



Seasonal changes in ovarian maturation of an Asian Cat fish, *Mystus vittatus* (Bloch, 1797) in captivity

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ABSTRACT

Mystus vittatus, an indigenous fish species of West-Bengal are economically important due to its delicious taste. The ovarian histo-morphology and its annual changes and the spawning periodicity of this catfish were studied collected monthly in one year period of November, 2012 to October, 2013. The fecundity, gonado-somatic index, different stages of ovarian development (nucleus changes, oocyte diameter and formation of yolk vesicles, yolk granules and lipid droplets through histological observation) of *M. vittatus* were also observed in this article. By gross morphology and its seasonal changes of ovary especially on the changes in cell structure of this species, five maturity stages were distinguished to categorize the developing ovary: immature, mature, early ripening, ripe and spent. Our investigation has revealed that gonado-somatic index (GSI) in *M. vittatus* began to increase in March and peaked in July then declined sharply in September-October. This cat fish attained sexual maturity at 10-11 cm in length, 10-12 g in weight which was confirmed at the peaks of gonado-somatic index (GSI) and fecundity. The fecundity of *Mystus vittatus* has shown highly significant correlation with body length, body weight and ovary weight.

Keywords- *Mystus vittatus*, gonado-somatic index, fecundity.

INTRODUCTION

The Asian striped dwarf cat fish, *Mystus vittatus* (Bloch, 1797), locally known as "Bujuri Tangra", is a minor bagrid occurring in the eastern and north eastern regions of India having good market demand due to its high nutritive value as well as

great consumer preference. The species support a strong capture fishery in freshwater as well as in estuarine waters in India. However, during last few decades its wild population showed a steady declining phase mainly due to unavailability of proper breeding ground and favorable conditions (Chakraborty *et al.*, 2007). Besides that, its resources are also under strain due to over exploitation and habitat degradation by anthropogenic interferences. The fish also have good marketing value as ornamental fish and already gained a significant status in the international ornamental fish market. The fish were also randomly caught by fishermen for exporting as ornamental fish outside the country. This was another reason for diminishing availability of this priced and economic fish. All this aspect requires a need of production of this fish seed and to know its biological properties thoroughly. Knowledge of biological properties of any species is of paramount

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importance for management of its population and to assess the availability for their culture purpose. The available information on the biology of *Mystus vittatus* is scarce. Only a few researchers have studied on breeding biology of this species (Rao and Sharma, 1984; Hoque and Hossain, 1993; Rao et al., 1999; Mukherjee et al., 2002; Chakraborty et al., 2007; Islam et al., 2011).

Keeping all this in view, the present study was designed to observe maturation stages of ovary of *Mystus vittatus* macroscopically as well as microscopically which is very essential for its breeding biology. Studies on fecundity and gonado-somatic index as reported here are important tools to understand the demography of fish population as well as to make efforts to increase the amount of yield (Gupta and Banerjee, 2013).

MATERIAL AND METHODS

Fingerlings of *Mystus vittatus* (about one month old) were collected from Ichamati river and its adjoining market (near Majhdia, West-Bengal) in the month of July – August, 2012, transported to Vidyasagar college, P.G. campus, Kolkata and were maintained in cemented stocking tank (size- 104 cm X 56 cm X 50 cm). The fishes were fed daily with proper supplementary feed consisting of mustard oil cake, rice bran , fish meal (1:1:1 ratio) and live *Tubifex* @ 5 % of body weight per day in two splitting parts in order to ensure proper growth and development . For the study of fecundity, GSI and stages of maturation of ovary, 10 female fishes were sampled twice a month from November, 2012 to October, 2013 to observe the changes of maturation and reproductive status. The ovaries of each specimen were dissected out very carefully from the female body after removal of excess blood, fat bodies and were weighed. Then the ovaries were pieced into smaller parts and fixed in 10 % buffered formalin solution for 24 h. The materials were then processed under histological techniques following Roberts (2004). The weight, color and physical state of the ovaries were also noted visually in every month. Gravimetric method (Lagler, 1949) was used to determine the fecundity of fishes. It was estimated on the basis of number of eggs in the ovary. The formula used in the method was as follows:
Absolute Fecundity =

$$\frac{(n_1 + n_2 + n_3) \times \text{Ovary weight}}{W_1 + W_2 + W_3}$$

Gonado-somatic index (GSI) was calculated according to method cited by Parameswarn et al. (1974).

