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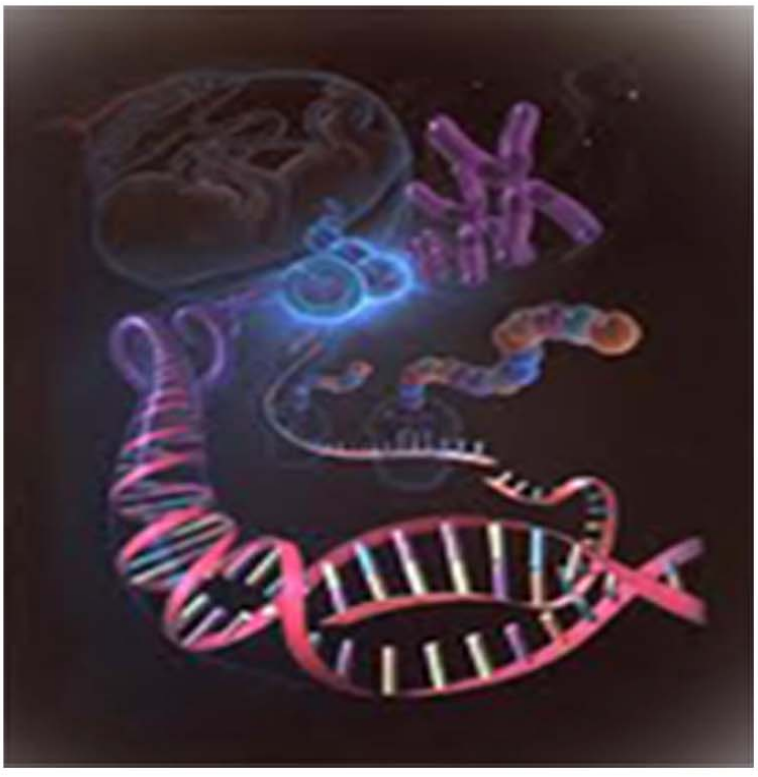

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Research Paper

REPRODUCTIVE BIOLOGY AND GONAD HISTOLOGY OF MUD EEL, *MONOPTERUS CUCHIA* (HAMILTON, 1822)

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A study was performed to understand the reproductive biology and gonad histology of freshwater mud eel *Monopterus cuchia* for a period of 6 months. Gonadosomatic Index (GSI) and ova diameter were found to range between 0.31 to 1.85 and 0.53 mm to 4.20 mm, respectively. The GSI value chromonologically increased and found to peak in June (1.85). The highest ova diameter reached in June. Mean absolute and relative fecundity were 645.13±92.12 and 2.52±0.29, respectively. The percentage of Early Perinucleolus (EPN) stage was highest in March (7.67±1.53%) while Late Perinucleolus (LPN) stage was highest in April (31.00±2.65 %) and Cortical Alveoli (CA) stage appeared from April and reached to maximum in May (10.67±2.08%). Vitellogenic stage (VG) was found in ripe phase, prevailed from May to June and chronologically increased and highest percentage of ripe oocytes were recorded in June (33.67±3.51%).

Keywords: Threatened fish, Mud eel, GSI, Gonad histology, Fecundity

INTRODUCTION

Monopterus cuchia (Hamilton, 1822), is an obligate air-breathing mud eel inhabiting the swamps and derelict water bodies. Thus *M. cuchia* commonly known as mud eel or swamp eel which belongs to Synbranchidae family under Synbranchiformes order. This is a delicious and nutritious freshwater fish species. The species commonly occurs in Bangladesh, Pakistan, Myanmar, Nepal and India (Jingran and Talwar, 1991). Especially in Bangladesh, mud eel is generally available in open water resources such as haors, baors, beels, canals and floodplains (Hasan *et al.*, 2012).

M. cuchia is a threatened fish species of Bangladesh (IUCN, 2000). Abundance of this species in nature has been declined due to heavy fishing pressure, habitat destruction, aquatic pollution and indiscriminate uses of pesticides. Though the fish is only consumed by the tribal people in Bangladesh. The fresh blood of cuchia is directly consumed to cure weakness, anemia, asthma (Jamir and Lal, 2005; Kakati *et al.*, 2006), some people consumed gall bladder of cuchia to cure asthma, anemia, piles and diabetes (Saikia and Ahmed, 2012; Chakravarty and Kalita, 2012). However, *M. cuchia* is commercially important due to its high demand for export.

