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Reproduction characteristics and length- weight relationships of the sand whiting (*Sillago sihama)* in the south coastal of Iran (Persian Gulf and Oman Sea)

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ABSTRACT

Sillago sihama is one of the most common recreational and commercial species in the local fishery in the Persian Gulf and Oman Sea. Various aspects of the reproductive biology of the S. sihama are studied to describe gonad development, spawning season, sex ratio, ovarian histology and fecundity. A total of 676 fish with a total length of 10.44-25.4 cm (Males), 11.17-24.8 cm (Females) and the total body weight ranged between 11.86-124.53 g (males) and 13.96-114.05 g (females) were used for this study. Sex ratio defined as the proportion of females to males was 1.2:1; microscopic and macroscopic gonad analysis and monthly variation of GSI in female component of the S. sihama shows a reproductive season from April and May and continued until June. The ova diameter frequency distributions in mature component indicated that the species exhibits a synchronous-group and monocyclic ovary characterized by deposition in a single batch of eggs per year (total spawner). The size at which 50% of the population attain sexual maturity (Lm50) is 138 mm for females and 132 mm for males. Maximum absolute fecundity is estimated to lie between 21345-73781 eggs for specimens between 13-24 cm in total length and 14.01-110 g body weight. Relative fecundity was 384.29-832.65 eggs per g body weight. There was a linear relationship between absolute fecundity and fish size for all Species tested.

Key words: *Sillago sihama*, Reproductive cycle, Fecundity, Sex ratio, GSI,

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INTRODUCTION

Sillago sihama, ladyfish and sand whiting, belonging to the family Sillaginidae is a valuable brackish water species for domestication and aquaculture. The *S. sihama* has a wide global distribution of the Sillaginidae, although considered limited to the Indian water and Pacific Oceans. *S. sihama* is an extremely valued recreational and one of the leading species for aquaculture production due to its rapid growth rate and high level of flesh quality.

Despite its recreational values and commercially prized, there were limited studies done in the aspects of the reproductive cycle of the *S. sihama* to provide valuable knowledge concerning stock assessment and management in the Persian Gulf except two by [13] and [4] which have studied the food habits and some aspects of reproductive biology in Persian Gulf. Related studies from Karwar on the Indian western coastline carried out by [16] . More recently, some attempt to gather data on spawning season of *S. sihama* carried out by [19] in Zuari estuary- India. In addition, maturity stages development of this specimen from Gulf of Mannar have been revealed by [7].

In regards to its importance in the fishery and the absence of any published informative data on the fishery of this species from the Persian Gulf and Oman Sea, the current study based on the reproductive outcome and spawning patterns of the *S. sihama*. In particular, examine the sex ratios and length at first maturity, maturity stages and gonadosomatic indices (GSI) for fish from the entire Persian Gulf and Oman seacoast.

MATERIALS AND METHODS

2.1. Study site and sampling:

Random monthly samples were obtained from commercial gillnet vessels and fishery-independent collections using hook and line from September 2009 to August 2010 in two locations: Bandar-e- Jask (25º 36′ N and 57º 48′ E) and Bandar-e-Abbas (27º 11′ N and 56º 16′ E) in the north coast of the Persian Gulf and Oman sea (Figure 1).

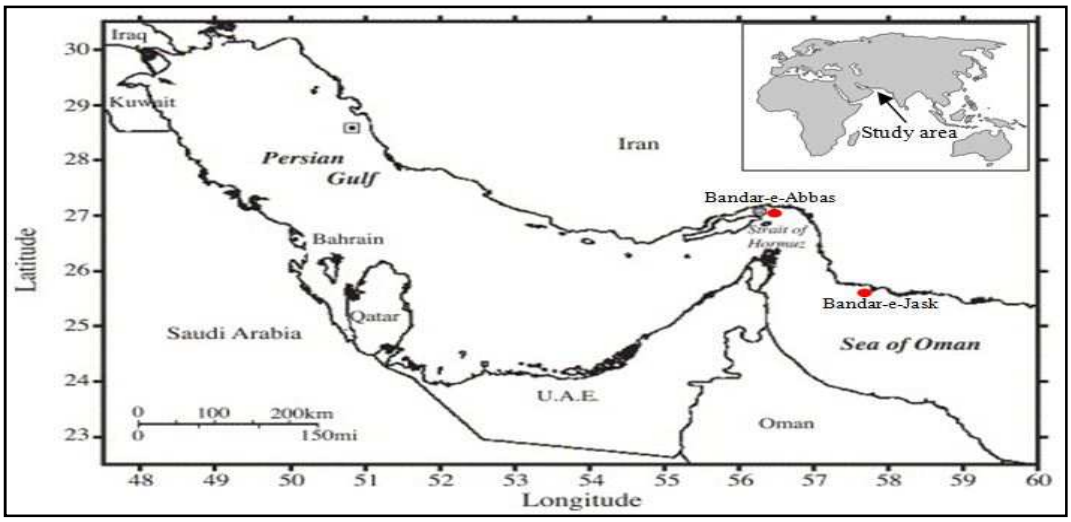

Figure 1: Location of the sampling sites (●) in the Persian Gulf and Oman Sea.

