

Reproductive traits of the obtuse barracuda *Sphyraena obtusata* Cuvier, 1829 in the Gulf of Mannar, south-east coast of India

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ABSTRACT

The reproductive biology (sex ratio, maturity and fecundity) of the obtuse barracuda *Sphyraena obtusata* Cuvier, 1829 was investigated based on 698 specimens (323 females and 375 males) sampled along the Pamban Coast in the Gulf of Mannar during the years 2018 and 2019. The total length of the samples ranged from 11.8 to 41.6 cm and the overall sex ratio (male:female) was 1:0.86. The length at first maturity (Lm_{50}) was estimated to be 31.26 cm for females and 31.12 cm for males. Monthly changes in the gonadosomatic index (GSI) values and reproductive phases revealed a protracted spawning, extending from September to February with a peak in January. However, the presence of mature females throughout the year indicated year-round spawning behaviour of the species in the region. Detailed histological studies of the ovary further confirmed that the species is a continuous batch spawner. The estimated absolute batch fecundity ranged between 20,520 and 2,22,422 oocytes and the relative batch fecundity ranged from 261 to 754, with an average of 505 ova per gram body weight of fish. The ova size ranged from 0.03 to 0.84 mm (Mean±SE: 0.43±0.04). Fecundity positively correlated with the total length and total weight of the fish, indicating larger females produced a greater number of ova compared to the smaller ones.

Keywords: Batch spawners, Bay of Bengal, Fecundity, Gonadosomatic index (GSI), Histology, Maturity, Reproductive dynamics, Sex ratio

Introduction

Barracudas (Family: Sphyraenidae), also known as pikes, are popular food as well as sport fish in tropical and subtropical waters. They form important component of the commercial fish catch along the Pamban Therkuvadi region, south-east coast of India and are mostly harvested year-round using fish trawls. The barracuda fishery in India is constituted by 10 species of which the obtuse barracuda Sphyraena obtusata Cuvier, 1829 is dominant in the fishery. Along with S. obtusata, other species that were found to contribute significantly to the fishery are S. barracuda, S. forsteri and S. jello (Kasim, 2000). Detailed studies on the reproductive biology of the fish provide essential information on sex ratio, maturity stages, length at first maturity, fecundity, spawning season and spawning periodicity which are important inputs for stock assessment, ultimately leading to sustainable management of the resource (Dias-Neto, 2010; Tsikliras et al., 2013; Trindade-Santos and Freire, 2015). Earlier reports on the reproductive biology of barracudas from India include those on S. putnamae (Rajesh et al., 2020), S. obtusata (Rajesh et al., 2021) and S. jello (Premalatha and Manojkumar, 1990). However, detailed information on the reproductive biology of obtuse barracuda from the Pamban waters in Tamil Nadu on the south-east coast of India, is lacking. Hence, this study on reproductive biology including the microstructure of ovaries of *S. obtusata* was undertaken to support framing appropriate sustainable management measures for the species along the south-east coast of India.

Materials and methods

Sampling and laboratory handling

Monthly sampling was done at the Pamban Therkuvadi Fish Landing Centre (Fig. 1) (9°16' 45.683"N; 79°12' 19.298"E) located on the Gulf of Mannar coast, between January 2018 and December 2019. The total number of samples collected was 698, consisting of 323 females and 375 males. The collected specimens were immediately placed in insulated ice boxes and then transported to the laboratory for further detailed analysis.

Sex ratio

Sex of the individual specimen was identified macroscopically and the distribution of sex ratio (male:female) was estimated based on the number of males and females in the sample. Month-wise and size-wise



Fig. 1. Map showing the sample collection area (Red dot) Pamban Therkuvadi Fishing Harbour, Gulf of Mannar

homogeneity of sex ratio (1:1) was tested using chi-square test with the incorporation of Yates correction factor (Zar, 1996).