Gonado-Somatic Index (GSI) =

$$\frac{\text{Gonad weight} \times 100}{\text{Body weight}}$$

The stages of gonadal maturation and the seasonal changes in the proportion of the oocyte development within the ovaries were noticed and maturity stages were studied by microscope and macroscopic observations.

RESULTS

Fecundity of ovary:

The fecundity showed maximum in the months of June – July and minimum in September – October respectively. It was counted nil in November-December (Table-1) as it was not formed. The maximum average absolute fecundity was 7782.35 with an average length of 10.96±0.38 cm and weight 13.85±0.42 g and minimum average absolute fecundity 596.75 was observed in fish having an average length of 8.64±0.21 cm and weight 10.93±0.32 g (Table-1). Figure-1 illustrated the relative fecundity of fishes in different months. From January it increased gradually to the maximum in July then it slowly declined for the remaining months.

Table-1. Seasonal fluctuations of fecundity of *Mystus vittatus*

Month	Mean Length (cm)	Mean Weight (g)	Mean Ovary weight (g)	Absolute fecundity
November	6.5	5.06	0.08	0.00
December	6.65	6.19	0.12	0.00
January	8.07	8.36	0.34	2705.09
February	8.22	8.40	0.43	4900.25
March	8.87	8.90	0.78	6040.55
April	9.95	10.36	1.33	6400.65
May	10.87	11.45	1.74	6599.76
June	10.92	11.55	2.06	6984.50
July	10.96	13.85	3.01	7782.35
August	10.94	11.75	1.6	4196.89
September	10.93	9.72	0.41	2387.02
October	10.93	8.64	0.18	596.75

Figures-2-4 showed the regression analysis between body length, body weight and ovary weight with absolute fecundity. All the graphs showed linear relationship. Correlation value between length and absolute fecundity (r = 0.58), body weight and absolute fecundity (r = 0.83), ovary weight and absolute fecundity (r =0.84) showed positive correlation.

Gonado-somatic Index (GSI):

The Gonado-somatic index is an indicator of the state of gonadal development, maturity and spawning period in teleost fishes (De Vlaming, 1972).

Figure.1 showed monthly variations of relative fecundity.

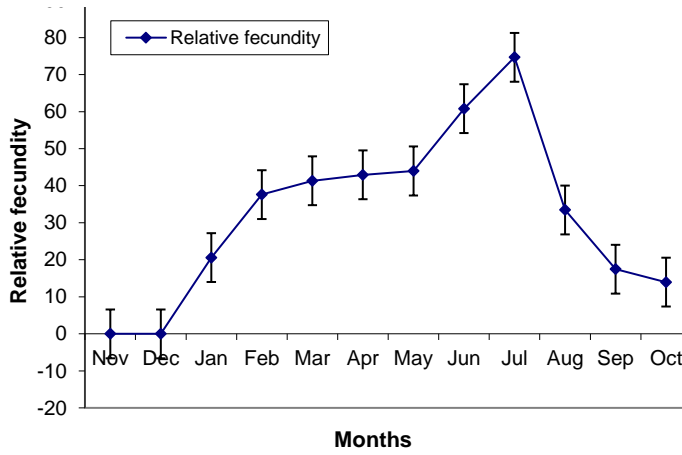


Figure.2 shows the regression between length & absolute fecundity. $r = 0.58$

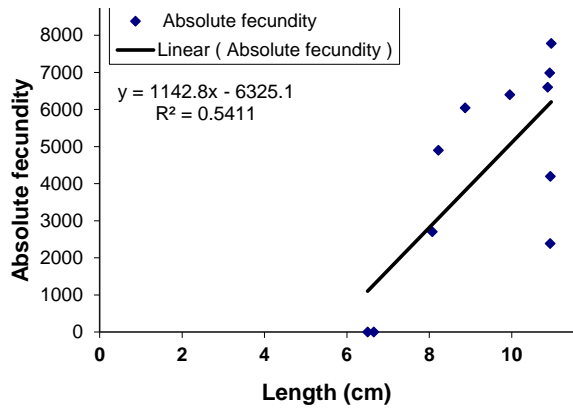


Figure.3 shows the regression between body weight & absolute fecundity.