2.2.Biological Data Collection:

All specimens were measured to the nearest 0.1 mm of TL and weighed to the nearest 0.01 g.

Monthly sex ratio departure from the expected 1:1 ratio was testified. The total length-weight relationship was calculated by regression between these variables for each sex using [11] method:

$W = aL^b$, Where: W = Weight of fish (g), L = Length of fish (cm), a = y-intercept or the initial growth coefficient, b = Slope or the growth coefficient.

2.3. Morphological and histological Analysis:

Maturity stages were assessed according to criteria [3]. Macroscopic classification was established by gonadal inspection following the five-step method description (Table 1). Histological Analysis for female specimens was examined with fixed Gonad tissues in Bouin solution, then the samples were dehydrated in ethanol of rising concentrations (from 70 to 95%), exposed in xylene and embedded in paraffin blocks. The samples were sliced into 5-µm thickness using a rotary microtome then dyed using the hematoxyline-Eosin (Table 2).

Table 1: Visual (macroscopic) maturity stages and descriptions for *S. sihama* ovaries from Persian Gulf and Oman Sea

Stage	Described stage
I	Ovary and testis occupy nearly a third entire abdominal cavity. Tiny and close to the spinal column. Testis and ovaries inenarrable with the naked eye, thin, pinkish, ribbon like
II	Ovary and testis occupy most of the abdominal cavity. Ovary greyish, semi-transparent; eggs apparent with magnification device .Testis translucent and white-coloured.
III	Ovary and testis occupy nearly two-thirds entire abdominal cavity. Ovary pinkish- yellow together with granular appearance. Testis darker than before appearing pale reddish coloured.
IV	Ovary and testis occupy the entire length of the abdominal cavity. Ovaries expanded and including large transparent eggs. Testis milky in colour and fewer transparent with the convolution much less firm
V	Ovaries shrank getting loose walls. The ovary may possess very few ripe darkened or translucent eggs. Testis brighter, pale white and shrunken in size

Table 2: Sexual microscopic stages and descriptions for *S. sihama* ovaries from Persian Gulf and Oman Sea.

Stage	Description
I	Basically oogonia (og) and primary growth oocytes (po) that has significant pinkish nuclei are mixed together. Ovary walls are narrow.
II	Primary growth oocytes and oogonia can be found. Ovary walls are thin in new spawners. Cytoplasm about 50% occupied with yolk granules. primary growth oocytes and oogonia are present. Finally, oocytes are totally filled up with yolk, cortical alveoli are pushed towards the cell membrane (cm) and nucleus gets started migration in direction of the micropyle.
III	The symptoms of this stage can be pointed to final growth oocytes and hydrolyzed yolk granules (hyg), primary growth oocytes and oogonia. The nucleus at the end of this stage is not observable.
IV	Post ovulatory follicles (pof), remaining growth oocytes that has hydrolyzed yolk granules, primary growth oocytes (po) as well as oogonia existing in spawning stage. Post ovulatory follicles have taken place in the circumferential tissue
V	Primary growth oocytes and oogonia are existing. Spent phase includes several Post ovulatory follicles with growth oocytes. The ovary wall turn to thick from the shrinkage of the ovary after spawning

2.4. Length at first sexual maturity

Based on [9], the length at which 50% of fish individuals are at sexual maturity (L50) is known as the length of the first maturity. Frequency of mature specimens was plotted against different size group (10 mm) in male and females.

2.5. Gonadosomatic index:

Gonadosomatic index was calculated for individual fish thus:

$$\text{GSI} = \frac{\text{Gonad weight (g)}}{\text{Fish weight (g)}} \times 100 \quad [2, 17]$$

2.6. Fecundity:

To estimate the number of eggs in the ovaries, Gravimetric method was used for determination of absolute fecundity; therefore, ovaries were rinsed with water and placed in Gilson's fluid to dissolve the connective tissues. Three subsamples taken from the anterior, middle and posterior parts of the ovary. Samples were weighed and average number of eggs in each subsample were directly counted, the mean value was considered with equation given below [14]:

$$F = n \times \frac{G}{g}$$

Where: F= absolute fecundity, n= Average number of eggs in each subsample, g = subsample weight (g), G = Ovarian dry weight (g).

Relative fecundity was calculated by the following equation [5]:

$$R = \frac{F}{TW}$$

Where: R= Relative fecundity, F= absolute fecundity, TW= Total body weight (g)

2.7. Ova diameter:

Ova Diameter (ED) was deliberated to the nearest micron using a visual micrometer set within the eyepiece of a light microscope. Length at first maturity was determined by plotting the rates of matured fish specimens against their length whenever half of female specimens were mature [15].

2.8. Statistical analysis

Regression analysis was used to determine the relationships between Fecundity (F) with total length (TL) and total weight (TW). In order to determine the differences of the gonad weight and GSI value of different maturity stages of the male and female fish, one-way ANOVA and group t-tests for independent samples were used. The statistical analyses were carried out with SPSS software package and a significance level of 0.05 was applied.

