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Research Article Breeding Biology of Mola Carplet, (*Amblypharyngodon mola*, Hamilton, 1822) in Semi-Natural Condition

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Abstract

Background and Objective: The mola carplet, *Amblypharyngodon mola* is a representative small indigenous fish species that is enriched with different vitamins and minerals. The main objective of this study was to determine the reproductive biology of *A. mola* for better scientific management and conservation in hapa based culture, set up in a large earthen pond. **Materials and Methods:** The experiment was conducted of three treatment with two replicates along hapa size varied treatment: T_1 (2 m³), treatment: T_2 (10 m³) and treatment: T_3 (20 m³) with two replicates each and the stocking density in those hapas were 50, 250 and 500g, respectively in April, 2012 to March, 2013. Some biological characteristics, gonad weight, Gonadosomatic Index (GSI) and condition factor (K) of both sexes were observed. **Results:** Cycle of gonadal maturation and month-wise variations were documented with their fecundity, size-frequency of intra-ovarian oocytes in females. Throughout the suitable physic-chemical parameters, monthly microscopic examination of gonad revealed five maturity stages of female *A. mola* but male showed same four stages. Ripe ova have been observed for nine consecutive months (April to December) which referring extended the breeding season. Considering the stocking density and hapa sizes, *A. mola* bred the highest frequency in T₃ during the culture period. Different size classes of intraovarian oocytes have portrayed the batch-wise (asynchronous) development of oocytes in *A. mola* throughout the culture period. **Conclusion:** This evidence suggests that *A. mola* has its inherent regulating mechanism of oocyte development adapted to the annual life of the fish, in spite of their single spawning habit, they discharge all oocytes in different batches throughout the spawning season.

Key words: Reproductive biology, gonadosomatic index, water quality, semi natural environment, spawning habit, oocytes

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Mola carplet (Amblypharyngodon mola), is locally known as moa, mowka, moraru, mouchi, mowrala, mola, maya and molongi in different region of Bangladesh. Its body moderately compressed, dorsal profile more convex than ventral, snout rounded, covered with skin and caudal deeply forked. Lateral line incomplete and extended up to 15 scales. Scale small, silvery color, a dark band runs on both sides of the body from head to tail, dorsal and anal with black edge¹. The A. mola is an indigenous small fish widely distributed in the rivers, canals, haors, baors, lakes, ponds, beels, floodplains, slow moving streams and paddy fields of Bangladesh, India, Myanmar and Pakistan² and is also reported from Afghanistan³. It is a popular food fish in Indian sub-continent because of its good taste and high nutritive value⁴. Very little focus has been given on the role that nutrient-dense small fish can play in preventing micronutrient deficiencies. Whole small fish with bone is a commonly consumed food by the poor people in developing countries having extensive fisheries resources, including Bangladesh. Moreover, small fish is culturally acceptable, can be collected on small bulk and wellliked by most household members including children^{5,6}. It is evident that like other population of fishes, natural abundance of A. mola is decreasing due to natural and other factors. Despite potential threats to small fishes, this species is yet to be identified and therefore the A. mola is categorized as least concerned by IUCN7. In recent times, it has also got its entry in ornamental fish trade and has been reported to be available in ornamental fish markets with moderate demand and availability⁸. Although various aspects of culture and farming of the species are known9-17, most farmers of the subcontinent do not include this species in freshwater aquacultures, because it is self-recruit species and in confined water compete with major carps for space and natural food as well as supplementary feed¹⁸. However, Roos et al.¹⁹ observed that A. mola could be cultured successfully in small seasonal ponds in polyculture with carps subject to availability of good seeds and minor modification of the culture techniques. The success of any fish species is ultimately determined by the ability of its members to reproduce successfully in a fluctuating environment. Reproductive parameters such as size at first maturity, spawning frequency, fecundity, sex ratio and recruitment are of great value in fishery prediction and formulation of management measures^{20,21}. No work has been done on the reproductive biology of *A. mola* in hapa system. A study of the reproductive biology of A. mola appears to be

essential in the sense that it may provide information and clues for the successful culture and capture of this species. In view of the economic importance and food value of this fish, this study was designed to collect the information of the reproductive biology of *A. mola* for better scientific management and conservation of this important fish species.