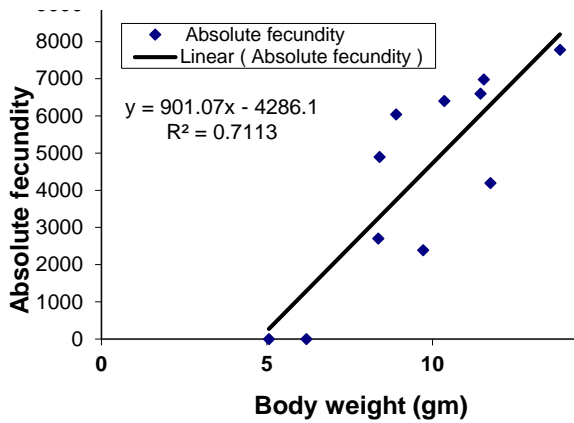


Figure-4 shows the regression between ovary weight & absolute fecundity. $r = 0.84$

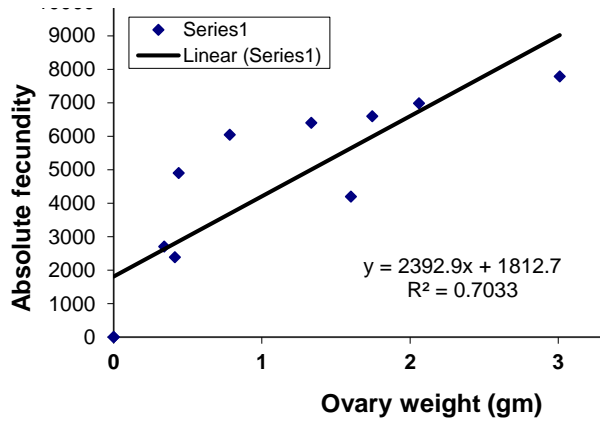


Figure-5 showed the monthly fluctuation of gonado-somatic index (GSI) in female *Mystus vittatus*.