RESULTS

During investigation period, 676 S. *sihama* individuals were examined. The total length of the specimens ranged from 10.44-25.4 cm (males), 11.17-24.8 cm (females) while the total body weight ranged between 11.86-124.53 g (males) and 13.96-114.05 g (females). Out of the total *S. sihama* specimens assessed throughout the study (N=676), 304 (44.9%) specimens were males, 372 (55.1%) were females (Figure 2). The overall sex ratio during the investigation period was in prefer of females (Female/Male=1.2:1); it significantly different from the hypothetical

distribution of 1:1 ($p<0.05$). In general, the number of males was significantly lower than females. Except for June and July, females were the highest frequency.

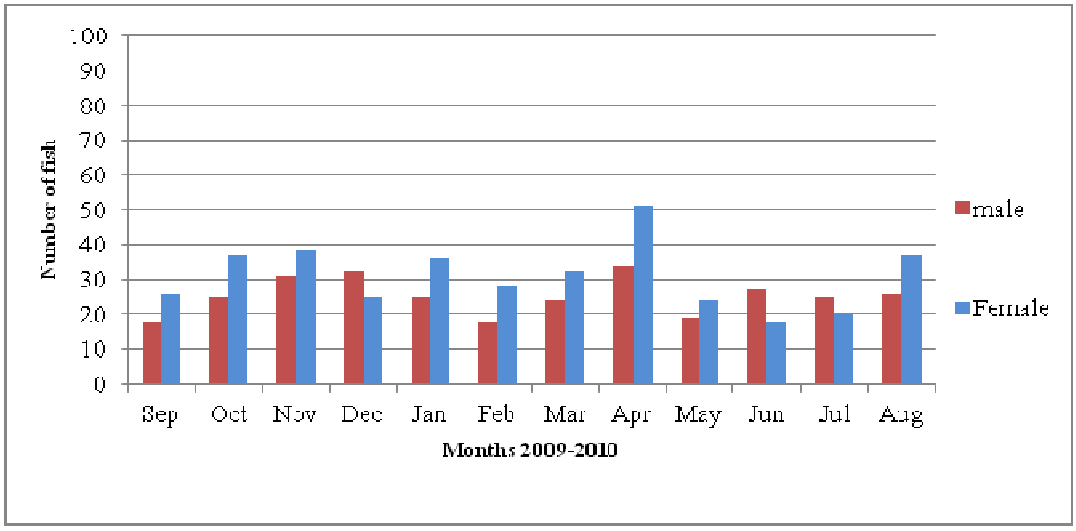

Figure 2: Monthly Variation of Sex Ratio for *S. sihama* (Female: Male) from Persian Gulf and Oman Sea.

Length- weight relationships were analysed for both sexes separately. Due to the significant difference between the male and female slopes of Weight-Length regressions, these equations are shown below:

Females: $W= 0085TL^{2.98}$ $R^2= 0.970$ $P< 0.05$ (Figure 3)
Males: $W = 0.0075TL^{3.03}$ $R^2 = 0.966$ $P< 0.05$ (Figure 4)

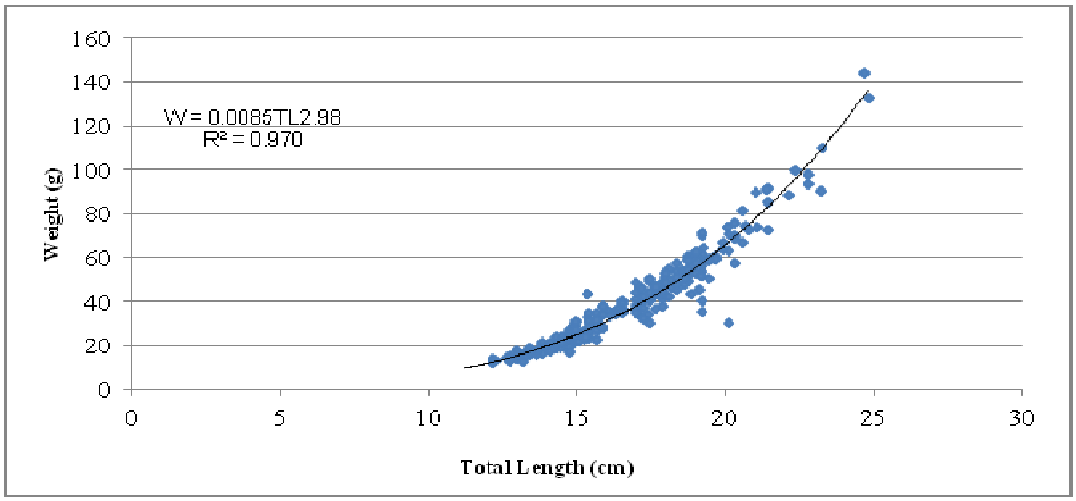

Figure 3: Length–weight relationships of *S. sihama* (Male) sampled from Persian Gulf and Oman Sea.