MATERIALS AND METHODS

Study area and design of the experiment: This study was started from April, 2012-March, 2013 in the Fisheries Field Laboratory, Bangladesh Agricultural University, Mymensingh. The experiment was carried out in a 43 decimal pond (1725 m²). The treatments consisted of three sizes of hapas which showed in Table 1 and Fig. 1.

The hapas were made of fine mesh net and hanged in water on bamboo poles. The commercial floating feeds Mega were applied at 10% body weight of *A. mola* twice in a day.

Water quality monitoring: The water quality parameters were determined once in a month. Water quality measurement and sample were collected between 08:00-10:00 hours in plastic bottles of 250 mL each with stopper and marked with hapa number. Water temperature was recorded with a Celsius thermometer. Dissolved Oxygen and pH of water samples were measured by a HACH 40 d Multi-parameter. Alkalinity of water was measured by the titration method with the help of 0.02 N H₂SO₄ and methyl orange solution. The concentration of nitrate-nitrogen (NO₃-N) and nitrite-nitrogen (NO₂-N) were determined by HACH kit (DR-2010, a direct reading spectrophotometer) using NitraVer-6 and NitriVer-3 powder pillow. Ammonia-nitrogen (NH₃-N) was also determined by HACH kit with Rochelle salt and Nessler reagent. The same HACH kit and Phosver-3 powder pillow was used to determine phosphate-phosphorus (PO₄-P). Chlorophyll-a was measured by spectrophotometer (Spectronic GENESYS 5) after acetone extraction following Starling²².

Sampling of Mola: Total fishes of each hapa were measured every month in the morning for weighing (electric balance, College B2002-S) and counting separately. During sampling hapas were cleaned by soft brushes for the removal of fouling

Table 1: Experimental design with size of hapa and stocking density

	-		
Treatment	Replication	Hapa size (m³)	Stocking density (g)
T ₁	R ₁ , R ₂	$1 \times 2 \times 1 = 2$	50
T ₂	R ₁ , R ₂	$2 \times 5 \times 1 = 10$	250
T ₃	R ₁ , R ₂	$4 \times 5 \times 1 = 20$	500

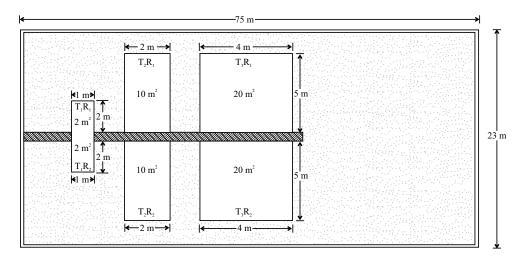


Fig. 1: Design of the experiment

algae, which sometimes blocked the net meshes of hapa. Some *A. mola* fishes were died during sampling time due to their fragile nature. Dead fishes were collected and used to determine of their reproductive characteristics.

Sex determination and fecundity estimation: Male and female fishes can be clearly distinguished in the breeding season. In the early stages, it was difficult to separate them from each other. For breeding purposes, it was necessary to separate male and female accurately through observation of some external characters. Males and females were different in color, males were comparatively brighter than females. The females were larger in size and in case of mature female, the abdomen was soft and swollen, pelvic fins were smooth and caudal fin was deeply forked. Mature females with distended abdomen had been easily recognizable during the spawning season. The length (cm) and weight (g) of fishes were measured by a measuring scale and an electric balance (College B2002-S), respectively. In every month, about 5 to 20 fishes were recorded both male and female for the determination of reproductive characteristics. Before weighing, the specimen was washed with water and left exposed to air and the excess of moisture was dried off with the help of a blotting paper. Usually, more than 1g fishes were selected for gonad separation. The collected fishes were dissected out by scissors starting from anus to lower jaw and the belly was opened. The whole internal mass (stomach, intestine and ovary) were removed carefully from the ovarian wall by means of fine forceps and soft brush and then gonad was weighed (g) by a high precision electric balance (PG 503-S Delta Range, Max 510 g d = 0.01/0.001 g, METTLER TOLEDO). General feature and structure as well as month-wise size, shape and color of gonads of the experimental fish were

recorded by open eye and electronic microscope. For fecundity estimation, the paired ovary of the individual fish was removed and carefully placed on a petridish. The ovary was washed and cleaned with distilled water. The weight of the ovary was taken by using electric balance. The ovary was then preserved in 10% buffered formalin for fecundity estimation. According to Nikolsky²³, male and female gonads have been grouped into different gonadal stages of development by using macroscopic and microscopic observation. And also some other information for the gonadal maturity stages has been gathered following Azadi and Mamun²⁴ and LeCren²⁵ study. Spawning periodicity has been determined by monthly evaluation of the Gonadosomatic index (GSI), condition factor (K) and mean monthly ova diameter. GSI and K value have been measured using the following formulas:

Gonado somatic index (GSI) =
$$\frac{\text{Gonad weight (g) \times 100}}{\text{Total body weight (g)}}$$

Condition factor (K) = $\frac{W \times 10^5}{W^2}$

L3

where, W is body weight (g) and L is body length (mm)

For greater efficacy over the other methods the gravimetric method²⁶⁻³⁰ was applied in the present study. Prior to estimation, oocytes from samples (0.01g) obtained from three portions (anterior, middle and posterior) two ovarian lobes randomly were counted and measured to determine whether they had significant differences between locations. Results showed that oocytes were uniformly distributed and in the succeeding analysis, samples were taken from the middle portion. A section of about 2 mm long was cut from

middle part of the right ovary, weighing to nearest 0.01 g after removal of formalin from the ovary surface with tissue paper and put into a Petri dish with a small amount of water. All the eggs were separated from each other with needle and measured along their longest axis under a stereo microscope. Oral saline solution was used when necessary for the separation of sticky eggs. Absolute fecundity was estimated on the basis of total weight of the ovaries using the following formula:

$$F = \frac{WO{\times}N}{W}$$

where, F is fecundity, WO is total weight of ovary, W is the weight of the sub-samples of ovary and N is the number of ova counted in the sub sample.

Ova diameter was calculated every month on ovaries collected at random from 10 mature fishes with the help of ocular and stage micrometers. Diameters of 10 ova were measured at random from anterior, middle and posterior region of each ovary and then the mean diameter was calculated.

Data analysis: All data were analyzed using SPSS for windows (version 20: SPSS Inc, Chicago, USA). One way analysis of variance (ANOVA) was performed and the mean values were compared using Tukey's test as post-hoc test.

RESULTS

Water quality: Temperature was found to range from 22.00-34.00 °C during the experimental period in hapa. The mean pH fluctuated between 7.32 and 8.02 and most of the small hapa was recorded two times in T_1R_1 and once in T_1R_2 throughout the experiment. Whereas, *A. mola* bred more frequently in treatment T_2 and T_3 than T_1 . The highest breeding frequency (four times) of *A. mola* was observed in T_1R_2 .

Table 2: Mean values (±SE) and range of water quality parameters in different treatments

time it was above 7.5. Total alkalinity ranged from 99.75-101.41 mg L⁻¹ (Table 2). The concentration of dissolved oxygen (3.47-3.54 mg L⁻¹), ammonia (0.127-0.167 mg L⁻¹), nitrate-nitrogen (0.014-0.046 mg L⁻¹) and nitrite-nitrogen (0.007-0.014 mg L⁻¹), phosphate-phosphorus (0.09-0.93 mg L⁻¹) were recorded during the experiment. The values of chlorophyll-a varied from 89.08-93.87 mg L⁻¹ in all the treatments. There was no significant differences in different parameters among the treatments. All water quality measurements were within the appropriate range for culture and also breeding of *Amblypharyngodon mola*.

Trend of fluctuation on number of fishes: During the culture period in hapa, several fishes were increased in different hapa due to their breeding. Considering the stocking density and hapa sizes were varied the total no of fishes obtained during the sampling. Next to breeding *A. mola* fingerlings were increased in every hapas. Although, the number of fishes decreased after the breeding of 1st and 2nd week. Stocking after an immediate month in May some fishes bred first time in T₃R₂ hapa (Fig. 2). The breeding performance of *A. mola* in

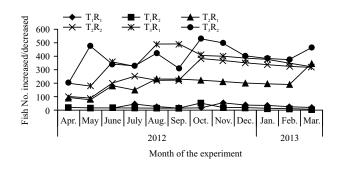


Fig. 2: Month-wise obtained number of fish increased/decreased trend in different treatment T_1R_1 : Treatment 1 and Replication 1, T_1R_2 : Treatment 1 and Replication 2, T_2R_1 : Treatment 2 and Replication 1, T_2R_2 : Treatment 2 and Replication 2, T_3R_1 : Treatment 3 and Replication 1, T_3R_2 : Treatment 3 and Replication 2 which is refer to Table 1