Length at first maturity (Lm_{50})

The relative frequency in the percentage of matured individuals under each length group was used to estimate the length at first maturity (Lm_{50}) . The proportion of mature females in the sequential length group was used for the logistic regression analysis following the model given by King (1995) as: $p=e^{(a+b(T/L))/1}+e^{(a+b(TL))}$, where, 'p' is the predicted proportion of mature animals, 'a' and 'b' are the estimated coefficients of the logistic equation and TL is the total length. The coefficients were estimated using the least-squares approach after regression, utilising linearised log-transformed P and TL values. The highest probability estimates of the parameters for the given equation were derived using the SOLVER procedure in MicrosoftTM Excel. The inverse of the coefficients' ratio (-a/b) was used to estimate the Lm₅₀.

Reproductive dynamics

The total body weight (Tw) and gonad weight (Gw) were obtained to the nearest 0.1 g using an electronic weighing balance. The reproductive phases were identified based on the microscopic and macroscopic observations of gonads. The reproductive phases of the female gonad were determined using a five-stage maturity scale (Brown-Peterson *et al.*, 2011) as: (i) Early developing, (ii) Developing, (iii) Spawning capable, (iv) Regressing and (v) Regenerating phases. The ovaries were preserved

in customised Gilson's fluid (Bagenal, 1978) for further fecundity and histological studies.

The reproductive activity was assessed based on the gonadosomatic index (GSI) covering both males and females separately using the equation: GSI=(Gw/Sw)*100 where, GW and SW represent gonad and somatic (gutted) weights, respectively. In the annual reproductive cycle, the peak spawning season was inferred with both monthly GSI values and the occurrence of various stages of maturity in different months of the year. The percentage of spawning-capable and regressing phases in each month was used to determine the seasonality of spawning over the course of the months. Significant differences in the occurrence of spawning females and GSI over months were estimated using the Kruskal-Wallis test.

A total of 35 female gonads collected from different individuals were analysed using histological techniques. The excess fluid on the gonads was patted dry with blotting paper and weighed to the nearest mg. Gonads were fixed in 10% neutral buffered formalin (NBF). Sub-samples were taken from the anterior, middle and posterior portions of the gonads and processed following standard procedures for gonad histology. Based on the gonad microstructure using histology, the gonads were classified into different developmental stages (Brown-Peterson *et al.*, 2011) and were used to confirm the spawning pattern of *S. obtusata*.

The gravimetric method was employed to determine the fecundity of *S. obtusata* ripe female gonads (n=35) by following the method of Murua *et al.* (2003). A sub-sample of 0.001 g was taken from the ovary and placed in a

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modified Gilson solution (Simpson, 1951) and then counted and measured with the help of a stereo zoom microscope (20x) (Nikon DS-Fi2, Japan). Ovary subsamples were removed from the anterior, middle and posterior portion of the ripe ovaries and absolute fecundity was calculated using the equation:

$$\frac{\text{Absolute}}{\text{fecundity}} = \frac{\text{Weight of the ovary * No. of oocytes in the sample}}{\text{Average weight of the sample}}$$

Relative fecundity was stated in terms of the number of oocytes per unit weight (g) of the fish. Using the least square method (Snedecor and Cochran, 1967), regression relation was fitted between absolute fecundity and total length (TL) and weight. The relationship between fecundity and the total length, total weight and gonad weight assessed using the empirical equation, $F=ax^{b}$, where, F=fecundity, a=constant, X=variable (fish length and fish weight or ovary weight) and b=correlation coefficient. The constants 'a' and 'b' were derived by the method of linear least squares. The relationship was transformed into a straight-line logarithmic form based on the equation, logF = loga + b logx (Zupa *et al.*, 2013). For oocytes and ova diameter, a total of 300-400 oocytes were measured from each ripe ovary using a calibrated ocular micrometer under a compound microscope.

Results

Size distribution

The TL of females (n=323) ranged between 11.8 and 41.6 cm, with a mean \pm SE of 28.95 \pm 2.6 cm. For males (n=375), TL ranged from 17.7 to 35.7 cm with a mean length of 27.11 \pm 1.6 cm (Table 1). The estimated modal length was 26.7 cm for females and 27.2 cm for males, respectively. Fish having TL ranging between 20 and 35 cm dominated (93.0%) the commercial catch (Fig. 2). The mean TL of females was not significantly different from that of males (independent t-test, p<0.01).

