

Reproductive biology of under threatened fish species *Ompok bimaculatus* (Bloch 1794) from Vamsadhara River, Andhra Pradesh.

Comment [D1]: Reproductive biology of near threatened Indian Butter catfish *Ompok bimaculatus* (Bloch, 1794) from Vamsadhara River, Andhra Pradesh

ABSTRACT

Ompok bimaculatus (Bloch 1794) is a near threatened catfish (IUCN, 2025), of freshwaters, belongs to the family Siluridae of the order Siluriformes. Due to its rich lipo-protein content and soft bony structure it becomes delicious and nutritiously highly valuable food fish species. At present *Ompok bimaculatus* facing decline in the natural populations, due to several anthropogenic factors such as indiscriminate fishing during the breeding season, unscientific use of pesticide in agricultural fields, gradual siltation in the freshwater habitat, depletion of water velocity as a result of decrease in water volume etc. Because of such drastic reduction in its population and a very restricted distribution in the freshwater ecosystem, Present work aims to an experimental analysis of reproductive biology of *Ompok bimaculatus*. Standardized knowledge on factors like size at maturity, spawning, sex-ratio, ova diameter studies and fecundity are essential pre-requisites in fishery management and conservation.

Comment [D2]: Recommended to re arrange sentence scientific sequence

Keywords: Food fish, Maturity, Gonado somatic Index, Gastro Somatic Index, fecundity, spawning,

Comment [D3]: This will come in food and feeding biology, recommended to add other terminology relevant to reproductive biology

Introduction

The fresh water fish fauna of India is a group of nearly 2500 species, comprising into three main families known as, Cyprinidae, Siluridae and Channidae, inhabiting in the inland ecosystems (Arthi et al 2013). However, majority of fishes are yet to be analysed well. Growth of the fish, length and weight parameters, have positive influence on reproduction. In general, the fecundity is more closely related to length than the somatic weight or ovary weight. Fecundity is elucidated in simple terms as number of potential eggs in the ovary. The discharge of eggs is known by a process called 'spawning'. Studies on the reproductive biology of fish is essential in evaluating

Comment [D4]: Recommended to Update data due to old record, and Suggested cross check number of fresh water fish species in India is less than 1000 and In India, 3523 fish species belonging to 1097 genera and 272 families of 55 orders are recorded as per recent studies of ZSI

Comment [D5]: Italics

Comment [D6]: Recommended reference of species recent data: Citation: Kosygin, L., Mohapatra, A., Bineesh, K.K., Sharma, I., Jadhav, S.S., Khyriam, D. (2024). Fauna of India

the commercial potentialities of its stock, life history, cultural practice and management of small fish's species (Eyo et al. 2014). Reproductive biology i.e., fecundity, spawning, sex ratio, etc. are among the import aspects of the biology of fishes which must be understood to explain the variations in the level of populations as well as to make efforts to increase the amount of yield. (Gupta, et al. 2014).

Comment [D7]: Italics

Comment [D8]: Italics

Materials and methods

40 female individuals of *Ompok bimaculatus* species were collected randomly throughout the year from Mahendranaya a small tributary of Vamsadhara River, Andhra Pradesh and local markets. The collected fish weight and length were taken at the sampling site, afterward preserved in 4% formalin, further transferred and preserved in 10% formalin for laboratory studies.

Comment [D9]: Size of the samples very low and for reproductive biology both male and female population scientifically represents in data and materials and methods references and map of study area and original image of fish species suggested to add

Comment [D10]: Suggested to cross check & update as Mahendranaya is not a small tributary of the Vamsadhara River; it is considered a major tributary of the Vamsadhara River, originating in the Gajapati district of Odisha and joining the Vamsadhara in Andhra Pradesh, Mahendranaya tributary originates from the Mahendragiri Hills. It joins the Vamsadhara river upstream of the Gotta barrage.

Comment [D11]: Original size of Ova might shrink as the smallest oocytes increased ~11 % in diameter and ~15 % in weight when fixed in formalin, whereas the largest oocytes decreased about 4 % in diameter and 7 % in weight. However lowest concentration suggested 2% neutrally-buffered formalin concentration was chosen because it was the lowest possible concentration that would ensure proper oocyte preservation while minimizing changes in oocyte size and appearance.