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The reproductive biology of a fish might be defined by its reproductive traits and it also expresses the combination of the species-specific reproductive mode (Winemiller and Rose, 1992; Murua and Saborido-Rey, 2003; Morgan, 2008). Doha and Hye (1970) opined that knowledge on reproductive physiology of any fish species is essential for evaluating the commercial potentialities of its stock, life history, culture practice and management of its fishery. Reddy (1979), mentioned the determination of breeding season is an essential part of biogenic investigations of fishes.

Vitellogenic stage (VG) of oocyte as well as increased gonad weight, GSI value and gonadal histology indicate the maturity of mud eel and also definite the breeding season. Very little information is available on the reproductive biology of mud eel. So a scientific research attempt was made on to evaluate the GSI value, ova diameter and fecundity of *M. cuchia*.

Therefore, this study will give information about reproductive biology of mud eel in Bangladesh ecological condition.

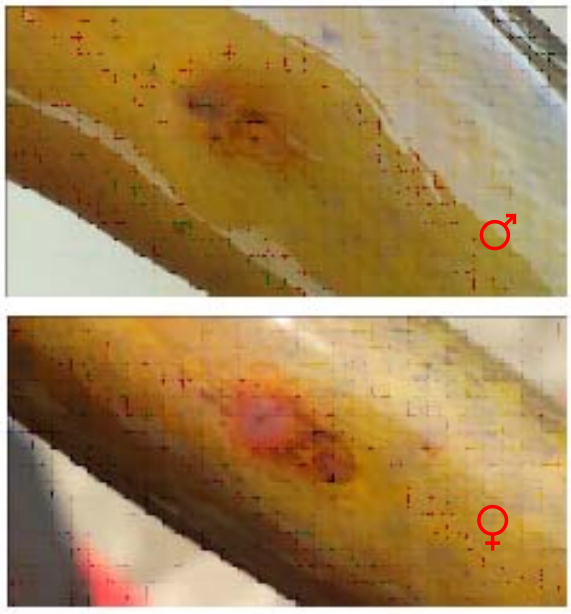
MATERIALS AND METHODS

The present study was conducted for a period of six months from January to June 2012 in the Freshwater Station, Bangladesh Fisheries Research Institute, Mymensingh, Bangladesh. Live 400 *M. cuchia* samples weight varied from 200 to 400 g were collected from different area of Mymensingh district during November to December, 2011 and kept them in hapa for conditioning and then the eels were transferred in the cemented cistern where eight inches thick clay was used as soil layer. Live halencha and water hyacinth were used in cistern as shade and shelter for *M. cuchia*. The eels were fed with live tilapia fry, SIS and fish paste. After sunset fish paste was applied on the surface of water hyacinth.

Collection of Gonad Sample

During experimental period *M. cuchia* was sexually separated (Plate 1), although reports are not available on sexual dimorphism of eels. Narejo *et al.* (2003) reported that in *M. cuchia* there are no secondary sex characters for sexing. They identified male and female eels on external characters during breeding season. During the experimental period only female gonad was used. For the experiment ten gravid eels were randomly collected in each month from the cistern using plastic bowl where compost and earth warm were used. Collected eels were transferred to the laboratory and weight was recorded in gram. The peritoneal cavity was opened and collects the gonad. Collected gonads were preserved in Bouin's fixative for further analysis.

Plate 1: Sexual Dimorphism of Mud eel (*M. cuchia*)



Determination Procedure of Breeding Parameters and Reproductive Biology

Fecundity, fertilization, hatching, incubation time, yolk sac absorption time and larval size are the breeding parameters. Among the breeding parameters fecundity and GSI value and ova diameter were observed according to the following method:

Gonadosomatic Index (GSI)

Gonads status and weight weighed on electronic balance before being preserved in Bouin's fixative for subsequent studies. Total body weight of selected fishes in each month was considered to calculate the mean Gonadosomatic Index (GSI). GSI was calculated according to the following formula:

Weight of gonad

$$GSI = \frac{\text{Weight of fish}}{\text{Diameter of ova}} \times 100$$

After dissecting the ovary, representative sample from each part of anterior, posterior and middle portion of the ovary was collected. The ova from each samples were separated using physiological saline solution (0.65% NaCl) in a petridish and then spreaded on a glass slide to measure the diameter of ova under a microscope using an ocular micrometer. The units of the ocular micrometer which were visible in the microscope were standardized with a stage micrometer for measurement of ova diameter in micrometer (µm). Approximately 100 ova from each sample were measured and determined the ova diameters.