Table 1. Seasonal dynamics of reproduction in S. obtusata



Fig. 2. Size distribution of female and male S. obtusata in the fishery

Sex ratio

Females constituted 46.3 and males 53.7% of the samples, with an overall male:female ratio of 1:0.86, which did not differ significantly from the expected ratio of 1:1 (chi-square df = 1; p>0.01). Variations in sex ratio were observed in all the months. However, the monthly sex ratio revealed significant differences during January, April, June, July, October and November (chi-square df = 1; p<0.01) (Table 1) where males dominated the fishery.

Length at first maturity (Lm_{50})

For females, 19.6 cm was the measured minimum size at maturity (MSM). The Lm_{50} was estimated at 31.26 cm TL in females (Fig. 3) and 31.12 cm TL in males (Fig. 4). A higher number of females in 'Developing' and 'Early Developing' were observed in the size range between 11.8 and 26.8 cm TL and a higher number of 'spawning capable' females were observed in the size range between 21.8 and 36.8 cm TL (Fig. 5).

Spawning seasons

The monthly mean GSI in females ranged from 0.81 to 3.51. GSI-based studies indicated a long spawning season from September to February for the species, with peaks

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	Total length (cm)	(cm) Sex ratio				Spawning seasonality			
Months	(Mean±SE)	Male	Female	M/F	χ2 (p-value)	Spawning capable	Spawning	GSI (Female)	GSI (Male)
	(n=698)					Female (%)	capable Male (%)	(Mean±SE)	(Mean±SE)
Jan	27.62 ± 0.37	46	19	1:0.41	11.22(0.001)	9.23	26.15	3.51±0.23	1.62 ± 0.11
Feb	28.26 ± 0.52	27	30	1:1.11	0.16(0.691)	28.07	14.04	3.15±0.23	1.17 ± 0.09
Mar	29.44 ± 0.59	23	42	1:1.83	5.55(0.018)	10.77	1.54	2.09 ± 0.20	$1.19{\pm}0.15$
Apr	29.09 ± 0.52	18	43	1:2.39	10.25(0.001)	0.00	0.00	0.81 ± 0.11	$0.48{\pm}0.06$
Jun	26.28 ± 0.57	17	39	1:2.29	8.64(0.003)	32.14	0.00	1.69 ± 0.15	1.07 ± 0.09
Jul	27.13 ± 1.01	3	32	1:10.6	24.03(0.000)	34.29	0.00	2.66 ± 0.52	0.41 ± 0.11
Aug	25.63 ± 0.52	32	24	1:0.75	1.14(0.285)	21.43	8.93	2.02 ± 0.23	$0.93{\pm}0.10$
Sep	27.74 ± 0.40	36	29	1:0.81	0.75(0.385)	24.62	21.54	3.35 ± 0.23	1.07 ± 0.08
Oct	27.41±0.36	65	22	1:0.34	21.25(0.000)	10.34	16.09	2.87 ± 0.39	$0.79{\pm}0.08$
Nov	27.45 ± 0.28	67	16	1:0.24	31.34(0.000)	13.25	34.94	$3.10{\pm}0.36$	1.23 ± 0.06
Dec	30.87 ± 0.37	41	27	1:0.66	2.88(0.090)	39.71	23.53	3.26±0.26	1.41 ± 0.11



Fig. 3. Length at first maturity (Lm₅₀) of female S. obtusata



Fig. 4. Length at first maturity (Lm_{50}) of male S. obtusata



Developing, Early developing, Regenerating, Regressing, Spawning capable

Fig. 5. Distribution of maturity stages across length classes in female *S. obtusata*

in January (3.51), September (3.35), December (3.26) and November (3.10) while April had the lowest reproductive activity (0.81). Males exhibited major spawning activity in January (1.62), December (1.41), November (1.23) and February (1.17) (Table 1). The monthly analysis of maturity phases of female *S. obtusata* revealed the presence of 'spawning capable' females almost throughout the year. Maximum number of spawning females were observed in December (39.71%) and July (34.29%) and maximum number of spawning males were observed during November (34.94%) and January (26.15%) (Table 1). Samples could not be collected during May due to the fishing ban in place from April-June along the coast. The presence of all maturity stages almost throughout the year indicated that the species is a year-round spawner.