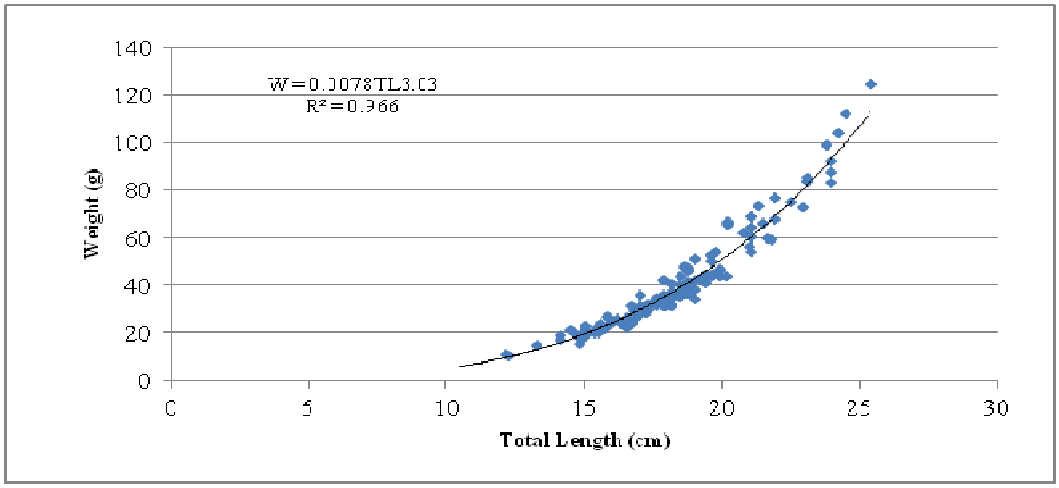

Figure 4: Length–weight relationships of *S. sihama* (Female) sampled from Persian Gulf and Oman Sea.

The maturity stages in the both sexes revealed that the morphological structure of the testis and ovary during immature and maturing condition (I, II) were during September to February (Figure 5 - 6). The results showed a significant increase during December to March for ripe ovaries (III). Spawning specimens began to appear in March and reaching to higher percent in late April and early July (IV). By July and August most of the individuals were completely spent (IV). While the highest percentage of the ripe testis (Stage IV) were observed in March and gradually decreased until July. Spent phase (V) of testis maturity formed during May and reach to the highest percentage in August.

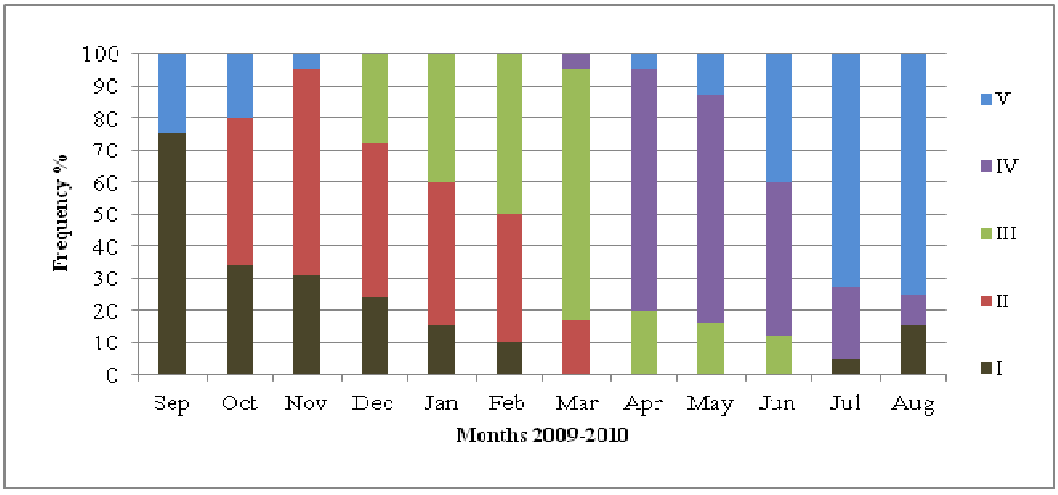

Figure 5: Monthly variations of ovarian maturity stages of *S. sihama* from Persian Gulf and Oman Sea.

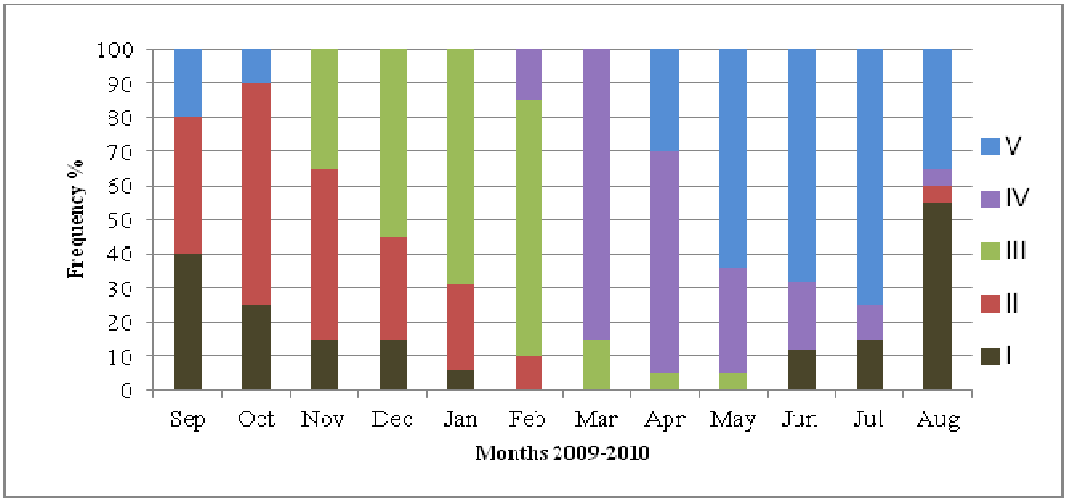

Figure 6: Monthly variations of testis maturity stages of *S. sihama* from Persian Gulf and Oman Sea.