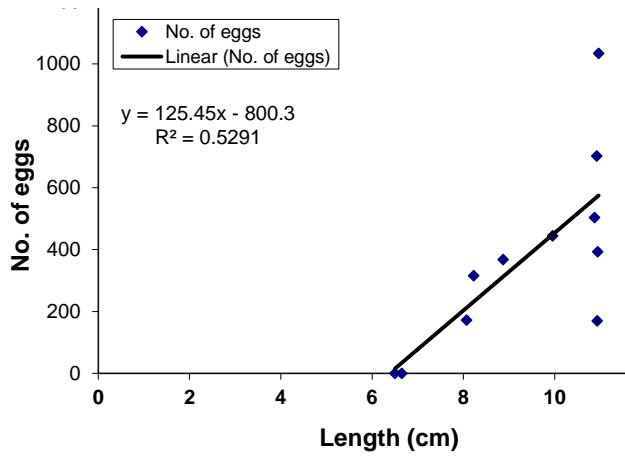


Figure-6 shows the regression between length and GSI. $r = 0.67$

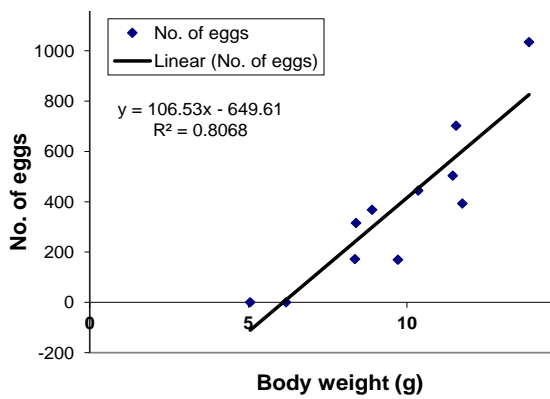


Figure-7 shows the regression between bodyweight and GSI. $r = 0.88$

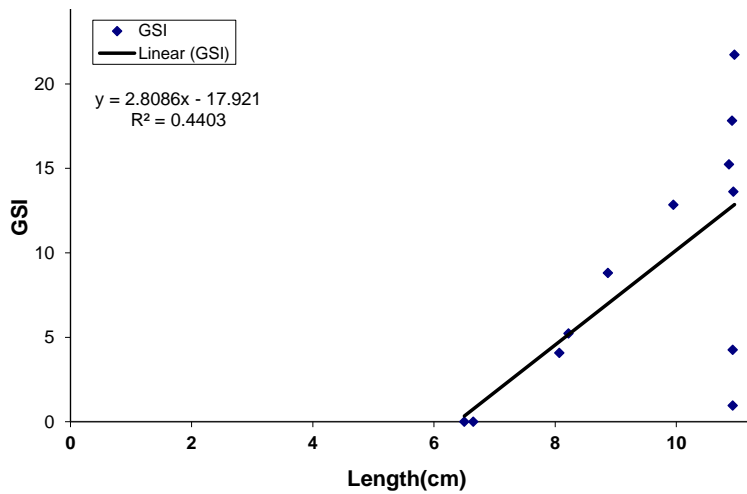


Figure-8. Shows the regression between ovary weight and GSI. $r = 0.98$.

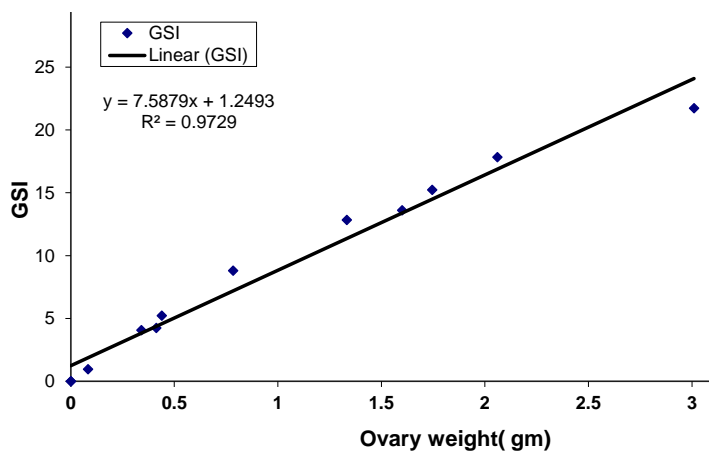


Figure-5 showed that GSI increased steadily from March, reaching peak in July, then declined for the rest of the months. The female fish showed changes in weight of gonad corresponding to three spawning period (Pre-spawning, spawning and post-spawning).

In the pre-spawning period (January-April), there was a gradual increase in the gonado-somatic index reaching a peak during spawning period (May-July) and then it was a declining phase during post spawning period (August-October) (Figure-5). GSI value recorded nil in November and December as development of gonad took place after these months. It is familiar, that GSI increased with the maturation of fish being maximum during the period of peak

maturity (Parameswarn *et al.* 1974). Arockiaraj *et al.* (2004) studied in *Mystus monotonus* and suggested that GSI values were relatively more stable over the year for the juveniles than for the adult fishes.

The data of GSI recorded in each month and has been represented graphically which showed linear relationship with length, body weight and ovary weight in Figures-6-8 respectively. All the regression analysis of GSI showed linear relationship with length ($y=2.81138 x-17.507$, $R^2=0.4466$), body weight ($y= 1.9928 x- 7.7977$, $R^2=0.7804$) and ovary weight ($y= 7.4729x+1.8261$, $R^2=0.9535$). In above three cases, it has been observed that ovary weight ($r=0.98$) and body weight ($r=0.88$) were highly correlated with GSI in comparisons to body length ($r=0.67$). Strongest

correlation ($r=0.98$) was found between ovary weight and GSI. It was revealed (Figures-6-8) that with the increase of fish length, body weight and ovary weight, GSI of *M. vittatus* was also increased steadily.

Maturation of ovary:

Maturation stages were observed every month to draw a clear picture of the spawning period. It was observed that the female attained maturity at 10-11 cm body length and body weight 10-12 g.

Based on visual observation under microscope (Olympus MIPS Camera microscope), five maturation stages could be distinguished in the ovary of female: Immature, mature, early ripening, ripe and spent. The onset of sexual maturity was first observed in January. During February-March, weight of the ovary increased and occupied half of the body cavity. At the end of June, it changed its color from cream to deep yellow and occupied $2/3^{rd}$ of the body cavity. Ovary in this stage was richly supplied with blood vessels. After releasing ova, it became reduced in size (October-November).

Histology of ovary showed the different phases of development of ovaries during oogenesis. The process of oogenesis of *M. vittatus* have been classified into five phases (I-V) according to ovary development following Arockiaraj *et al.*, 2004.

Immature stage (I):

Oogonia of this period were very small, spherical cells. Each cell contained thin, indistinct peripheral basophilic cytoplasm with a large nucleus. The primary oocyte was mostly polygonal or hexagonal shape and varied in diameter 25-88 μm (Figure-9).