Histological observation of gonad

The structural changes observed in the oocytes of S. obtusata are shown in (Fig. 6). Immature ovarian tissues were observed to have oogonia and primary growth (PG) oocytes (Fig. 6a). Developing ovarian tissues were distinguished by the presence of cortical alveoli (CA) in the cytoplasm of the oocytes. The presence of cortical alveolar oocytes signified the start of vitellogenesis (Vtg.) and the density of this in the cytoplasm was used to demarcate the two stages of vitellogenesis, Vtg.1 and Vtg. 2 (Fig. 6b). A mature ovary that is spawning capable is characterised by the appearance of Vtg. 3 (Fig. 6c). However, batch spawners had oocytes in all stages of vitellogenesis and are not restricted to Vtg. 3 (Fig. 6f). Post-ovulatory follicular complex (POF) and atretic oocytes (A) were observed in the regressing phase of the ovary when the active reproductive cycle ends (Fig. 6d). The regenerating phase was categorised by ovarian tissue comprising only oogonia and PG oocytes (Fig. 6e). During this stage of maturity, fishes are sexually mature but without any reproductive efficiency. The presence of different developmental stages in ovarian tissues confirmed that this species is a multiple spawner releasing ripe eggs in batches during the spawning period.

Fecundity

The estimated absolute fecundity ranged from 20,520 to 2,22,422 oocytes (Mean±SE: 1,01,527±9,588) for specimens measuring between 23.5 and 37.7 cm and weighing from 78.68 to 295 g. The relative fecundity ranged from 261 to 754 ova per gram body weight (Mean±SE: 517±32) with an average of 547 ova. Fecundity showed positive correlation with total length ($R^2 = 0.90$) (Fig. 7a) and body weight ($R^2 = 0.88$) (Fig. 7b) indicating that larger females produced a greater number of eggs as compared to smaller ones. The ova size ranged from 0.03 to 0.84 mm (Mean±SE: 0.43±0.04).

Discussion

Sustainable exploitation of any fish needs a systematic study of its reproductive dynamics, such as sex ratio,

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Fig. 6. Histological sections showing different phases of *S. obtusata ovary*. (a) Immature, (b) Developing, (c) Maturing, (d) Regressing, (e) Regenerating and (f) Batch spawners. OW - Ovarian wall, PG - Primary growth oocytes, CA - Cortical alveolar oocytes, Vtg.1 - Vitellogenic 1, Vtg. 2 - Vitellogenic 2, Vtg.3 - Vitellogenic 3, POF - Post-ovulatory follicles, A - Atretic oocytes, MB - Muscle bundle

(e)



Fig. 7. Relationship of fecundity with (a) Total length and (b) Body weight in S. obtusata

length at first maturity, maturity phases, spawning season and fecundity. A similar length range for the species in the fishery has been reported in earlier studies (Premalatha and Manojkumar, 1990; Kasim *et al.*, 1990; Jaiswar *et al.*, 2004; Sivashantini *et al.*, 2009; Najmudeen *et al.*, 2015). The preponderance of the specimens was between 20 and 35 cm, even though the smallest size in the fishery was 11.8 cm, revealing the lack or occasional presence of juveniles and sub-adults in the area where trawls are used for fishing. The reason may further be attributed to the spawning behaviour of barracudas, generally in deep waters at the edge of the continental shelf, as reported by Kasim (2000).

(d)

The present study on *S. obtusata*, revealed the overall sex ratio (male:female) to be 1:0.86, indicating slight dominance of males in the population. A higher

proportion of males in the *S. obtusata* population has been reported earlier by Rajesh *et al.* (2021) along the southwest coast of India; in *S. obtusata*, *S. chrysotaenia* and *S. jello* by Okera (1982) in the northern Australian waters and in *S. flavicauda* by Allam *et al.* (2004) in the Egyptian Mediterranean waters.

