

## REPRODUCTIVE BIOLOGY OF BOMBAY DUCK, (*HARPADON NEHEREUS*) IN MON COASTAL WATER

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### Abstract

The reproductive biology of *Harpadon nehereus* from Mon coastal water was investigated during May (2019) to April (2020). *H. nehereus* have a prolonged spawning season with two peaks during May to June and December to January. GSI showed peak values in May (4.16), June (5.16), December (4.83) and January (5.69) in female. The Kn values ranged from 0.989 (August) to 1.004 (October) in male and 0.998 (July) to 1.027 (December) in female. The overall sex ratio (1:0.66) showed significant difference at 5% ( $p < 0.05$ ) level and males outnumbered females. The length at first maturity of females was estimated as an average of 230 mm. The fecundity varied between 23,625 and 117,498 with an average fecundity of 55054 ova. The fecundity indicated higher relationship with ovary weight ( $r = 0.934$ ) rather than total length ( $r = 0.724$ ) and body weight ( $r = 0.713$ ). The mature ovaries (from stage IV to VI) of *H. nehereus* contained mature ova which are distinctly separated from the immature stock.

**Keywords:** *Harpadon nehereus*, reproductive biology, spawning, fecundity.

### Introduction

*Harpadon nehereus* (Hamilton, 1822) popularly known as Bombay duck, forms one of the most important commercial fish along the Mon coastal areas. These species was mainly caught by bag-net locally in shallow water of estuaries within the depth of 10 to 40 meter. The fishery sector of Sepalar, Setse and Zeephyuthaung coastal areas are important to the local populace where artisanal fishery is developed including the production of dried fish, dried shrimp, fish paste and fish sauce as their business. Bombay duck is a very soft fish and due to its highly perishable body composition, a large amount of the catch particularly during the peak fishing season is sundried and a small portion is sold fresh in the markets.

Reproductive parameters such as size at first maturity, sex composition, spawning frequency and spawning season, fecundity and ova diameter studies can help in the fisheries forecast (Bal and Rao, 1984). The knowledge on length at first maturity and spawning season detects when and at which length the fish should be protected and therefore it is important for the proper management and conservation of fish stocks (Hunter *et al.* 1992).

Several biological information such as length-frequency distribution, length-weight relationship and food and feeding habit *H. nehereus* along the coast of Mon state have been studied by Tint Swe (2011) but there was no record, so far, on the reproductive biology of this species. Thus, the objective of this study is to provide the first information on reproductive biology of *H. nehereus* including maturity stages, spawning season, gonado-somatic index (GSI), relative condition factor (Kn), length at first maturity, sex ratio, fecundity and ova diameter measurement.

### Materials and Methods

#### Study areas

The present study on the reproductive biology of *H. nehereus* was conducted at three stations in Mon coastal area, namely Sepalar (Lat. 16° 14'N, Long. 97° 32'E) in Chaungzone

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Township, Setse (Lat. 15° 56' N, Long. 97° 35' E) in Thanbyuzayat township and Zeephyuthaung (Lat. 15° 19' N, Long. 97° 51' E) in Ye Township.

### Sample collection and study period

A total of 843 fish samples (508 males and 335 females) ranging in size from 180-305 (mm) total length and 19.55 - 206 (g) body weight were collected monthly from Sepalar, Setse and Zeephyuthaung fish landing sites during May (2019) to April (2020). The samples were preserved in ice box until it reaches to the laboratory.

### Maturity stages and spawning season

After recording the total length and body weight by using digital clipper (0.01 mm) and digital balance (0.01 g), ovaries were separated from fresh specimen. The maturity stages of female were identified based on macroscopic examination like appearance of ovary (shape, size, color, condition, extend of yolk formation) and microscopic structure such ova diameter measurement (Clark, 1934). Maturity stages were classified into seven stages such as immature (stage I), maturing (Stage II), developing (stage III), mature (stage IV), pre-spawning (stage V), ripe (stage VI) and spent (stage VII) followed by Mackie and Lewis, (2001). Samples of ovary were weighted to nearest 0.01g with digital balance and preserved in 5% formalin for further study. Spawning season was ascertained by the proportion of the maturity of stage IV, stage V and stage VI (mature, pre-spawning and ripe) females, and gonad-somatic index values during the different months of the study period.