Fecundity

The term "fecundity" can be expressed as the number of eggs laid in a single in one season by the species. For the estimation of fecundity gravimetric method was used according to Blay (1981) and gonad was removed as early as possible. For fecundity estimation, 10 gravid female were (50-65 cm in length) were used. Sample of anterior, middle and posterior region of ovary was weighed separately and the number of ova present in each sample was recorded and then fecundity was estimated on the basis of total weight of the ovary. The absolute fecundity was calculated as suggested by Grimes and Huntsmen (1980). It was obtained by using the following formula:

$$F = N \times G\ g^{-1}$$

where, F = fecundity, N = no. of eggs in sub sample, G = total weight (g) of ovary and g= weight (g) of the sub-sample.

The relative fecundity was estimated by simply dividing the absolute fecundity with total body weight (g).

Histology of Gonad

The ovary tissues were fixed in aqueous Bouin's fluid and then histological process was followed for slide preparation. According to Humason (1979), the gonadal tissues were serially sectioned at a thickness of 4 µ and stained with hematoxylin-eosin. Nuclear diameters and physical feature of the oocytes were observed under microscope and different developmental stages were identified.

RESULTS AND DISCUSSION

Gonad Observation

M. cuchia ovary was cylindrical shaped with single lobed lying in the body cavity. The size and extent of occupancy of the body cavity were found to

vary with size and maturity of the females. Silas and Dawson (1961) also reported that in mature eel, *Amphipnous indicus has* single-lobed ovary to below the liver. Ovary showed the highest weight in June and also indicating the mature stage. The immature ovary was compact and cream color whereas matured one was yellowish in color (Plate 2).

Plate 2: Physical Appearance of *M. cuchia* Ovary

Gonadosomatic Index and Ova Diameter

GSI, the indicator of the status of gonadal development and maturity of any species. During observation period of this experiment, GSI value ranged from 0.31±0.83 to 1.85±0.91. The highest value of GSI was recorded in June and lowest was recorded in January. GSI value chronologically increased from January to June and monthly variation of GSI value are graphically presented in Figure 1. Whereas Rahman *et al.* (2009) reported that in spiny eel maximum GSI value (female) was recorded in March and minimum was in May.

The observed ova were spherical and uniform in diameter. The ova diameter progressively increased from 0.53 to 4.2 mm. During the study period ova diameter were: 0.53-0.86 mm (January-February), 1.50-2.75 mm (March to April), 3.50-4.20 mm (May-June). Monthly variations of ovary weight and ova diameter of *M. cuchia* are presented in Figure 2. Similarly, Narejo *et al.* (2003) in *M. cuchia* the ova diameter was found to vary from 0.30 mm to 4.00 mm and Munshi *et al*. (1989) reported that *M. cuchia* lays large yolky eggs (4.539±0.107 mm diameter) during May-June. From the above observation of GSI value and ova diameter, it can be concluded

Figure 1: Monthly Variation of Gonado-Somatic Index (GSI) of *M. cuchia* with Peak in June

