) and 4ml of distilled water. The homogenate was centrifuged at 2000 rpm for 10 minutes. The supernatant was taken in separating funnel and 5ml each of distilled water and chloroform was added and mixed well. After overnight separation the lower layer was collected in pre weighed ceramic bowl, dried in nitrogen stream and weighed. The concentration was expressed as percentage of lipid on dry weight basis.

2.3 Statistical analysis

Significant monthly differences in water content, carbohydrate, protein and lipid content of *L. marginalis* were tested using a One-way ANOVA with DUNCAN *post hoc* test. All statistical analyses were performed using SPSS (version 20.0) at $P < 0.05$ level of significance.

3. Results

The proximate composition such as protein, carbohydrate and lipid contents of *L. marginalis* tissue are presented in Fig. 47-50. The results of the present study revealed that the protein composition were high 38.88% followed by carbohydrate 21.25% and lipid 13.62%.

3.1 Water content

During the study the water content showed significant monthly fluctuations (ANOVA, $P<0.05$, Fig. 50). The water content was ranged from 74.66 to 81.88%. In 2014 water content was increased during January to July and then decreased up to November with a single fluctuation in August, when the water content was increased. In 2015 the water content showed several monthly fluctuations with similar trend as 2014 (Fig. 50).

3.2 Carbohydrate content

Figure 51 shows the significant seasonal variations in carbohydrate content of *L. marginalis* during the study period (ANOVA, $P<0.05$). At the first sampling month in January 2014 the carbohydrate content in the tissues of the *L. marginalis* was 19.83% and gradually increased 21.25% in the month of March. A sharp decline of carbohydrate was observed during March and April (17.75%) (Fig. 51). A slow decline of carbohydrate level was evidenced during April and July, indicating a high energy catabolism of the mussel. A progressive recovery of carbohydrate reserve was observed during July (16.25%) to December (20.1%) (Fig. 51).

3.3 Protein content

The protein content of the mussel showed significant monthly fluctuations and the average value was ranged 14.04-38.88% (ANOVA, $P<0.05$, Fig. 52). Protein content gradually increased during January (20.79%) and June (38.88%) and then decreased onwards at reaching the lowest value in the month of November (14.04%) (Fig.52). Protein content again increased from November to onwards. A higher protein level observed during June and July could be linked with the ripe gamete of the mussel and lower level of protein during August and October could be linked with the spawning release of gamete of the mussel (Fig. 52).

3.4 Lipid content

In the present study the lipid content showed significant monthly fluctuations and the average lipid content was ranged from 3.01% to 13.62% (ANOVA, $P<0.05$, Fig. 53). At the first sampling month in January, 2014 the lipid content was 5.3% and gradually increased to 9.59% in the month of February and then decreased up to April and then again increased to 12.06% in the month of June. A rapid decline of lipid content was observed during June to September (3.01%). Seasonal cycle of lipid content during January to July, 2015 was similar as January to July, 2014 (Fig. 53).