### Gonado-somatic index (GSI)

Gonado-somatic index provides information about the spawning period and GSI was calculated for monthly by using the following formula, (Nikolsky, 1963).

$$\text{Gonado-somatic index (GSI)} = \frac{\text{Weight of gonad}}{\text{Weight of fish}} \times 100$$

### Relative condition factor (Kn)

The relative condition factor was calculated by using the formula:

$$Kn = W/W' \text{ (Le Cren, 1951)}$$

Where, Kn = Relative condition factor, W = Observed weight, and W' = Calculated weight.

### Sex ratio

To know the homogeneity of the distribution of males and females, Chi-square test (Snedecor and Cochran, 1976) was applied.

$$\text{Chi-square (x}^2\text{)} = \frac{\sum (O - E)^2}{E}$$

Where, 'O' is observed frequency and 'E' is expected frequency of males or females per month or length groups.

### Length at first maturity

Length at first maturity was estimated by plotting the percentage of cumulative frequency against the length groups. The length at which 50% of fish attained sexual maturity was considered as length at first maturity (Lm<sub>50</sub>). (Beverton and Holt, 1957).

## Fecundity

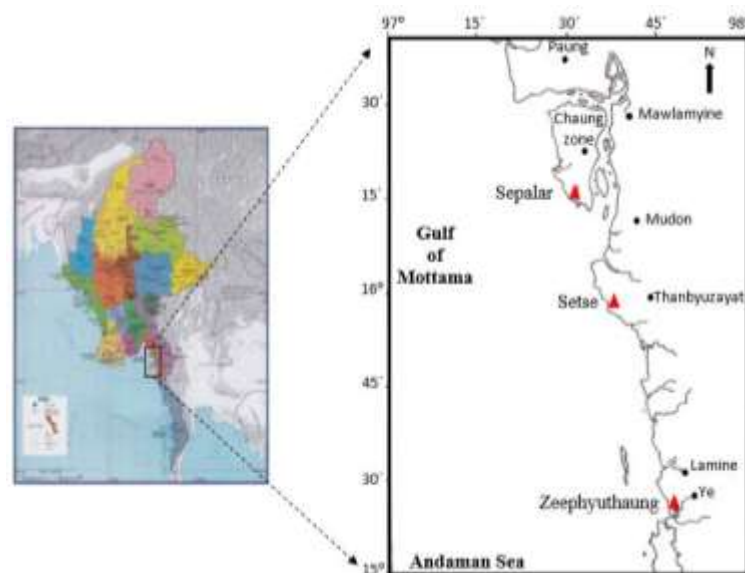
For estimation of fecundity, twenty ovaries in the V (mature) and VI (ripe) stages of maturity were selected. An amount of 0.05g of ovary from the anterior, middle and posterior regions of each ovaries was taken by using digital electronic balance. The sub-samples were placed on the counting slide with aid of few drop of water. The numbers of mature ova were counted and average number of ova of the three portions determined. Fecundity was calculated by using the following formula, Fecundity = Total weight of gonad/ sub-sample weight of gonad\* No. of ova in the sub-sample. The relationship between fecundity (F) and total length (TL); body weight (BW); ovary weight (OW) and their respective correlation coefficients (r) were expressed by least square method:

$$\text{Log } F = a + b \text{ Log } X$$

Where: F = fecundity, a = constant, X = variable (Total length; body weight; and ovary weight) and b = regression coefficient (The exponent).

## Ova diameter

Twelve ovaries were selected for the ova diameter study, where two each belong to stage I to VI. Random samples of ova from three portions of each ovary were taken and the diameters of nearly 200 ova were measured in straight line under the microscope using calibrated ocular micrometer. Ova diameter measurements were grouped into 80  $\mu\text{m}$  class interval size groups and expressed in percentage of the total number of ova for each ovary (Clark, 1934).