Comment [D12]: Add reference Author

Maturity stage

Maturity stages were dispensed based on the degree of development of testes in males and ovaries in females. Cycle of gonadal maturation has been studied by macroscopic and microscopic monthly examination of the different maturation stages of gonad; male and female gonads have then been grouped into different gonadal stages of development according to Nikolsky (1963). The percentage of mature individuals relative to immature and premature individuals was calculated for each age group and fitted to the logistic model $Y = [1 + e^{-(aX + b)}]^{-1}$, where Y is the proportion of individuals that were mature at age X, a and b are empirical parameters.

Gonado-Somatic Index (GSI %)

The gonads expand to mature as the fish grow. Till the maturity reaches the ripened stage, the relationship of growth of gonads with that of fish is directly proportionate. However, with spawning, as the ova are delivered and the gonad is spent, the weight of it is likely to decrease. Hence, the GSI steadily increases till the development of gonads into ripening. Then there is a noteworthy decline with spawning. This index is calculated for both males and females

separately and the monthly mean value was then plotted. Gonado-somatic index (GSI) was determined following the equation of Parameswarn et al. (1974):

Comment [D13]: Italics

$$\text{GSI} = (\text{Weight of gonad} / \text{Weight of fish}) \times 100.$$

Fecundity

Gravimetrically the fecundity of the fish was observed. The external connective tissues were detached carefully from the surface of the ovaries. With the help of blotting paper moisture of the ovaries was removed. Weight of the ovaries was recorded with fine electronic balance. Then 0.01 g of each ovary was taken out separately from anterior, middle and posterior regions of each ovarian lobe. The number of mature and immature eggs for each portion was sorted out and counted. The fecundity of the studied fish was observed adopting the methodology of LeCren (1951):

$$\text{Fecundity} = (\text{number of eggs in the sample} \times \text{weight of gonad}) / \text{weight of the sample}.$$

Relationship of fecundity with various body dimensions such as body length, body weight, ovary length and ovary weight were transferred into log transformed and least squares regression equation (Bagenal 1978):

Ova diameter

At different stages of the maturity the diameter of the ovum was measured with ocular micrometer under stereoscopic Olympus microscope along the longest axis of the ovum (Clark 1934).

Comment [D14]: The condition factor (K) suggested to be calculated from the relationship of Fulton [17]: $K = 100 \times (W/L^3)$

Comment [D15]: Length at first sexual maturity (L50) recommended to be calculated for both male and female

Comment [D16]: logistic model $Y = [1 + e(aX + b)]$ formula written in materials but not used in results section, if in sufficient data wait for some time suggested to collect data male with corresponding female fish population

Results

Determination of Maturity stage

Four maturity stages of ovaries have been recognized on basis of microscopic and histological studies which are stated below:

Comment [D17]: Prior to this sex ratio suggested to be estimated for each month from the proportion of females to males articulated as the percentage of the total sample. For this purpose the body colour and other morphological features were noticed under binocular microscope

Comment [D18]: Recommended title and table on Gonadal maturity stages of *O. binaculatus* both male and females samples used in present study

Stage I (Immature): Ovaries translucent, colorless and thread like in appearance. Ova are not visible to naked eyes, but under microscope ova are irregular in shape, with a clear nucleus and transparent as yet yolk is not formed.

Stage II (Maturing): Ovaries yellowish white in colour and still thread like in appearance; ova are visible to naked eyes; under microscope ova are spherical in shape, partly opaque in appearance due to commencement of yolk development.

Stage III (Mature): Ovaries light yellowish in colour and enlarged in size; ova are clearly visible to naked eyes; under microscope spherical in shape and opaque in appearance except the transparent periphery (Fig. 1).

Stage IV (Ripe): Ovaries deep yellow in colour; with maximum size. Under microscope, ova are spherical in shape and opaque due to huge amount of yolk present. In this stage, ova are with their full size and start to become liberated through oviducts on putting light pressure on the abdomen.

Stage V (Spent): Ovaries pale whitish in colour; almost thread like in appearance resembling the Stage I ovary; distinguishable only with the presence of the pigmented covering of the ovary. Under microscope, ova are visible, very small in size, irregular in shape.

Immature gonads (Stage I) have been observed from March to May; highest percentage being observed in March while lowest percentage in May. Stage II (maturing gonad) first have been observed in May and available till September; highest percentage being observed in July and lowest percentage in September. Stage III (mature gonad) have been observed from March to October; highest percentage being observed in September and lowest percentage in March.

Ripe (Stage IV) have been observed from April to December with highest percentage being observed in November and lowest percentage in April. Spent (Stage V) have been observed in October to December; highest percentage being observed in December and lowest percentage in October.