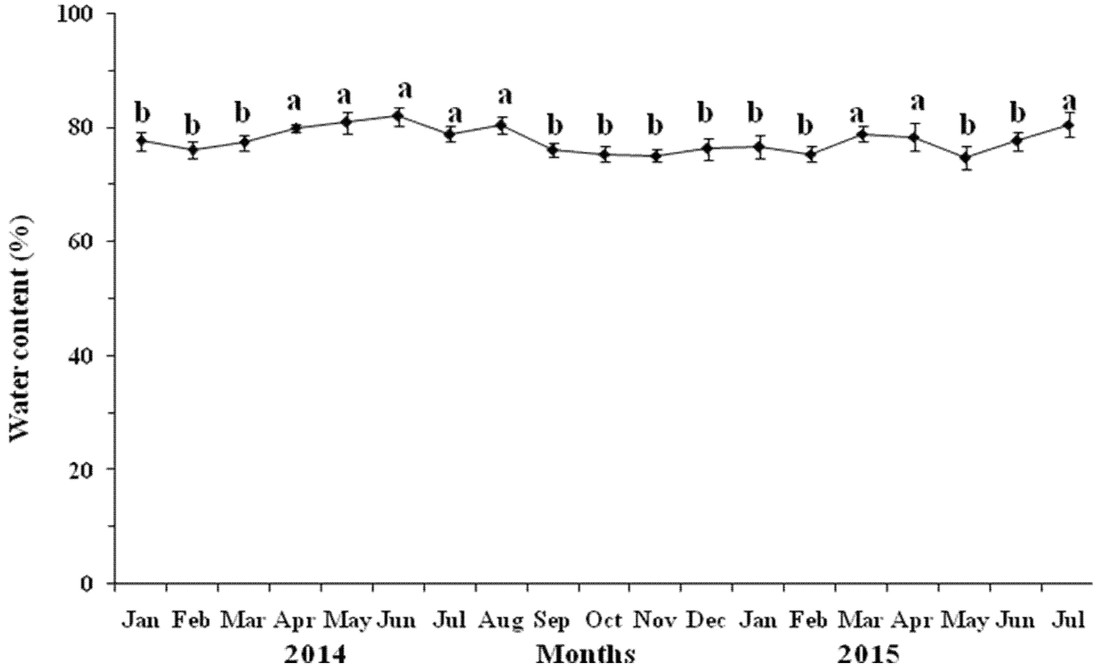

Fig. 50. Monthly variations of water content (%) in the tissue of the *L. marginalis* during the study period. Vertical bar represents standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

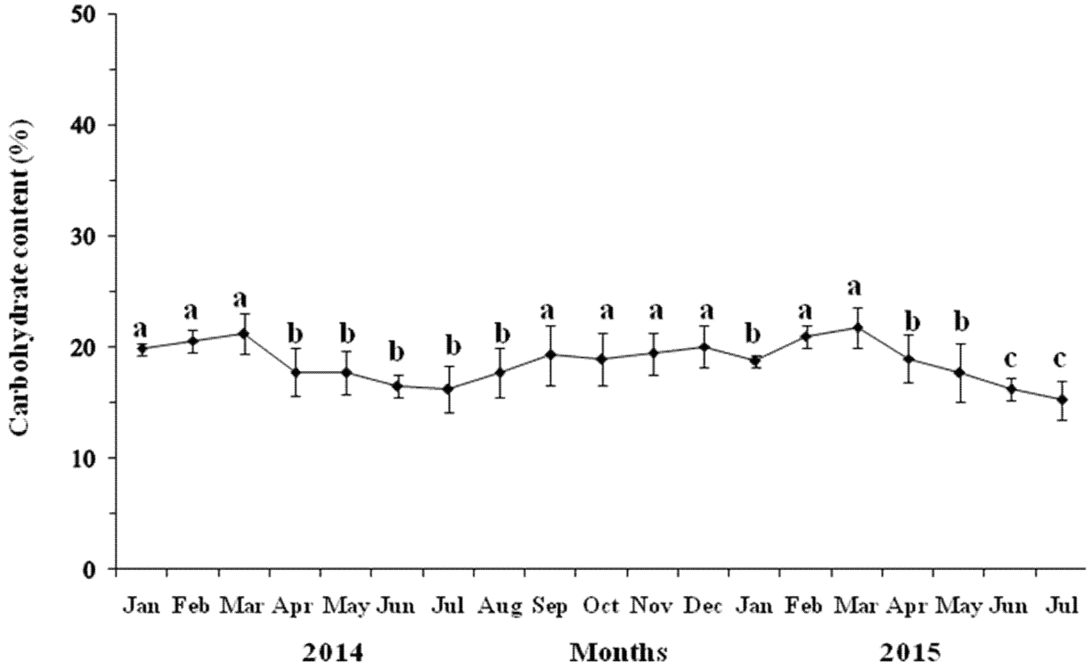

Fig. 51. Monthly variations of carbohydrate content (%) in the tissue of the *L. marginalis* during the study period. Vertical bar represents standard error and different

alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

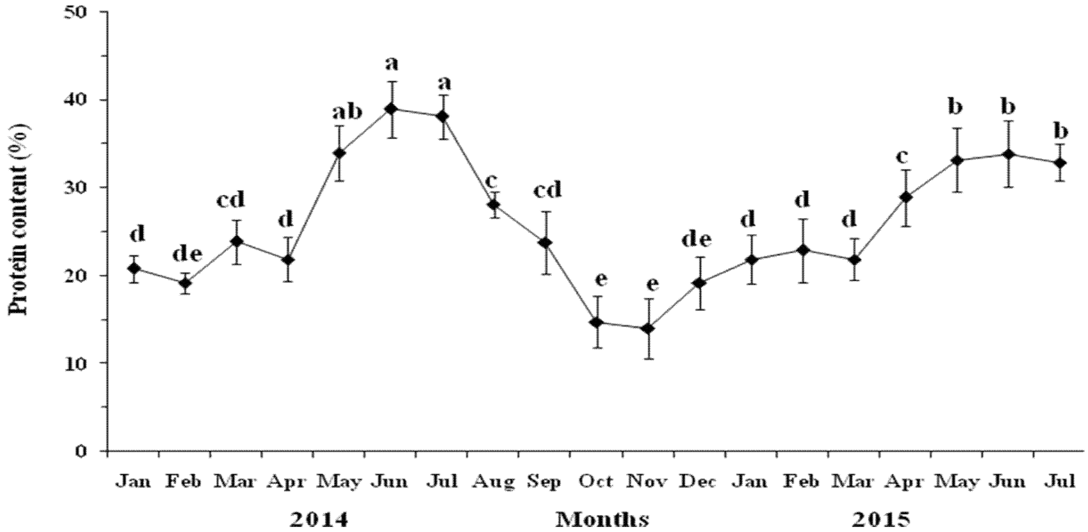

Fig. 52. Monthly variations of protein content (%) in the tissue of the *L. marginalis* during the study period. Vertical bar represents standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