**Figure 1** Map showing the study sites of *H. nehereus* in Mon coastal water

## Results

### Maturity stages and spawning

Based on a total of 335 females ranging from 180 to 305 mm total length and 22.33 to 206 g body weight, the maturity stages and spawning season were examined. From the result of monthly percentage distribution of maturity stages, the maturity stages I–III ovaries were observed throughout the study period and the maximum percentage for stage I was recorded in April, stage II in October and stage III in February, respectively. Stage IV ovaries were also observed all the month except in April and the peaks values were observed in December and January. The percentage occurrence of stage V ovary showed the lowest value in September and the highest in

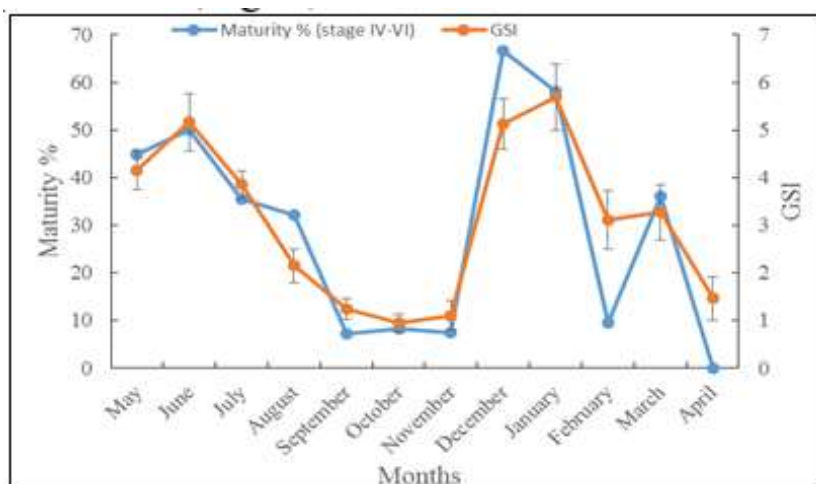
June followed by December and January. Stage VI ovaries occurred from May to August, November to January and March showing two peaks in June and December. Samples of stage VII ovary were recorded in June, August to November and February to March showing three similar peaks in August, September and February. The lowest percentage were observed in March and November (Table 1.). The proportion of maturity (%) for stage IV, stage V and stage VI indicated the peaks in May (44.82%), June (50%), December (66.66%) and January (58.06%) and the lowest values were observed in September (7.14%), October (8.33%), and November (7.41%). There was no record in April (Fig. 2).

**Table 1 Monthly percentage distribution of maturity stages in female *H.nehereus***

Months	No. of fish	Maturity stages (%)						
		I	II	III	IV	V	VI	VII
May	29	6.90	20.69	27.59	13.79	17.24	13.79	
June	36	5.56	13.89	19.44	11.11	22.22	16.67	11.11
July	48	4.17	22.92	37.50	14.58	16.67	4.17	
August	28	10.71	17.86	25.00	17.86		14.29	14.29
September	28	28.57	28.57	21.43	3.57	3.57		14.29
October	24	16.67	41.67	20.83	8.33			12.50
November	27	29.63	29.63	25.93	3.70		3.70	7.41
December	33	3.03	9.09	21.21	30.30	21.21	15.15	
January	31	3.23	9.68	29.03	29.03	19.35	9.68	
February	21	23.81	14.29	38.10	9.52			14.29
March	25	28	12	20	24	4	8	4
April	5	40	40	20				

### Gonado-somatic index (GSI)

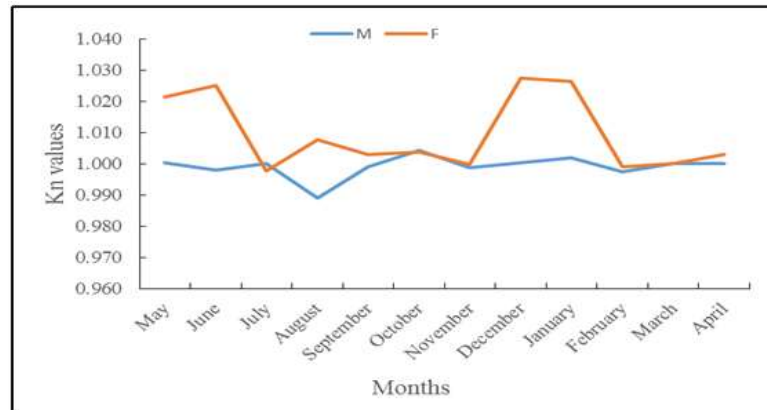
Monthly gonado-somatic index of female *H. nehereus* varied between 0.94 to 5.69 and the peak values were observed in May (4.16), June (5.16), December (4.83) and January (5.69). The values were found to be lower in September (1.24), October (0.94), November (1.1) and April (1.46) as expressed in (Fig. 2).