Table 2: Mean values (\pm	SE) and range of water quality pa	rameters in different treatments			
Parameters	T ₁	T ₂	T ₃	F-value	Level of significance
Temperature (°C)	29.37±0.62 (22.00-34.00)	29.37±0.55 (23.00-34.00)	29.29±0.60 (22.00-34.00)	0.006	NS
рН	7.92 (7.33-8.54)	7.97 (7.37-8.71)	8.02 (7.32-8.88)	0.289	NS
Alkalinity (mg L ⁻¹)	101.41±3.44 (75.00-130.00)	101.33±3.12 (74.00-125.00)	99.75±3.43 (72.00-134.00)	0.079	NS
DO (mg L ⁻¹)	3.47±0.15 (2.34-5.12)	3.54±0.16 (2.36-5.34)	3.50±0.15 (2.31-5.02)	0.056	NS
$NH_3 (mg L^{-1})$	0.139±0.12 (0.05-0.31)	0.167±0.03 (0.03-0.99)	0.127±0.01 (0.01-0.24)	0.776	NS
$NO_3 (mg L^{-1})$	0.046±0.012 (0.01-0.22)	0.029±0.006 (0.01-0.14)	0.014±0.004 (0.00-0.06)	0.681	NS
$NO_2 (mg L^{-1})$	0.007±0.001 (0.00-0.02)	0.013±0.004 (0.00-0.10)	0.014±0.003 (0.00-0.06)	1.510	NS
$PO_4 (mg L^{-1})$	0.93±0.08 (0.20-1.55)	0.87±0.09 (0.13-2.08)	0.09±0.07 (0.15-1.54)	0.107	NS
Chlorophyll-a (µg L ⁻¹)	93.87±3.96 (66.00-134.00)	89.08±3.77 (55.00-123.00)	94.54±3.84 (69.00-133.00)	0.594	NS

**Here, DO: Dissolve oxygen, NH₃: Ammonia, NO₃: Nitrate nitrogen, NO₂: Nitrite nitrogen, PO₄: Phosphate phosphorus; °C: Degree centigrade, mg L⁻¹: Milligram per Liter, T: Treatment

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Table 3: Monthly mean (\pm SE) body weight, total length, gonad weight, GSI (%) and condition factor (K) of male f

Month	Body weight (g)	Total length (cm)	Gonad weight (g)	GSI (%)	Condition factor (K)
April, 2012	1.431±0.018	5.166±0.149	0.019±0.001	1.363±0.129	1.079±0.087
May, 2012	1.542 ± 0.042	5.380±0.489	0.022 ± 0.001	1.472±0.067	0.988±0.014
June, 2012	1.761 ± 0.032	5.520±0.075	0.033±0.001	1.891±0.051	1.058±0.051
July, 2012	1.613±0.042	5.510±0.034	0.028±0.001	1.677±0.062	0.998±0.011
August, 2012	1.613±0.042	5.520±0.064	0.024±0.001	1.520±0.073	0.958±0.015
September, 2012	1.651±0.049	5.670±0.097	0.028±0.001	1.717±0.037	0.910±0.030
October, 2012	1.589±0.047	5.680±0.066	0.023±0.001	1.495±0.077	0.966±0.026
November, 2012	1.519±0.033	5.350±0.054	0.021±0.001	1.415±0.071	0.991±0.024
December, 2012	1.494±0.030	5.370±0.026	0.020±0.000	1.343±0.035	0.964±0.013
January, 2013	1.431±0.032	5.360±0.042	0.016±0.000	1.143±0.045	0.928±0.011
February, 2013	1.302 ± 0.041	5.020±0.032	0.015 ± 0.000	1.179±0.033	1.027±0.023
March, 2013	1.518±0.023	5.380±0.032	0.019±0.001	1.258±0.070	0.975±0.019

Here, GSI: Gonadosomatic index

Table 4: Monthly mean (±SE) body weight, total length, gonad weight, GSI (%), fecundity and Condition Factor (K) of female fish