The results of the histological study indicated that the ovary of *S. sihama* consists of an ovarian wall and numerous developing oocytes within follicles, which were embedded in a mass of connective tissue. The histological observation of female's specimens shown in Figure 7. Vesicle (cortical alveoli) and Yolk Granules or Vitellogenesis Stages were present during the study period except Jun and July while Maturation, hydration, and ovulation stages were found from March to August. These results were similar to the monthly macroscopic changes in maturity stages for *S. Sihama*.

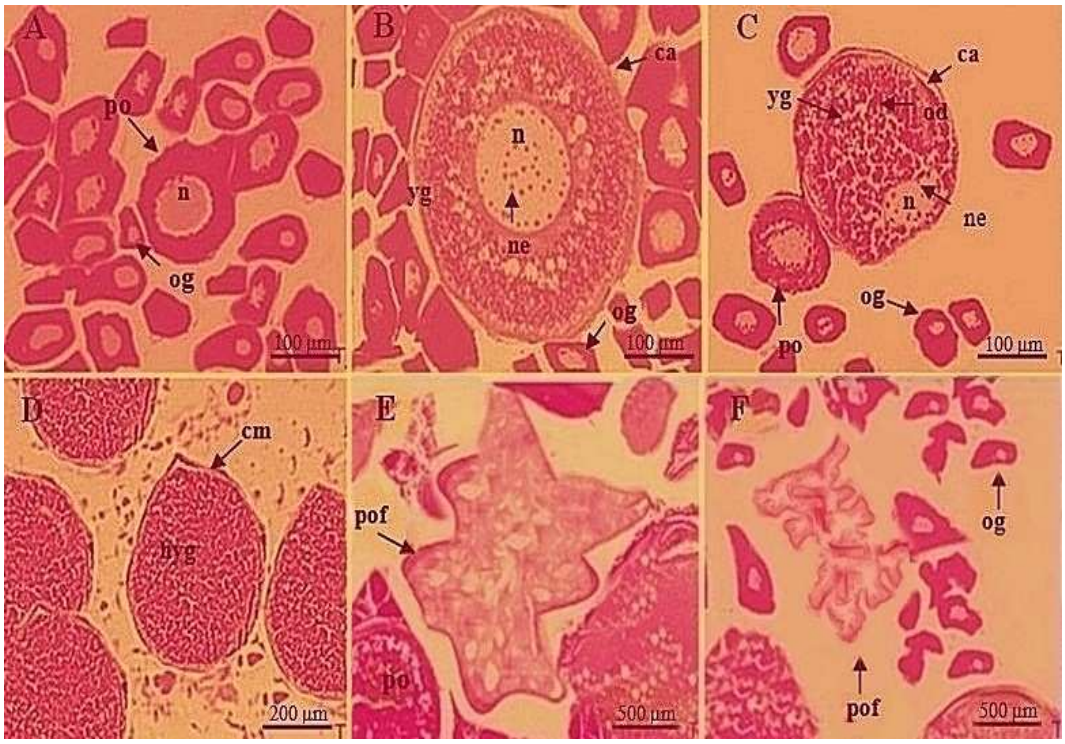

Figure 7: Histological section of ovary in S. sihama . (A) Immature (B) Maturing stage; (C) End of maturing stage; (D) Ripe stage; (E) Spawning stage; (F) Spent stage; Description: . n: nucleus; ne: Nucleolus; og: oogonia; po: primary growth oocytes; ca: cortica Alveoli; yg: Yolk globule; od: Oil droplet; cm: cell membrane; hyg: hydrolysed yolk granules; pof: post ovulatory follicles.

Immature ova diameter ranged from 0.02-0.1 mm, while the ova from the maturing phase ranged from 0.07-0.21 mm at the beginning and to 0.21-0.31 mm at the end. In ripe stage ova diameter varied between 0.31-0.43 mm, up to a whopping 0.53 mm in few instances. Fully mature ova in spawning phase varied in diameter from 0.41-0.55 mm,

only a few specimens were observed up to 0.65 mm (Figure 8). The frequencies of occurrence of ova at various diameters plotted against different months, confirmed which immature ova was presented throughout august - December and mature ova in the month of February-May. Since Jun, the development of mature ova reduced quantitatively (Figure 9). Furthermore, find out the spawning period depending on ova diameter.