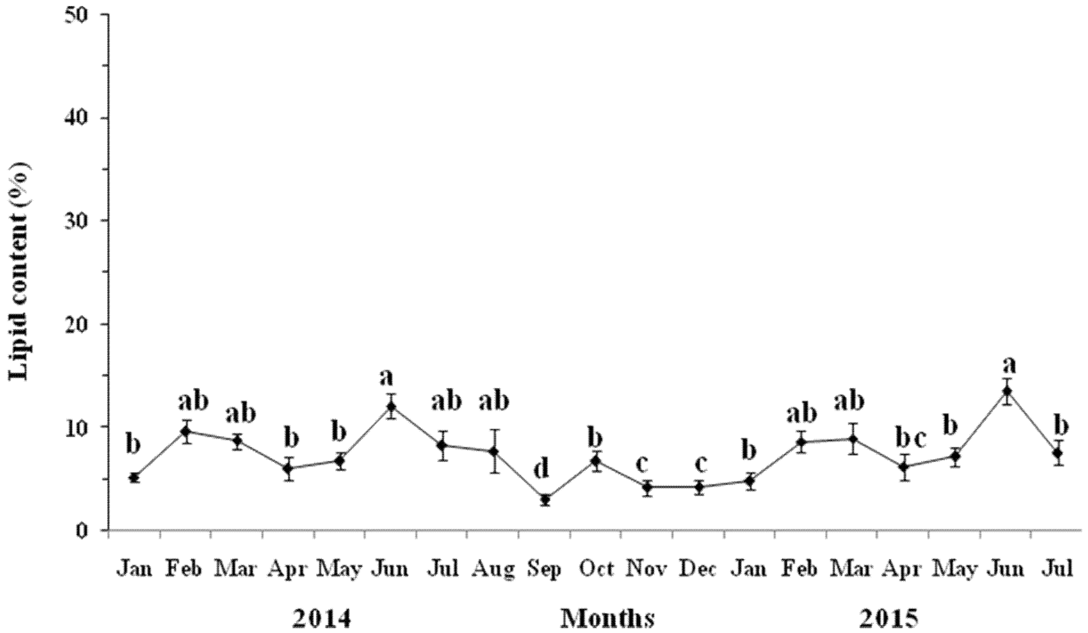

Fig. 53. Monthly variations of lipid content (%) in the tissue of the *L. marginalis* during the study period. Vertical bar represents standard error and different alphabets indicate the significant variation (ANOVA, Duncan's, P <0.05).

4. Discussion

Seasonal variation in the nutritional contents of the whole body tissues of *L. marginalis* was found to be, proteins (14.04-38.88%), lipids (3.01% to 13.62%) and carbohydrate (15.2%-21.7%) of dry tissue weight. Shetty et al. (2013) observed the seasonal changes in the biochemical composition of freshwater bivalves, *Parreysia spp.* from Tungabhadra River, Karnataka, India and reported proteins (41.2%-60.8%), lipids (3.8%-8.2%) and carbohydrate (14.79%-42.3%) for *P. favidens* and for *P. khadakvaslaensis* proteins (40.6%-57.2%), lipids (3.2%-7.6%) and carbohydrates (18.3%-40.2%) of total dry weight, which are more or less similar to the findings of the present study.

Protein is essential for the sustenance of life and exists in largest quantity of all nutrients as a component of the human body (Okuzum and Fujii, 2000). Carbohydrates are a group of organic compounds including sugars, starches and fiber, which is a major source of energy for animals. The lipids are highly efficient as source of energy, in that they contain more than twice the energy of carbohydrate and proteins. The present investigation also revealed the maximum level of protein content in *L. marginalis* body tissue in compare to lipid and carbohydrate. Periyasamy et al. (2000) also reported the maximum level of protein content in *D. incarnatus* body tissue from Cuddalore Southeast coast of India.

Seasonal cycle of biochemical composition in bivalve mollusks is closely linked to the annual reproductive cycle (Gabbot, 1975; Ruiz et al., 1992; Kang et al., 2003; Ojea et al., 2004; Ngo et al., 2006; Yang et al., 2011). Carbohydrates are considered as the main energy source and its variation is inversely related with the gonadal development (Gabbott, 1975; Camacho et al., 2003). In the present study the gonad development of *L. marginalis* seemed to occur along with the utilization of reserve carbohydrate originating from food consumed previously. The seasonal cycle of total carbohydrate content followed by the gametogenic cycle, the higher reserve observed during January and March when most of the mussels were in early and late gonad maturation condition (i.e. pre-vitellogenic stage) as reported in GI and GSI results. And then carbohydrate contents decline during April and July, indicating that the bivalve transformed the carbohydrate reserves into gonad particles during this period as a consequence of vitellogenesis (Deslous-Paoli and Héral, 1988; Marin et al., 2003; Ojea et al., 2004; Yang et al., 2011). After completion of spawning, the carbohydrate level again increased during August and December.

Seasonal changes in protein reserves are also closely linked with gonad maturation and may reflect the protein accumulation in oocytes; in addition, protein contributes to energy maintenance during the periods of reduced glycogen levels (Berthelin et al., 2000; Dridi et al., 2007). In this study, protein level increased with the advancement of gonad maturation and the higher content observed in June, when most of the mussels were in ripe condition as mentioned in histology result in Fig. 44-45. This higher protein level also coincided with higher egg content of the oysters as reflected in the reproductive effort (GSI) results in Fig. 49. The protein level sharply declined from July to onwards at reaching the lowest level in the month of November which was coincided with the spawning of the mussel. Several studies have reported that protein is the major component of the oocytes of bivalves, accounting for 40–50% of egg weight (Kang et al., 2003; Ngo et al., 2006; Yang et al., 2011). Ngo et al. (2006) reported that the summer protein maxima in the tissue of oyster coincided with GSI peaks in Gosung Bay, Korea, and postulated that seasonal changes in protein reserve were closely linked to the annual reproductive cycle of the oyster. In the present study protein represented a high level during this monitoring and the monthly highest protein level observed in June, corresponded with the highest GSI and could be associated with protein-rich eggs.