Month	Body weight (g)	Total length (cm)	Gonad weight (g)	GSI (%)	Fecundity	Condition factor (K)
April, 2012	3.398±0.106	6.660±0.130	0.337±0.014	10.115±0.507	4093±195.22	1.178±0.066
May, 2012	3.778±0.079	6.725±0.097	0.528±0.017	14.029±0.439	5760±241.03	1.256±0.041
June, 2012	5.369±0.152	7.405±0.088	0.915±0.058	16.751±0.710	9860±481.89	1.329±0.043
July, 2012	4.937±0.187	7.310±0.099	0.710±0.046	14.318±0.670	7881±412.86	1.257±0.026
August, 2012	4.519±0.178	7.005±0.110	0.561 ± 0.025	12.393±0.159	6216±394.17	1.315±0.036
September, 2012	5.278±0.219	7.335±0.129	0.816±0.057	15.283±0.676	8812±722.10	1.360±0.037
October, 2012	3.147±0.086	6.500±0.695	0.384±0.017	12.301 ± 0.586	4180±354.12	1.140±0.853
November, 2012	2.893±0.139	5.935±0.135	0.266±0.018	9.240±0.473	3016±216.08	1.400 ± 0.064
December, 2012	2.862 ± 0.084	6.030±0.064	0.210 ± 0.008	7.397±0.318	2541±161.35	1.320 ± 0.043
January, 2013	3.058±0.047	6.240±0.086	0.173±0.006	5.648±0.159	2210±123.60	1.275±0.042
February, 2013	3.250±0.108	6.600±0.061	0.257±0.009	8.067±0.388	3200±213.60	1.128±0.028
March, 2013	3.398±0.125	6.750±0.080	0.295±0.013	8.757±0.325	3625±265.35	1.104±0.035

Here, GSI: Gonadosomatic index

Gonadal maturation, gonadosomatic index (GSI) and condition factor (K): The highest and lowest gonad weight was found 0.033 ± 0.001 g (June, 2012) and 0.015 ± 0.000 g (February, 2013) in male *A. mola*. The mean GSI value of male has shown two peaks; once in June and another in September. The GSI value of male *A. mola* was found from 1.143 ± 0.045 (January, 2013) to 1.891 ± 0.051 (June, 2012). The mean values of condition factor (K) in male *A. mola* were found from 0.910 ± 0.030 (September, 2012) to 1.079 ± 0.087 (April, 2012) (Table 3).

The lowest and highest gonad weight of female *A. mola* were found 0.173 ± 0.006 g (January, 2013) and 0.915 ± 0.058 g (June, 2012), respectively. The GSI value of female *A. mola* were found from 5.648 \pm 0.159 (January, 2013) to 16.751 \pm 0.710 (June, 2012). The mean values of condition factor (K) in female *A. mola* were found 1.140 \pm 0.853 (October, 2012) to 1.400 \pm 0.064 (November, 2012), respectively (Table 4).

Four maturity stages of testes have been recognized which are as follow:

• **Immature:** Testes were very tiny and thread-like in appearance; whitish. The two lobes were fused, appearing like a single structure.

- **Mature:** Testes enlarged in size and weight and palewhitish. Two lobes close but not joined together.
- **Ripe:** Testes enlarged in size and weight and yellowishwhite. Milt came out on putting light pressure on the abdomen.
- **Spent:** Testes darker, but still milky, opaque, flaccid and shrunken in size.

Five maturity stages of the ovary have been recognized which are as follows:

- Immature: Ovaries were small, pale pink and thread-like in appearance. Ova was not visible to naked eyes, but under microscope, ova were irregular in shape with a central nucleus and transparent as yet yolk was not formed.
- Maturing: Ovaries were larger than previous stages, white-yellowish in color and still thread-like. Ovaries were visible, under microscope ovary was round and slightly opaque due to the deposition of yolk at the central part.
- Mature: The ovary became enlarged and light yellowish. Ova was clearly visible to naked eyes, under microscope it was spherical in shape and the nucleus was visible due to heavy deposition of yolk.

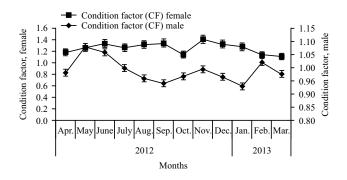


Fig. 3: Monthly variation (±SE) of Condition Factor (CF) of male and female *A. mola*

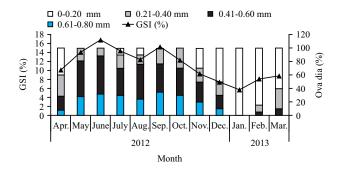


Fig. 4: Gonadosomatic Index (%) and Ova dia (%) of female *A. mola*

- Ripe: The ovary became deep yellow and grew in size. Under microscope, ova were found round in shape and opaque due to huge amount of yolk present. Eggs were extruded with slight pressure on the abdomen.
- **Spent:** Ovaries were pale whitish in color. Different stages of ova were found, mostly immature ones only with a few ripe ones. Ovaries were shrunken in size and flaccid.