**Figure 2** The percentage occurrence of maturity (Stage IV-VI) and GSI of female *H. nehereus* in different months

### Relative condition factor (Kn)

The Kn values in male showed slightly fluctuation from the minimum 0.989 in August to the maximum 1.004 in October. In female, the peak values were recorded in May (1.021), June (1.025), December (1.027) and January (1.026). The low Kn values were observed in July (0.998) and February (0.999). Average Kn values for male and female were 0.999 and 1.01.



**Figure 3** Monthly variation of relative condition factor in male and female

### Sex ratio

Since *H. nehereus* have no external differentiating characters between the two sexes, the abdomen of the specimens were cut open for distinguish the sex. The sex ratio was tested by 1:1 method of Chi-square ( $\chi^2$ ). The overall sex ratio of population (1: 0.66) showed significant difference at 5 % level ( $n = 843$ ,  $p < 0.05$ ). Monthly sex ratio was significant during September to October and January to April, indicating the predominance of males on females.

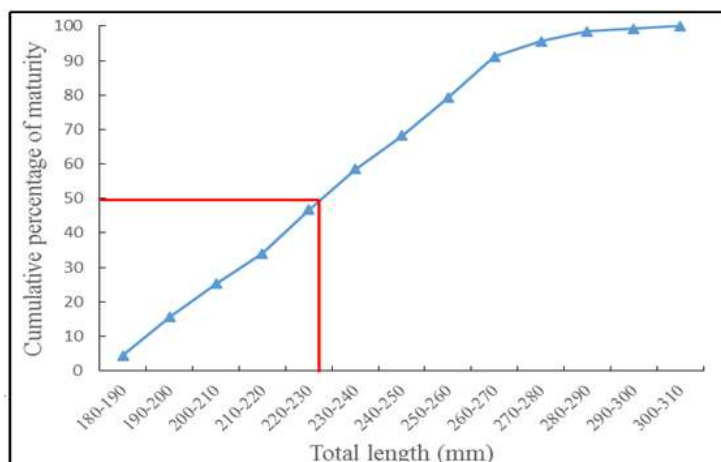
**Table 2** Monthly variation in sex ratio of *H. nehereus*

Months	No. of fish	Males	Females	Expected (E)	Sex ratio (M:F)	Chi-square
May	55	26	29	27.5	1 : 1.12	0.16
June	86	50	36	43	1 : 0.72	2.28
July	86	38	48	43	1 : 1.26	1.16
August	68	40	28	34	1 : 0.70	2.12
September	77	49	28	38.5	1 : 0.57	5.73*
October	72	48	24	36	1 : 0.50	8*
November	69	42	27	34.5	1 : 0.64	3.26
December	73	40	33	36.5	1 : 0.83	0.68
January	80	49	31	40	1 : 0.63	4.06*
February	79	58	21	39.5	1 : 0.36	17.32*
March	72	47	25	36	1 : 0.53	6.72*
April	26	21	5	13	1 : 0.24	9.84*
Total	843	508	335	421.5	1 : 0.66	35.5*

\* Significant at 5% level ( $P < 0.05$ ) with 1 df

### Length at first maturity

Length at first maturity of *H. nehereus* was determined based on 135 females ranged in size from 180 to 301 mm total length. Females in maturity stages IV and above were considered as matured fish and they were grouped into 10 mm length groups and analyzed into different maturity stages. The estimated length at which 50% of females *H. nehereus* reached sexual maturity was 230 mm.