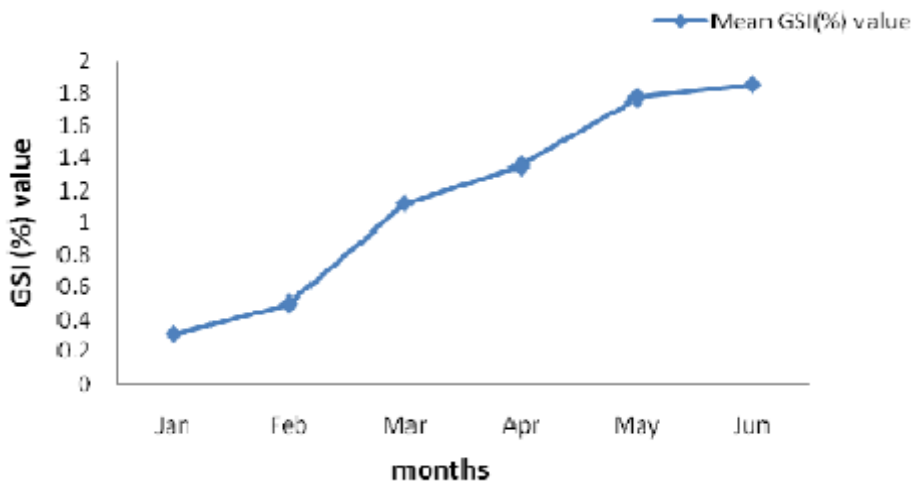


Figure 2: Monthly Variations of Ovary Weight and ova Diameter of *M. cuchia*

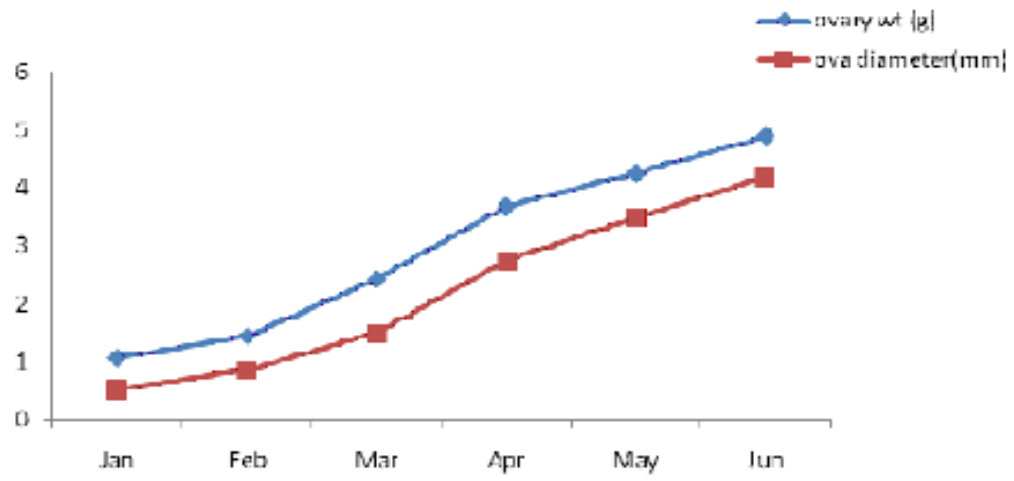
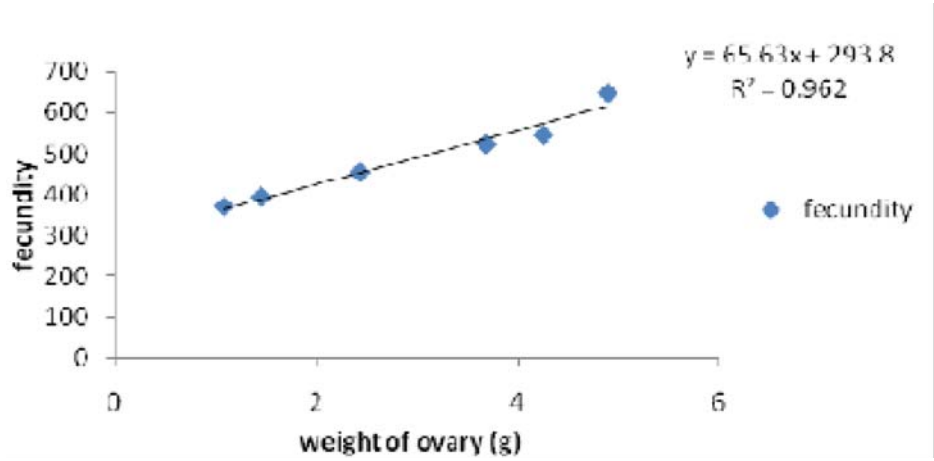


Figure 3: Relationship Between Fecundity and Ovary Weight of *M. cuchia*



that the peak spawning season of cuchia is May to June. Singh *et al.* (1989) and Narejo *et al.* (2003) described the spawning season of *M. cuchia* in May to June.

Fecundity

Among different breeding parameters fecundity is one of them. According to Lagler *et al*. (1956) fecundity of fish is a prerequisite of breeding program, i.e., fecundity is an indicator of reproductive potential of any species and Qasim (1973) mentioned that knowledge on the fecundity of a fish species is important for determining: spawning potential and its success.

During the present study, fecundity of eel was ranged 370-647 eggs from the body weight 225-380 g. The scattered diagram analysis shows a nearly perfect positive linear relationship between fecundity and body weight, where the calculated

values of regression coefficient, intercept and coefficient of correlation were 65.636, 293.88 and 0.481, respectively (Figure 3).

Nasar (1989) reported that fecundity of *M. cuchia* was ranged from 118-687 eggs and Narejo (2003) found 260-5890. Bromage *et al*. (1992) opinioned the fecundity varied with the seasons, climatic conditions and environmental habitat, nutritional status and genetic potential. Bagenal and Braum (1978) and Fagade *et al.* (1984) mentioned different causes for the variation of fecundity. The absolute fecundity (645.13±92.12) and relative fecundity (2.52±0.29) of the present study are shown in Table 1. Whereas Narejo *et al*. (2003) opinioned the average number of ova per gram of body weight was 4.61.