Figure- 9. Immature stage with primary oocyte
Mature stage (II):



Oocytes grew rapidly, showing follicular epithelium and increased in size (88-193.6 μm in diameter). The nucleus increased in size and number

of nucleoli (2-15 in number) appeared at the periphery of nucleus and varied in diameter 4 to 9 μm . The zona radiata was not thick in the growth phase. Balbini bodies appeared in the cytoplasm (Figure-10 and Figure-15).

Figure- 10. Matured ova



Figure-11. Early ripe stage of Ovary

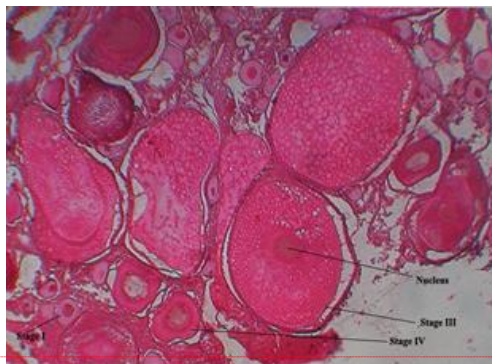
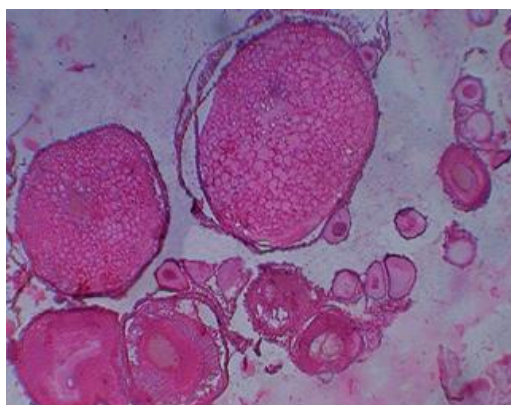


Figure-12. Ripe stage of ovary



Comment [u2]: Change 'Fig.10' to 'Figure-10 and Figure-15

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Early ripening stage (III):

Small vacuoles, assumed to be lipid made their appearance within the cytoplasm. The vesicles appeared randomly at various depths in the ooplasm. The nucleus increased in size. Inner layer of zona radiata became more conspicuous at this stage, coated with follicular epithelial layer. The diameter of the oocytes at this stage became 123.55-220.15 µm. This phase was also referred as vitelline phase (Figure-11 and Figure-15).

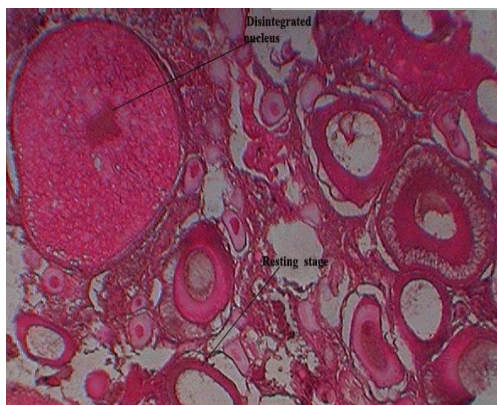
Ripe stage (IV):

Ovary was distended, released ova with slight pressure and was deep yellow colored. The oocytes increased to maximum size at this stage (338.30-476.35 µm in diameter). Yolk vesicles appeared and became randomly dispersed in the peripheral region of the cytoplasm. The yolk globules were small, spherical and located in between lipid vesicles. Soon they became larger in size and frequently fused with one another. The zona radiata reached to maximum thickness and differentiated into an outer and inner layer. The nucleus became irregular in size (Figure-12 and Figure-14).

Spent stage (V):

The phase is characterized chiefly by the migration of the nucleus towards the animal pole. The nucleus decreased in size and the nucleolar membrane disintegrated. The nucleoli were hardly distinguishable in the nucleus.

Figure-13. Spent stage of ovary



The cortical layer was found mixed with the yolk granules. After the germinal vesicles broke down, the oocytes ovulated in the ovarian lumen and the post ovulatory follicle remained in the ovary. The discharge of ripe ova during the spawning season was accompanied by a decrease in the number of the ripe ova. At the end of this period, spent stage

was noticed by the appearance of empty follicles (Figure-13).