**Figure 4** Length at first maturity of female *H. nehereus*

### Fecundity

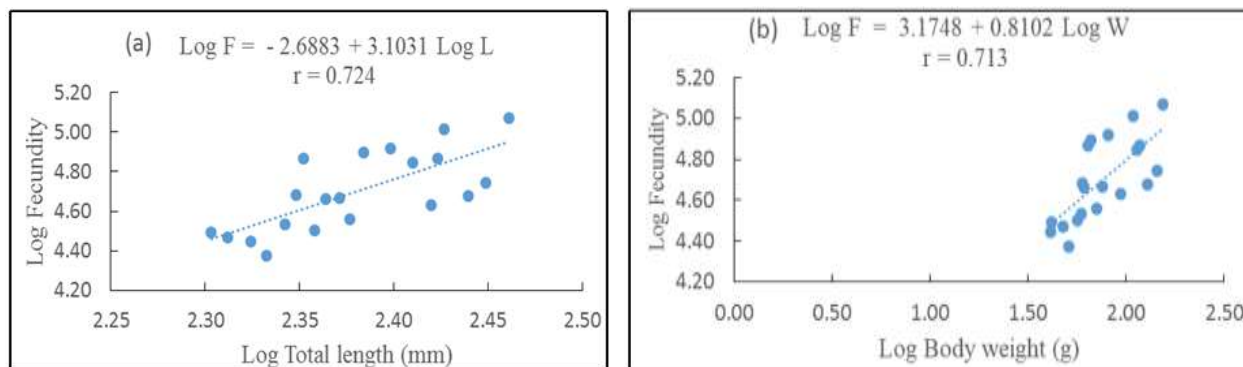
Fecundity was determined based on 20 specimens which ranged from 201 mm to 289 mm (TL) and 42.28 to 153.44 g (BW). The estimated fecundity varied between 23,625 and 117,498 ova with an average fecundity of 55054. The minimum fecundity was observed in fish with 215 mm (TL), 50.9 g (BW) and 3.36 g (OW). The maximum fecundity was recorded in fish with 289 mm (TL), 153.44 g (BW) and 16.78 g (OW). Regression equations of fecundity in relation to total length, body weight and ovary weight of fish were given as:

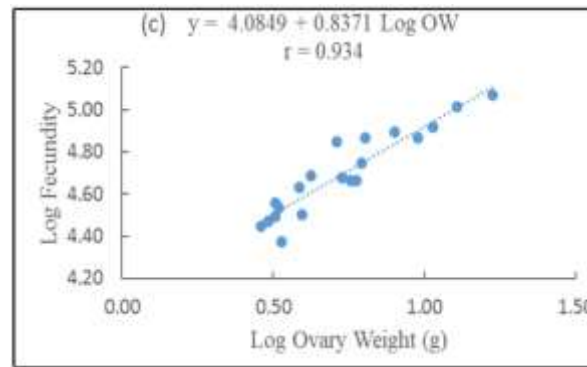
$$\text{Log } F = -2.6883 + 3.1031 \text{ Log TL}, r = 0.724$$

$$\text{Log } F = 3.1748 + 0.8102 \text{ Log BW}, r = 0.713$$

$$\text{Log } F = 4.0849 + 0.8371 \text{ Log OW}, r = 0.934$$

According to the correlation coefficient ( $r$ ) values from the above regression equations, there were highly positive relationship between fecundity against total length, body weight and ovary weight. This means that the fecundity increased according to total length, body weight and ovary weight.