Gonado-Somatic Index (GSI %)

The GSI value was low during September to April and increased monthly and attained its maximum level in June. In the female there were very low GSI (%) as observed during January

Comment [D19]: Microscopic examination of different size and at different stages of ova maturation stages in ovaries images suggested to be added at relevant section figures

Comment [D20]: Relevant monthly data variation in GSI table suggested to add

Comment [D21]: GSI month wise for male & Females length wise and body and ovary weight wise variation graph recommended to be added

to March. Thus, it represented the occurrence of pre-spawning period during January to March (Table:1).

The GSI value increased gradually to reach at the greater value in May and June (Spawning period), which was later on declined to minimum value.

Hence, it indicated that the pre-spawning period was developed during January to March. The GSI value enhanced gradually to a higher value in June (Spawning period) and then declined to minimum value in September (Table:1).

Fecundity

The fecundity of the studied fish individual varied from 3724 to 41552 eggs/fish. The mean value of eggs per gram of ovary varied in month from a minimum of 152 eggs/gm in September to a maximum of 2267 eggs/gm in July. The monthly variation of fecundity was calculated as mean sample. Fecundity was high during May- July, which declined sharply during August October and remained constant till January. So, fecundity was directly proportional to the length and weight of the studied fish species. Fecundity was also showing direct relationship with the weight of the ovary (Table:1).

Ova diameter

The diameter of immature eggs ranged from 0.211 mm- 1.345 mm. While before spawning the diameter of the eggs varied from 0.548 - 0.634 mm. In mature eggs the diameter ranged from 0.727 mm- 1.345 mm. The frequency of occurrence of eggs at different diameter showed that immature eggs were found out during February - April and mature eggs in the month of May-July (Table:1). Since August the development of mature eggs decreased quantitatively.

Discussion

Ompok bimaculatus the females were quantitatively dominant over males (Banik et al. 2012). Taiwo and Aransiola (2001) also noticed similar observations in some other catfish species. However, Ham (1981) pointed out that in cat fishes such dominance by female was due to certain environmental conditions while Fagade et al. (1979) pointed out the phenomenon because

Comment [D22]: Relevant Scientific terminology based data collected, analysed and presented and monthly data variation in GSI table suggested to add

Comment [D23]: Suggested to use absolute and relative fecundity if noticed data of absolute fecundity in present study

Comment [D24]: Re correct statement in researchers point of view

Comment [D25]: Ova diameter range

Comment [D26]: Italics

Comment [D27]: Not found/matching with suggested to add in reference

Comment [D28]: Not found/matching with suggested to add in reference

of population regulation. Chakrabarty et al. (2007) pointed out that the sexual maturity of *Ompok bimaculatus* discloses that during stage of maturity of the female the length reaches to a minimum of 17.0 cm. The present observation also depicts that the females are relatively longer (Banik and Malla 2011). For understanding the dynamics of the gonads and to assess reproductive performance of species information of the phase of gonad development is virtually important. In this species the stage of maturity is noticed with the volume of gonad. Variations in the form of gonad were found to be noticed during filli-form appearance of the immature stage to the development of mature lobular stage during the maturation process. The maturation process stops with the formation of wrinkle-shaped gonad after completion of spawning. At that period the gonad looks like dull and lobule. Lamai (1993) also observed similar findings on the reproductive cycle and gonad development stages in different fish species.

Comment [D29]: Italics

De laming (1972) deliberated the GSI (%) in teleosts and has used this parameter as indicator of the spawning period in teleost species. Further, considered the use of GSI (%) in reproductive biology which is noticed to be more appropriate when associated with other indicators of the reproduction under macroscopic observations (DeMartin and Lau 1999). Dmitincko (1970) pointed out that the GSI for female was always higher than that for male probably due to greater weight of the ripe female gonad. The fluctuations of GSI value confirmed that *Ompok bimaculatus* began to spawn in June and continued till August. In the present studies fecundity is relatively smaller in comparison to the other catfishes. Fecundity of *Silurus triostegus* in Ataturk Dam Lake varied from 6800 to 120,300 (Oymak et al. 2001).