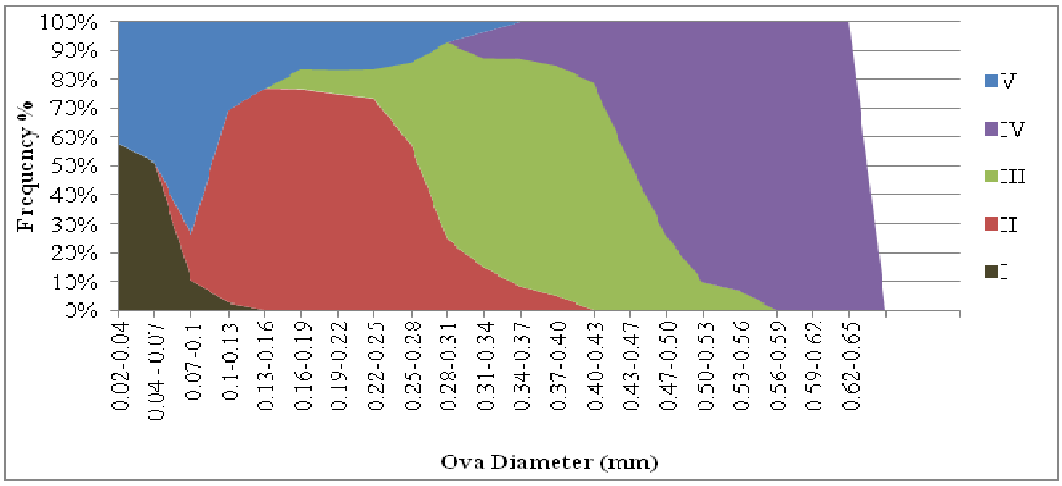

Figure 8: Size-frequency distribution of ova diameter for *S. sihama* from Persian Gulf and Oman Sea

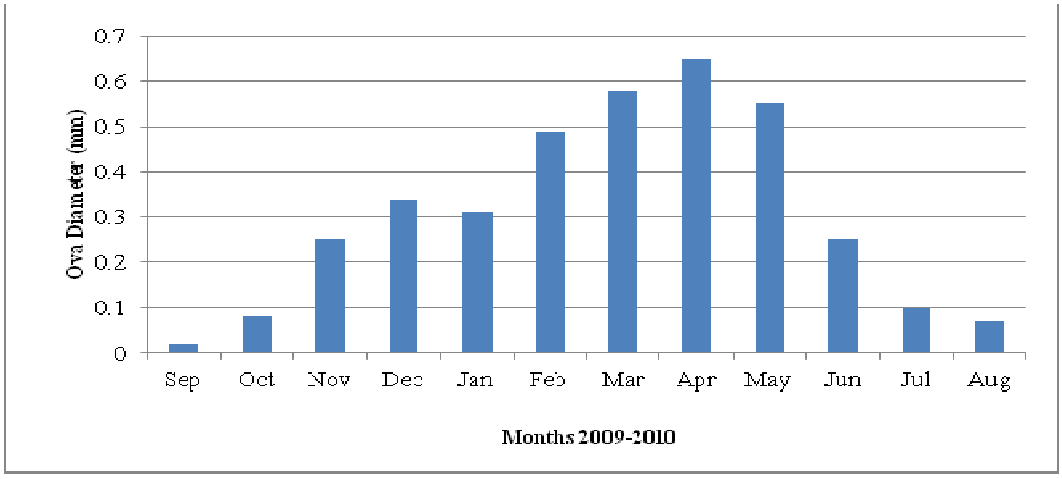

Figure 9: Monthly variation of ova diameter for *S. sihama* from Persian Gulf and Oman Sea.

Monthly gonadosomatic index variations shown both sexes followed nearly the same pattern. GSI presented the minimum values over the October and November period; lowest GSI was observed in October: 0.7 (Female) and 0.3 (Male).

Maximum values of GSI were found out during the March-May with highest GSI value in April (4.1) for females and it was 1.7 for males. Peaking was more noticeable for females (Figure 10). Based on the results of the current investigation, spawning occurred in April and May and continued until June, once the inactive phase of the maturity stages formed. There was no significant difference in GSI of male and female ($p>0.05$).

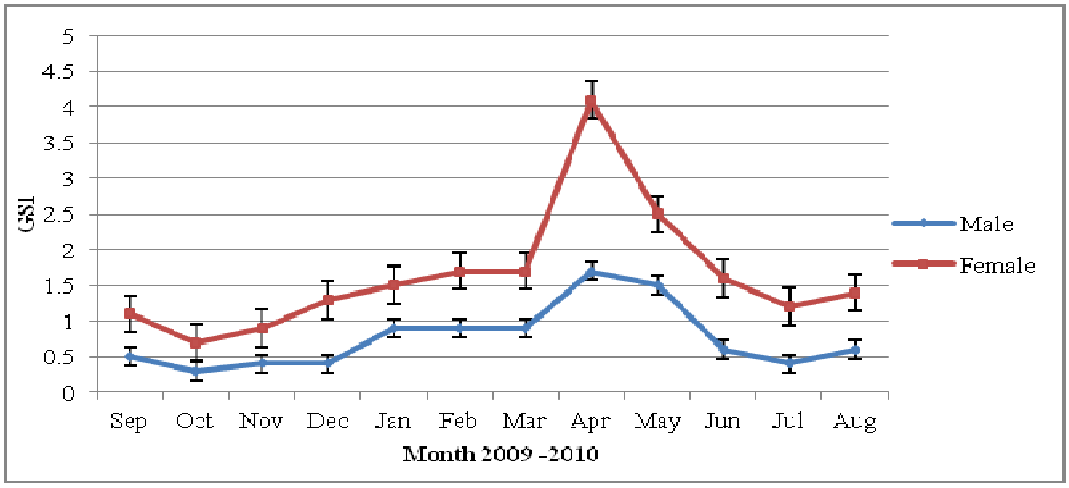

Figure 10: Monthly variations in gonadosomatic index (GSI) of *S. sihama* from Persian Gulf and Oman Sea.