(f)

According to a study by Allam *et al.* (2004), many *Sphyraena* species had male dominance in smaller length groups while females dominated larger length groups. For *S. chrysotaenia* and *S. sphyraena* from the Egyptian Mediterranean waters, identical findings were reported by Rizkalla (1985). The variation in sex ratio in barracudas could be due to the congregation of sexes for feeding, breeding and pre-spawning migration of males (De Sylva, 1963, 1973).

The length at first maturity for females of *S. obtusata* obtained in this study was larger than previous estimates of 21.3 cm in Karnataka by Rajesh *et al.* (2021) and 20 cm in Kochi waters by Premalatha and Manojkumar (1990). This could be a consequence of the fact that the current research was carried out in a different ecosystem and that various other variables, including stock differences, climate, food availability, migration and other environmental factors may have an impact (Sabrah *et al.*, 2018).

The observed monthly variation in GSI values indicated a protracted spawning season that lasted from September to February, which is consistent with previous research. The spawning season in this study was nearly identical to that reported by Rajesh *et al.* (2021), who reported the highest GSI values in November, April, October and March in Karnataka waters. *S. obtusata* spawns at its peak between October and March, while *S. jello* spawns between April and July in Kochi waters (Premalatha and Manojkumar, 1990). Observations made by Allam *et al.* (2004) on the spawning periodicity of smallerbarracuda species *S. chrysotaenia* (May to October), *S. flavicauda* (May to September) and *S. sphyraena* (April to August) from Egyptian Mediterranean waters of Alexandria, were similar to the findings of the present study.

According to Rajesh et al. (2021), the obtuse barracuda spawned throughout the year in Karnataka waters, with peak periods in October-November and April-May. According to Kasim (2000), barracuda breeding occurred in two phases in Thoothukudi waters, the first between October and February and the second between June and August. S. obtusata in Kochi waters spawned for six months from October to March, with a peak in November and December (Premalatha and Manojkumar, 1990). According to Bal and Rao (1985), the spawning season of S. obtusata in Mumbai waters lasted from November to February. The presence of spawning capable females in Pamban waters throughout the year, with the highest in February and November, agrees with Bal and Rao (1985) and indicates that this species has a year-round spawning behaviour. Moreover, histological studies supported the continuous and batch spawning phenomena, which are thought to be a population-sustenance adaptation.

The fecundity of *S. obtusata* was estimated to range between 20,520 and 2,22,422 oocytes in this study. Rajesh *et al.* (2021) estimated a fecundity range of 82,722 to 3,79,421 eggs in Karnataka waters, while Premalatha and Manojkumar (1990) observed fecundity for *S. obtusata* in Kochi waters in the range of 1,50,000 to 2,00,000. However, the fecundity of *S. obtusata* observed in the present study is comparable to that reported for *S. barracuda* from Florida Bay (Kadison *et al.*, 2010) and *S. chrysotaenia* (74,399 to 2,41,853), *S. flavicauda* (84,197

to 2,60,549) and S. sphyraena (46,778 to 1,03,453) from Mediterranean waters (Allam et al., 2004). The marginal differences observed in the fecundity are possibly due to differences in the ecological conditions or differences in representative samples. The ova diameter recorded in the present study, were in agreement with that reported by Rajesh et al. (2021) for S. obtusata (0.04 to 0.83 mm) in Karnataka waters and also on par with S. chrysotaenia (0.05 to 0.85 mm), S. flavicauda (0.05 to 1.20 mm) and S. sphyraena (0.05 to 1.50 mm) in Egyptian Mediterranean waters (Allam et al., 2004). Based on ova diameter, continuous graduation of oocytes, size and modes of oocytes Sphyraena species are reported as fractional batch spawners (Allam et al., 2004). Based on macroscopic observation of gonads, Rajesh et al. (2021) reported that S. obtusata was a batch spawner. The results of the present study indicated that S. obtusata is a continuous batch spawner.

S. obtusata is an economically important species in the Gulf of Mannar ecosystem and detailed microscopic and macroscopic studies of its reproductive features provide baseline information for understanding the population dynamics which would help in formulating strategies for sustainable exploitation and conservation of this species in the region.

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