Histology of Gonad

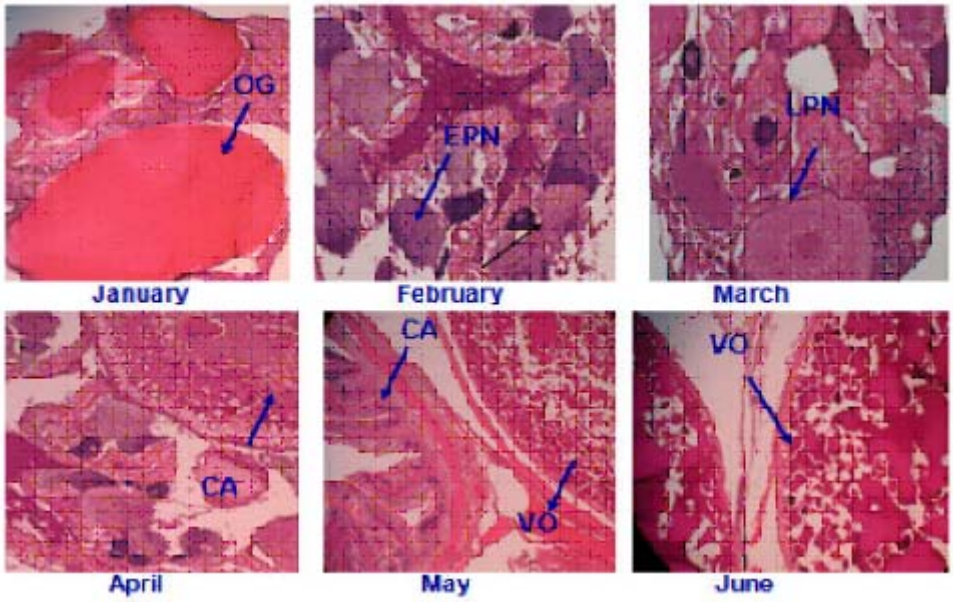
In case of seasonal breeders the changes of gonads are demarcated in different phases such as immature phases, maturing phases, mature phases, ripe phases, spent phases and resting phase. Mollah (1986) observed the oocyte maturation in catfish, *Clarias macrocephalus,* and identified seven stages: oogonia, chromatin nucleolus, early perinucleolus, late perinucleolus, yolk vesicle, early yolk-granule and late yolk-granule stage. During observation period the developmental stages of oocyte were differentiated on the basis of size and appearance of nucleolus and cytoplasm. Most of the oogonia (OG) were found in immature phase/early stage which prevailed from January to February. Early perinucleolus stage (EPN) and late perinucleolus stage (LPN) was distinguished in maturing phases which prevailed in the month of March to April. During these months oogonia gradually showed a decreasing trend to EPN and then LPN in gonads. The highest EPN stage was found in March (7.67±1.53%) with a decreasing trend in the following months. As a result, the percentage of LPN stage was highest in April (31.00±2.65 %) and then found to decrease gradually. Cortical alveoli stage (CA) was found in mature phases which basically prevailed in the month of May. In April some oocytes turned to CA but basically appeared in May and also highest percentage observed (10.67±2.08%). Vitellogenic stage (VG) was found in ripe phase, prevailed from May to June and chronologically increased and highest percentage of ripe oocytes were recorded in June (33.67±3.51%). Occurrences of advanced stage of oocytes indicate that mature ovary is almost ready to spawn. Biswas (1993) opined that the progressive change in the intra-ovarian diameter for a period not less than a year can give an idea of the spawning periodicity of the fish studies. Underdeveloped oocytes, early and late perinucleolar oocytes, yolk vesicle and early yolk granule oocytes were found during March in the ovarian tubule of *M. cuchia* (Alam *et al.*, 2012).

During the study period spent and resting phase were not seen. Different developmental stages of oocytes are shown in Plate 3. The developmental stages of oocytes seemed to be fairly similar to that of *Labeo rohita* and *Cirrhinus cirrhossus (*Jahan, 2008), *Puntius sophore* (Kohinoor 2000), *Notopterus chitala* (Kohinoor *et al.*, 2012).

Table 1: Mean Body Weight (±SD), Ovary Weight, Absolute and Relative Fecundity of *M. cuchia*

Mean Body Weight (g)	Mean Ovary Weight (g)	Mean Absolute Fecundity	Mean Relative Fecundity
225.8±28.03	4.82±0.59	645.13±92.12	2.52±0.29

Plate 3: Developmental Stage of Oocytes in Different Months (H & N × 220)



CONCLUSION

The variations in the gonad weight and GSI of the female cuchia fish reached to the peak during June indicating maturity of ovary and definite spawning season. While Vitellogenic stage (VG) of oocyte as well as highest ova diameter was observed in June also. The results of the present study may contribute to have successful breeding program and seed production technology of this fish species in captivity. Mass seed production and conservation of the available populations through proper management of the populations is recommended to save this threatened species from extinction.

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