The gonadosomatic index is the indicator of gonadal development status. In comparison month-wise condition factor (K) in both male and female were shown in Fig. 3.

Size frequency of intra-ovarian oocytes: Ova have been grouped in following four size classes: immature ova (0.0-0.20 mm), maturing ova (0.21-0.40 mm), mature ova (0.41-0.60 mm) and ripe ova (0.61-0.80 mm) (Fig. 4). The highest percentage of immature ova (0.0-0.20 mm) have been observed in January. Maturing and mature ova were observed round the year except January. But the highest percentage of mature ova was widely observed from May to September. The ripe ova were much prominent during June-July and September-October and also disappeared in January-March.

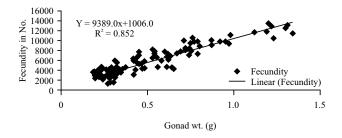


Fig. 5: Relationship between fecundity and gonad weight of *A. mola*

The relationship of fecundity with gonad weight was determined (Fig. 5). The scatter diagram obtained through plotting fecundity against gonad weight showed a positive correlation. In the process of analysis the equation was expressed as:

$$Y = 9389.x + 1006 (R^2 = 0.852)$$

DISCUSSION

Amblypharyngodon mola was stocked when the ponds were green and rich in dense plankton due to fertilization. Perhaps the beginning of the experiment causes of *A. mola* mortality due to their respiration. After the commencing of the experiment, *A. mola* bred first time in the month of May in big hapa (Fig. 2). Actually *A. mola* bred round the year except in December, January, February and April. From the three treatments, large hapas (Treatment T₃) were more suitable for *A. mola* breeding which seems like semi-natural pond condition. It has been suspected that the brood of mola die after breeding. A similar observation was reported by Babu and Nair³¹, Suresh *et al.*³², Ahamed *et al.*³³ and Saha *et al.*³⁴.

In male, the highest body weight, total length, gonad wt. and GSI were found in peak breeding month, June when *A. mola* bred in three hapas (Table 2). The result is in agreement with the finding of Kohinoor⁹ and Gupta and Banerjee³⁵ in male *A. mola*. In the present study of all the previous parameter again increased another peak period in September-October. The GSI of males showed two distinctive peaks once in June and another in September; whilst the lowest GSI value was observed in January. Shankar and Sarala³⁶ also reported higher GSI for *A. mola* in June.

In female, body weight, total length, gonad weight and GSI of females were observed high value similarly with male in June (Table 3). In females, GSI has been observed almost the

same trend just like male to reach peaks twice a year, once in June and another in September whilst the lowest value of GSI has been observed in the month of January. Das and Dewan³⁷ examined average mean length (6.5 cm) and weight (2.78 g) of *A. mola* in both sex which was more or less near about the present study. The mean GSI values of both sexes were more or less similar with Gupta and Banerjee³⁵ and Kohinoor⁹. LeCren²⁵, Olurin and Savage³⁸ and Nikolsky²³ revealed that GSI tends to increase with the initiation of breeding season which becomes maximum during the peak breeding period and declines abruptly thereafter when the fish became spent after gamete extrusion and/or re-absorption. During maturation, the GSI values of females have been observed to be much higher than males implying a greater proportion in body reserves for that allocation to the gonads³⁹.

Condition factor (K) was used for comparing the condition of fatness, or wellbeing⁴⁰ of fish based on the assumption that heavier fish of a given length is better condition. In the present study, monthly condition factor values of the female have shown two peaks; in October and November; lowest condition factor in March indicates first maturity. And also in male, lowest 'K' value perhaps second/third time maturation while the fish was in poor physical condition that report similar in landlock ayu, *Plecoglossus altivelis* fishes⁴¹. In male *A. mola*, inflection point on curve showing the diminution of 'K' with increase in length, which indicated the length at which sexual maturity was attained⁴². This feature also properly applied in many fishes⁴³⁻⁴⁵. The fluctuation of condition factors in different groups may be attributed to spawning and following recovery, gonadal development and also on the general condition of seasonal apetite⁴⁶.