Figure-14. Zona radiata and cortical vesicles in the ovary

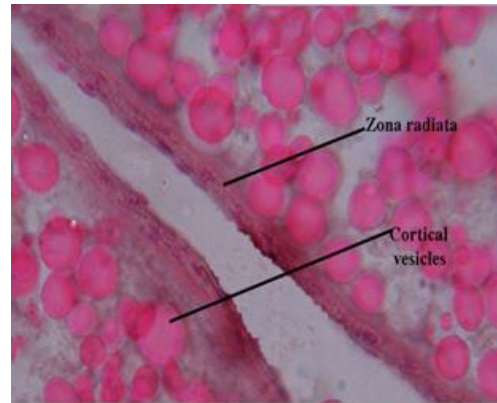
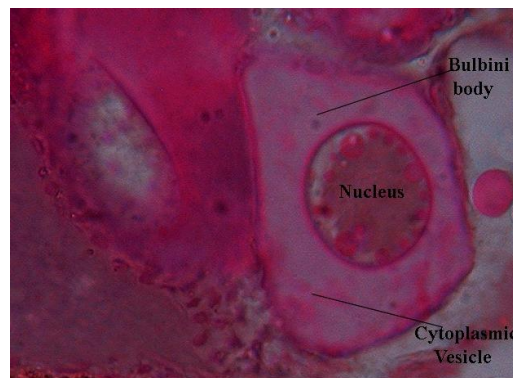


Figure- 15. Cytoplasmic vesicles, balbini bodies and nucleolei in the ovary



DISCUSSION

The female *Mystus vittatus* attained maturity at 10-11cm in length, 10-12g in weight. While *M. seenghala* a related species, matured at 7.0-7.8cm (Saigal and Motwarin, 1982). *M. monotonus*, however, matured at 13-14cm (14-16g) (Arockiaraj et al., 2004). This study showed that the ovarian maturation of *M. vittatus* peaked during April-July when the monsoon started. Interestingly, Arockiaraj et al. (2004) reported that in a similar species, *M. montanus*, it peaked during October-December at the onset of the North East monsoon. Such variations in spawning seasons (late June through August) were also observed in *M. tengara* (Rastogi and Saxena,

1968) when in *M. aor* in the North Indian rivers during April-August respectively (Khan, 1935).

The maximum fecundity of female *M. vittatus* (average length 10.87cm and weight 10.45 g) was estimated to be 7782.35 (Table-1). However, during a study on the breeding of *M. vittatus* with artificial hormones Chakrabarty *et al.* (2007) reported the female fecundity varied in between 1262 – 10734. Similar observation was made by Sarker *et al.* (2002) in *M. gulio*, the highest mean fecundity of female - 21589 (for 20-22cm in length) whereas the lowest was reported to be 11887 (for 10-12cm length). Fecundity varied from species to species depending on the environmental conditions, length, age, location, genetic potential etc. and even among the individuals of the same species (Alam and Pathak, 2010). In the present study, fecundity has been observed to increase with body weight and length of female. But the fecundity increased more with increase in body weight than increase in length. This may be due to the fact that compared to the body length the weight of the ovary increased more rapidly hence the body weight increased. This study also revealed that although the older fishes were more fecund but it were the younger fish that produced more ova per gram body weight (Figure-1) indicating that the younger fish had more capacity to produce ova. A similar trend was observed by Parameswaran *et al.* (1974) on Ophiocephalidae group of fishes.

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Gonado-somatic index (GSI) of the fish increased with body weight, length and weight of the ovary. The GSI was maximum when the fishes were fully matured as expected (Figure-5). The increase in GSI values of females indicated the gradual development of the ovary (Stage I-IV) during the months of January to July and it declined during November-December, indicated spent stage of ovary in *M. vittatus*, while in another observation in *M. monotonus* GSI peaked during November-December (Arockiaraj *et al.*, 2004; Londhe Sheetal D. and T. V. Sathe, 2015).

The growth activity of ovary was divided into five stages. These were immature, mature, early ripening, ripe and spent respectively which were confirmed by both visual and histological observations. In most teleosts growth of ovary followed a similar general pattern (Paritha bhanu and M. Deepak, 2015; Maddock and Burton, 1999; Knuckey and Sivakumaran, 2001; Arockiaraj *et al.*, 2004).

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Conflict of Interests:

The authors declare that there is no conflict of interests regarding the publication of this paper.

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