It presented males and females become mature at size 132 mm and 138 mm total length respectively. Almost all males approximately over 165 mm total length were mature, while, for females, it was 176 mm. Generally, males reach sexual maturity at a smaller size than female specimens (Figure 11).

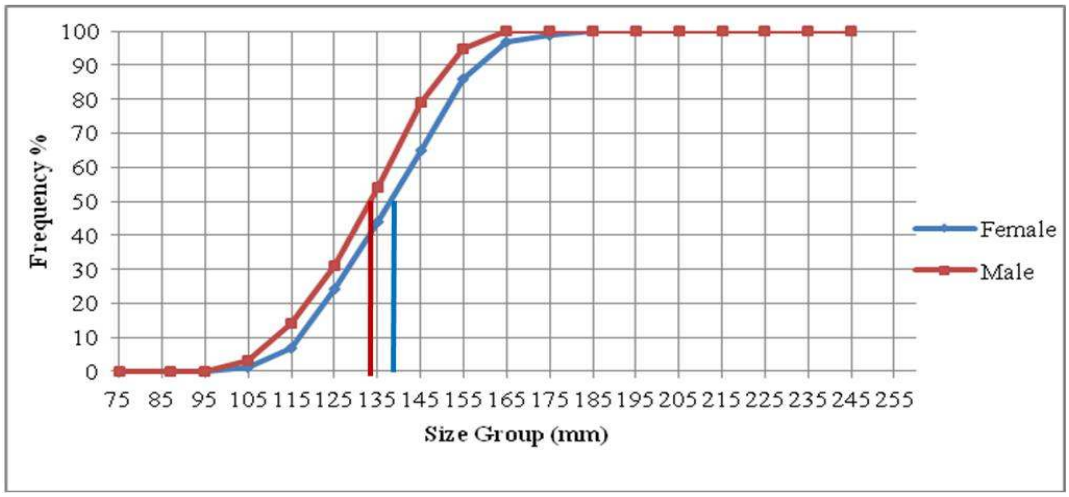

Figure 11: Length at first maturity (Lm50) of *S. sihama* from Persian Gulf and Oman Sea

The absolute fecundity was determined from 54 fish gathered from the spawning period. For absolute fecundity analysis, only ripe stages ovaries were considered. Absolute fecundity ranged from 21345-73781 ova per ovary of fishes of length and weight range 13-24 cm and 14.01-110 g respectively with mean value 45143 ± 3812. In addition, observed linear relationship between fecundity and total length and total weight.

Relative Fecundity (RF) was obtained as the number of eggs per unit weight (g) of fish. Lowest relative fecundity was 384.29 eggs in per gram related to species by mean total length 16.8 cm, mean total weight of 112.14 g while highest relative fecundity was 832.65 eggs/g related to species by mean total length of 21.3 cm and mean total weight 118.44 g.

Fecundity was positively correlated with length and body weight. The fecundity –Total length relationship is shown in Figure 12. The regression equation was F=5835TL-83510 (r = 0.517). The fecundity – body weight relationship is presented in Figure 13. The regression equation was F= 542.6W-10610 (r = 0.502).

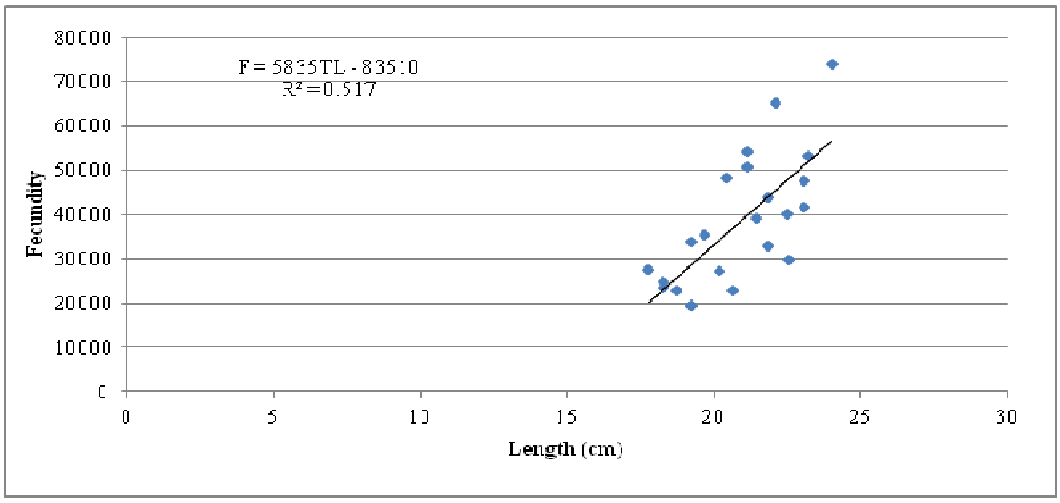

Figure 12: Fecundity - Total length relationship in *S. sihama* from Persian Gulf and Oman Sea.