The large number of eggs reported in June and July and the lowest in December and January. In the present study, the fecundity of *A. mola* ranged from 2210-9860, which more or less lies down within those previous findings. Dewan and Doha²⁸ reported the fecundity of *A. mola* to be ranged from 1,021-13,812. Misra and Jain⁴⁷ have documented the fecundity of *A. mola* to be ranged from 1,210-16,072. Azadi and Mamun²⁴ observed the fecundity to range from 1,280-13,679 with an average of 5,182.67 ± 3,731.51. Saha *et al.*⁴ revealed that the fecundity ranged from 1,291-12,737 with mean value of 5,752 ± 3,322. Gupta and Banerjee³⁵ obtained the fecundity of *A. mola* ranged from 1,014-9,690 with an average of 4,593. Mondal and Kaviraj⁴⁸ have documented fecundity range of 3,785-12,590 for the *A. mola*.

First mature gonad of males was observed in April and available till October and the highest was observed in June and the lowest in November. Whilst ripe gonad of males have been observed from June to December and the highest was observed in July while the lowest in December. Females with immature gonads have been observed from December to May; the highest (100%) was observed in January while lowest in August. Mature gonad of females has been observed from May to November and the highest was observed in June and the lowest in December. Ripe females have been observed from April to December with the highest was observed in September and the lowest in April. Spent females have been observed in November and December but the highest was observed in December and the lowest was in November. In both sex of A. mola, Hoque and Rahman49 observed five maturity stages (immature, maturing, immature, ripe and spent) whereas Azadi and Mamun²⁴ have reported, female consists three gonadal maturity stages (immature, maturing and ripening). Gupta and Banerjee³⁵ revealed five maturity stages (immature, maturing, mature, ripe and spent) for female and four maturity stages (immature, mature, ripe and spent) for male A. mola, those are similar with the present study. Suresh et al.32 reported A. mola breeding season from April to October in West Bengal, while Gupta and Banerjee³⁵ have observed mainly two spawning months in June and November, but continued through April to December. In West Bengal, this fish species bred in July as reported by Mondal and Kaviraj⁴⁸, but in Bangladesh, Rahman⁵⁰ reported May to October as its spawning months. Afroze and Hossain⁵¹ also observed August as the peak breeding season of A. mola. The breeding period of A. mola was with the ranges as mentioned by Parveen⁵², Hogue and Rahman⁴⁹ and Saha *et al.*⁴.

During the stocking month, the fishes bear four size of ova diameter where the higher (40%) of immature size among all. Then mature ova (0.41-0.60 mm) sharply rose in May (53%) and June (57%). The immature ova (0.00-0.20 mm) were absent in May-June and September-October which was the peak breeding time. The mature ova were present all of the months except January which is similar with Gupta and Banerjee³⁵.

The scatter diagram obtained from the fecundity and gonad weight showed a linear relationship. The value of coefficient of determination ($R^2 = 0.852$) indicated that the gonad wt. explained 85% of total variation in fecundity. The linear relationship between fecundity and ovary weight was observed in case of *A. mola* and puti agrees with finding of Mustafa *et al.*⁵³, Azadi *et al.*⁵⁴ and Das *et al.*⁵⁵.

Any surveillance electronic devices (CCTV camera, drone etc.) were not assigned for continuous field monitoring of this semi natural experimental culture setup; which could be considered as main limitation of this study. To overcome, a complete surveillance kit can be installed for further study of *A. mola* as well as other species to reveal the breeding biology of aquaculture species in semi natural condition.

CONCLUSIONS

In the present study some aspects of the reproductive biology of *Amblypharyngodon mola* has been revealed. The breeding season extends from April to December with two peaks in June and September. Fractional spawning behavior along with asynchronous oocytes development has also been observed and the mortality of some *A. mola* broodfish within a short period after breeding has development speculation that the broodfish of *A. mola* die after breeding. An intensive observation on the breeding biology may be further suggested in a large aquarium with close circuit camera.

SIGNIFICANCE STATEMENT

This study was the very first approach to determine the reproductive biology of *Amblypharyngodon mola* for better scientific management and conservation in confined hapa conditions in Bangladesh. This present finding will help to the related researchers, hatchery owners as well as fish farmers to select the appropriate broodstock for breeding in hatchery conditions and aquaculture of this species in the polyculture system. Thus a new auxiliary fish species for polyculture with carps may be arrived at which bring more nutritional and economic value to relevant practitioner.

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