Comment [D30]: Cross check reference correct spelling in author name

Variations in fecundity of fish species may be due to selectively different factors such as feeding, food abundance, species differentiation, nutritional resources etc. (Bagenal 1978). Peak season of fecundity of *Ompok* species coincides with the onset of rain fall and flood water during May to August (Vanderwaals 1974). Harding (1966) stated that most of the tropical fishes are adopted to breed on the rising flood conditions. Thus, allowing the juveniles to take full advantage of the flooded banks for smooth feeding purpose and thus protected from predation. The choice of a particular season in fishes for breeding is influenced by various factors among these are changes in water quality characteristics, inter specific interactions and occurrence of suitable spawning sites (Zaret 1980 and Ward and Samarakoon 1981). The present findings also reveal a significant

Comment [D31]: Italics

correlation between fecundity and ovary weight. The diameter of ova during peak season is ranged from 0.727 mm- 1.345 mm in the present observations. Douglas (1979) also noticed similar findings in *C. auratus*. The stage of the ovary of the studied fish species during October to January shows that the spawning period is lacking during a period from October to January.

Ethical statement

The fish species studied in the present study are not protected under The Wildlife Protection Act, 1972 (Last amended in 2013), Government of India, All the guidelines on animal use and care were followed accordingly.

Comment [D32]: Species photos and fishes capture gear used method not specified

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Comment [D33]: Reference year

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Comment [D35]: Reference not matching suggested to cross check add and arrange in reference section

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Comment [D36]: This reference not added text if missed to add suggested to add or if not referred can deleted

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Table 1: Total length (cm), total weight (g), weight of ovary (g), gonado-somatic index (GSI%), fecundity, ova diameter (mm) of 40 individuals of *Ompok bimaculatus*

Months	Total Length (cm)	Total Weight (g)	Weight of Ovary (g)	Gonado-Somatic Index (GSI %)	Fecundity	Mean Ova Diameter (mm)
January	22.3	42.15	2.340	5.55	3724	0.211
	28.2	73.57	3.992	5.42	4213	0.327
	29.7	85.43	3.794	4.44	5698	0.243
February	26.4	88.20	3.435	3.89	4327	0.578
	27.2	74.50	4.232	5.68	3767	0.427
	28.5	90.20	4.878	5.40	3778	0.573
March	26.2	87.52	4.987	5.69	5520	0.544
	27.4	95.20	5.125	5.38	6549	0.634
	29.2	99.00	4.997	5.04	7289	0.765
April	25.4	88.20	4.237	4.80	7554	0.727
	26.3	85.75	4.759	5.54	8452	0.654
	24.7	68.07	5.880	8.63	8642	0.731
May	27.8	70.52	7.244	10.27	9239	0.765
	20.2	35.20	6.528	18.54	9768	0.711
	19.5	54.70	7.525	13.75	10880	0.755
June	24.7	70.50	8.721	12.37	12747	0.757
	20.5	38.00	8.280	21.78	12837	0.848
	34.7	105.50	10.280	9.74	15554	0.886
July	34.5	107.50	15.322	14.25	17121	1.102
	31.2	136.50	16.250	11.90	18009	1.107
	36.0	142.53	17.242	12.09	22552	0.928
August	32.4	145.24	16.827	11.58	24196	1.064
	31.0	115.00	13.180	11.46	27220	1.115
	28.7	107.20	12.412	11.57	23186	0.949
September	29.5	107.23	11.742	10.95	25597	0.756
	25.7	96.80	8.720	9.00	20157	0.812
	31.5	124.07	12.252	9.87	28525	1.345
	32.7	127.15	12.774	10.04	29699	0.543

Comment [D37]: Total body weight of fish

Comment [D38]: Specify absolute or relative fecundity in numbers

Comment [D39]: Fecundity (Numbers)

Comment [D40]: The body weight of matured fish mentioned very low, than normal check actual data and specify the units

October	24.8	77.50	6.235	8.04	15872	0.992
	20.2	44.50	6.335	14.23	13785	0.976
	22.9	34.52	7.452	21.58	14293	1.042
	18.8	25.00	4.248	16.99	7934	0.875
November	19.7	27.07	5.990	22.12	13781	0.720
	18.4	23.50	4.087	17.39	9543	0.875
	20.5	28.50	5.209	18.27	11570	0.885
	21.7	32.72	4.528	13.82	9527	0.943
December	23.7	40.50	5.178	12.78	7190	0.925
	24.7	34.50	4.987	14.45	11192	0.942
	23.5	44.00	5.778	13.13	9934	0.875
	27.7	64.50	6.995	10.84	11123	1.175

Comment [D41]: Original Data cross check, add original body weight of fish and specify the units Length and weight of fish



Fig.1 Matured ovary of *bimaculatus*

Comment [D42]: Length and weight of Ovary suggested to be add

Ompak