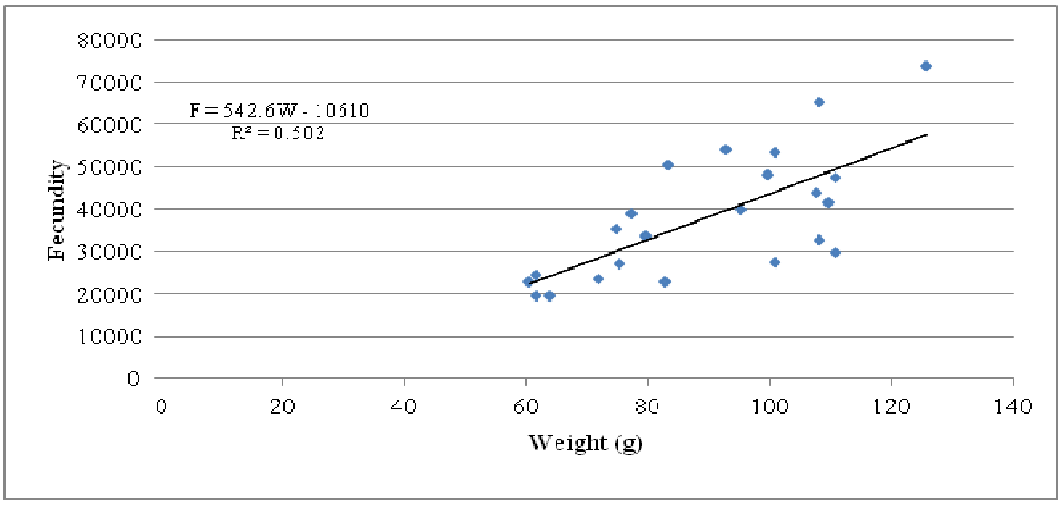

Figure 13: Fecundity - Total body weight relationship in *S. sihama* from Persian Gulf and Oman Sea.

DISCUSSION

In population structure, the length-weight relationship and b values attained for both sexes, Shows that the *S. sihama* species have isometric growth. To all specimens, the exponent b lies in between the expected range of 2.5-3.5 described in the majority of fish and shrimp of littoral and coastal [1, 11] and is nearby to 3 in both sexes showing which growth is isometric. Even though, the ratio of females to males of *S. sihama* revealed the prominence of females over males generally in most of the months throughout the year (September 2009 to August 2010) matching results have been described previously [16, 20].

In the present research, the highest GSI recorded for females, was greater (4.1) compared to males (1.7). Owing to an increasing number of ova within the ovaries of *S. sihama* in April and May, along with the temporal development of gonadosomatic index, present spawning time occurred during April to early June, including maturation, spawning and spent period. The findings about the reproductive cycle of *S. sihama* in this study are in line with observations of [4] who have examined *S. sihama* spawning period in the Oman Sea and [18] which it expanded around 3 months with the highest values of GSI from March to May. On the synchronization of spawning and ovary maturation, in the Southwest coast of India, indicated Spawning took place frequently, reach a high point between August and November. These differences may be related to changes in water temperature, salinity, and food supply, or perhaps to changes in maturity stage.

The findings of the histological experiments of ovary showed, the existence of oocytes at most of maturity stages, almost all oocytes of a matured group were generally spawned concurrently, but a few might be retained for

repetitive spawning. The results in this study are congruent with [8] and [10] arguments that monthly frequency distribution of oocytes in the *S. sihama* ovaries confirmed continuous spawning period.

In the current investigation, it may be ideally expressed that 130-140 mm size group is the size at which both the sexes reach sexual maturity. The size at first maturity in *S. sihama* stated by other researchers is 110-120 mm [4] 155 - 165 mm [19] 170-210 mm [20]. These differences might be the influence of geographical variations.

The results of the ova diameter experiments reveal that there was a moderate difference in ova size between female specimens. [12] determined the factors influencing on ova characteristics in S. *sihama* and expressed that the ova diameter changed with the stage development, temperature, salinity and timing of the spawning period and age of spawners.

The assessments of *S. sihama* fecundity in this study, since the amount of ova produced by fish specimens (21345 - 73781), were close to those expressed by [20]. Nevertheless, they are slightly below the number estimated. There is a significant difference in the determined value of fecundity recorded by different workers in *S. sihama*. [6] examined on the reproductive biology of *S. sihama* from the Mangalore waters and observed that the ovaries of ripe specimens included 11304-100593 ova per fish. [10] explained the maturity and spawning of *S. sihama* inhabiting in the Pulicat Lake. The fecundity of this species, changed from 31,678 to 2, 88,000. Many reasons have the effect of such considerable variations in fecundity records. These are fertility, the occurrence of spawning, parental care, ova diameter, population density, and even more significantly environment factors which include temperature, salinity and food supply [19].

CONCLUSION

Results of the current research clearly show that *S. sihama* has a long-term reproduction season as pointed out by GSI and large amounts of individuals in Mature and spent stages of maturity in the April-July. The above conclusion provides considerable issues for the management of the *S. sihama* in the Persian Gulf and Oman Sea. Recruitment fishing in earlier than the completed of spawning season and within two months after spawning peak can reduce spawning biomass.

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