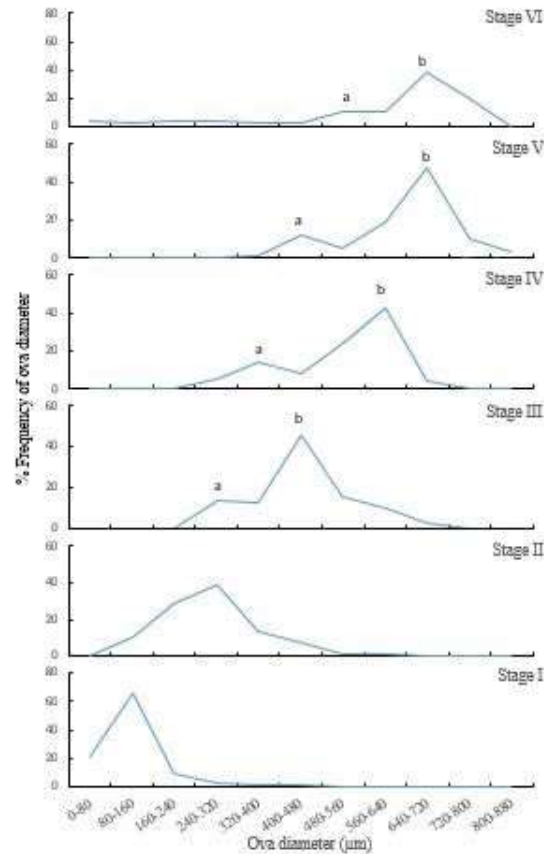




**Figure 5** Relationship between fecundity and (a) total length, (b) body weight and (c) ovary weight of *H. nehereus*

### Ova diameter

In stage I, most of the ova ranged from 57 to 300  $\mu\text{m}$  with a mode at 80-160  $\mu\text{m}$  (65.4%) and stage II showed ova diameter range from 160 to 440  $\mu\text{m}$  with a mode at 240-320  $\mu\text{m}$  (38.9%) size group for the immature stock. In stage III, most of the ova ranged from 240 to 600  $\mu\text{m}$  and two modes, one 'a' at 240-320  $\mu\text{m}$  (13.5%) and 'b' at 400-480  $\mu\text{m}$  (45.9%) were observed. In stage IV, most of the ova varied between 320 and 600  $\mu\text{m}$  and two modes were at 'a' 320-400  $\mu\text{m}$  (14.4%) and 'b' at 560-640  $\mu\text{m}$  (43.2%). All the ova belong to mature stock. Stage V showed two modes 'a' and 'b' at 400-480  $\mu\text{m}$  (11.9%) and 640-720  $\mu\text{m}$  (47.8%) size groups ranging the ova diameter from 400 to 680  $\mu\text{m}$ . In stage VI, two modes 'a' and 'b' were observed at 480-560  $\mu\text{m}$  (11%) and 640-720  $\mu\text{m}$  (37.8%) size groups. Ova in this stage is the largest varied between 560 to 720  $\mu\text{m}$ .



**Figure 6** Ova diameter frequencies for different maturity stages in *H. nehereus*

## Discussion

In the present study, all stages of maturity of *H. nehereus* were observed throughout the study period. The proportion of maturity (%) for stage IV, stage V and stage VI indicated the peaks in May, June, December and January and the low values were observed in September to November and April (Table. 1 and Fig. 2). It can be suggested that *H. nehereus* probably have a prolonged breeding season with two peaks during May-June and December- January. This result is in an agreement with the finding of Kham, *et al.*, (1992) who reported that peak spawning activity of *H. nehereus* was during December-January and June along the Saurashtra coast. Ghosh (2014) revealed that *H. nehereus* breed throughout out the year with peak in the summer period (April-July) along the Saurashtra coast. However, Johnson (2006) reported the peak spawning season was found to be in November and March in Mumbai water.

The study of gonado-somatic index (GSI) determined the reproductive conditions and spawning season of fishes. GSI of *H. nehereus* varied from 0.94 to 5.69 in female. The high GSI values were observed during May, June, December and January, and this found to be coincide with the peak spawning period. The low values were recorded in September to November when there was the predominance of immature (stage I) and maturing females (stage II) in the catch (Fig. 2). Kumar, *et al.* (2016) stated that GSI values of *H. nehereus* fluctuated between 2.158 and 8.173 and peak value was observed in February along Sunderband region. David (1963) observed the GSI peak which indicates active spawning.