Lipid variation has been related to gamete development with the highest levels of lipids accumulation during the gonadal ripening. In the present study the lipids also exhibited large fluctuations. The increase in lipid content may be due to the lipogenesis occurring in the ovary for production of gametes. Lipids have also been shown to provide energy during the period, when carbohydrate reserves are depleted (Beninger and Lucas, 1984). Accumulation and depletion of these stored reserves in bivalves also depends on the environmental influences on metabolic activities, and the quantity and quality of available food (Ansell, 1972) and has been well described by several authors (Zandee et al., 1980).

General conclusion

This study provides the comprehensive baseline data on growth pattern, condition index, meat yield index, sex ratio, spawning season, reproductive effort and biochemical composition of the freshwater bivalve mollusk *Lamellidens marginalis* (Lamarck, 1819) from northwest Bangladesh. During this study the mussel showed a significant linear growth pattern for length-length relationships and non-linear growth pattern for length-weight relationships ($P<0.05$). The higher condition index and meat yield during winter and summer indicated a better period of commercial exploitation of the mussel from the natural ground. During the study sex of the mussel was differentiable in all the months. The overall sex ratio 1: 0.82 (M: F) was significantly different from the expected ratio 1:1 ($\chi^2 = 5.90$, $P < 0.05$). Histology revealed five gonad development stages as early developing, late developing, ripe, spawning, spent both for male and female mussels. The mussel exhibited similar gametogenic patterns for both sexes with a continuous breeding throughout the year and a peak spawning pulse during July and August. In the present study the 6-7 cm size group mussels were more potential group in respect to reproductive performance. Gonadal index increased progressively reaching the maximal value (9.41± 1.23) in June, indicates the maximum ripeness of the gonad. Gonadal index sharply declined from July to afterwards at reaching a minimal value of 4.83±0.96 in September, indicates the peak spawning of the mussel population during this time. During the study the biochemical composition of the mussel was found to be, proteins 14.04-38.88%, lipids 3.01%-13.62% and carbohydrate 15.2%-21.7% of dry tissue weight. Seasonal cycle of the biochemical composition of the mussel was closely linked to the annual reproductive cycle. Further study is needed to identify the timing and brooding pattern for better understanding about the life history of this important freshwater bivalve.

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Bio-data

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For the development of academic carrier as well as research out-looking Miss Nahar entered in PhD course in 2013-2014, under the Department of Fisheries, Faculty of Agriculture, University of Rajshahi, Bangladesh. During the PhD course she worked on the growth indices and reproduction pattern of freshwater bivalve mussel, *Lamellidens marginalis*, under the scholastic supervision of Professor Dr. Saleha Jasmine and Professor Dr. Md. Mostafizur Rahman Mondol, Department of Fisheries, Faculty of Agriculture, University of Rajshahi, Bangladesh. During PhD Miss Nahar published following papers-

1. Nahar, D.A., Islam M.R., Islam M.S., Jasmine S., Mondol M.R.M. 2019. Growth pattern of freshwater bivalve mollusk *Lamellidens marginalis* (Lamarck, 1819) from the northwest Bangladesh. J. Bio-Sci. 27: 121-132.
2. Mondol, M.R., Hossen, M.A., **Nahar, D.A**. 2016. Length-weight relationships of 3 fish species from the Bay of Bengal, Bangladesh. Journal of Applied Ichthyology, 33: 604-606.
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