The high Kn values were recorded in May to June and December to January in female and this observation confirmed that the spawning period of *H. nehereus* was the highest during May to June and December to January (Fig. 3). Nair *et al.* (1983) reported that 'Kn' cycle of the female closely follows the seasonal pattern of GSI, the periods of heavy breeding activity showing comparatively high 'Kn' value and vice-versa in *A. commersoni*. Monthly variation in sex ratio showed that there was significant difference at 5% ( $p < 0.05$ ) level in September, October and January to April between males and females. The overall sex ratio of population indicated the dominance of males on females in the catch (Table. 2). Similarly, Gosh *et al.* (2009) found the sex ratio of *H. nehereus* as 1:0.99 along Saurashtra coast. Bapat (1970) suggested that the presence of large number of male in July-August was due to the fact that the spent females move out of the fishing grounds after the main spawning season in November-March.

The length at first sexual maturity is an important parameter in fisheries research to determine the optimum mesh size and minimum legal size that may be needed to maintain the suitable spawning stock and to ensure at least one spawning for the mature individuals. (Amin *et al.*, 2016). In this study, length at first maturity of *H. nehereus* was estimated as an average of 230 mm (Fig. 4). This result is consistent with the finding of Kurian and Kurup (1992) and Kurian (2000). The result of the present study on the length at first maturity of *H. nehereus* was smaller than that of Johnson (2006) estimated the length at first maturity of *H. nehereus* as 255 mm. The variations in the size at first maturity from different coast and from same area can be attributed to the temporal and spatial changes in ecological conditions as well as possible error in sampling (Jaiswar and Chakaborty, 2016).

Fecundity is the total number of eggs produced per fish. The fecundity of *H. nehereus* ranged from the minimum (23625) in 215 mm to the maximum (117498) in 289 mm total length. The result of the present study is in an agreement with Kumar, *et al.* (2016) reported that the minimum fecundity (18156) was recorded in the length group of 210–220 mm and that of maximum (92012) in length of 280–290 mm from Sunderban. The estimated fecundity in the present study was higher than that of Khan, *et al.* (1992) found the fecundity of *H. nehereus* varied between 17075 and 79 631 along the Saurashtra coast. According to the result of Johnson, (2006), the fecundity of *H. nehereus* ranged from 21,182 to 116,067 in 246–356 mm in Mumbai water.



Ghosh (2014) estimated that the number of ova varied from 8467 to 102,079. Bromage *et al.*, (1992) reported that fecundity varied with the seasons, climatic conditions and environmental factors, nutritional status and genetic potential.

The correlation coefficient values ( $r$ ) from the linear regression equations showed that the fecundity indicated higher relationship with ovary weight rather than that with total length and with total weight (Fig. 5). Thus, the present study on fecundity can be suggested that the weight of ovary is more suitable indices for estimating the fecundity than length and weight of fish. However, (Ghosh, 2014) stated that fecundity was more closely related with length of fish although the relationship between length, weight and gonads weight of fish was observed to be linear.

As a result of ova diameter measurement, two modes of ova representing the immature and mature were observed in maturity stage III to VI ovaries of *H. nehereus*. The mature ovaries (from stage IV to VI) contain mature ova which are distinctly separated from the immature stock. This observations probably indicated that *H. nehereus* spawns once a year with a prolonged spawning season. Similarly Bapat (1970) and Johnson *et al.* (2006) reported that the ovaries of *H. nehereus* have ova with two modes representing the immature and mature crop where individuals spawn once a year and the species breed throughout the year. This observation was not in an agreement with Ghosh (2014), suggested that the presence of one batch of mature ova, one batch of maturing ova and one batch of immature ova in a mature ovary showed the individual fish spawns continuously in a year. Maturation and spawning period varies in different fish species and even the same species from different waters. It depends upon several ecological and physiological factors mainly temperature and intensity as well as duration of light that control maturation of gonads (Khanna and Singh, 2003).

## Conclusion

In conclusion, *Harpadon nehereus* have a prolonged spawning season with two peaks during May to June and December to January and males were dominant in the catch throughout the year. The fecundity indicated higher relationship with ovary weight than total length and body weight, therefore the weight of ovary is more suitable indices for estimating the fecundity. The length at first maturity of female *H. nehereus* was estimated as 230 mm. The ova diameter measurement revealed that the fish spawns once a year.

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