



Hormonal Manipulation in Breeding of Pabda, *Ompok bimaculatus* (Bloch, 1794) Under Captive Condition

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ABSTRACT: *Ompok bimaculatus*, commonly known as butter catfish, is a significant food fish in India's aquaculture industry due to its high value and potential for diversification. Twelve separate induced breeding trials were carried out with 54 pairs of brooders, from September to January 2023. The most appropriate, reduced, and standardised hormonal dosages were 0.4 ml kg⁻¹ for male and 0.8 ml kg⁻¹ for female body weight, respectively. Fish spawned naturally in captive conditions. The relative fecundity ranged from 372.4 ± 2.85 eggs/g of body weight of the female. The ovulation for induced natural egg laying was 10±2 hrs at a temperature of 29±1°C. The fertilization rate was 82±9%. Fertilized eggs were distributed in the glass incubation chamber with an artificial aeration system. After fertilization, the egg hatched out in 18±2 hours at 29 ± 1 °C. In 57.5±3 hours, the yolk sac was fully absorbed, and the hatching rate was 87±3%.

Keywords: Pabda, Induced breeding, captivity, Natural spawning.

INTRODUCTION

Ompok bimaculatus (Bloch, 1794), also known as the butter catfish, is a significant freshwater fish with high medicinal and commercial value. This non-air-breathing, omnivore fish feeds on the surface level of water and typically consumes a wide variety of living things, including algae, plankton, mosquito larvae, crustaceans, aquatic insects, and protozoa (Bandyopadhyay, 2019). In Tripura, India, the butter catfish was designated as the official state fish. The fish is extensively dispersed and frequently encountered in freshwater environments, such as rivers, canals, reservoirs, and streams. The IUCN Red List (2010) listed this endangered species because of overexploitation, habitat loss, disease, pollution, river siltation, poisoning, and harmful fishing practices. In India, its market value and demand have both been strong. Pabda contains few intermuscular bones, fine flesh with a soft meat texture, high nutritional value, good taste, and omega-3 fatty acids, which lower blood pressure lessen the risk of stroke and maintain a healthy heart (Ghosh *et al.*, 2022). But in nature, they are becoming extinct. The catfish seed manufacturers saw a

decline in revenue of more than 50% (FAO, 2022). In India, one of the challenges faced in catfish farming is the lack of availability of catfish seeds (Jayasankar *et al.*, 2018). Thus, the seed production is crucial.

While there are huge reports on pabda breeding techniques including the stripping of female fish and the collecting of male fish testicles in captivity, non-sacrificial pabda breeding techniques are rare. This experiment details a study that shows how to successfully breed pabda and produce seeds in an artificial aquatic environment without sacrificing male fish.

MATERIALS AND METHODS

Collection and site for the experiment of *O. bimaculatus*. The brood fish of *O. bimaculatus* were collected from 'Bandhab hatchery' in Diamond Harbour, West Bengal (Latitude 22.228250, Longitude 88.183553) during August 2023 after conditioning. The experimental Site was Fish-breeding Training Centre, Panchpota, Garia, Kolkata-700152 (Latitude-22.467519, Longitude-88.419551).

Disinfection and Acclimatization. After transportation, fish will be disinfected by bath treatment

with Potassium permanganate (KMnO₄) @10ppm until fishes show the symptoms of stress. The glass chambers are sanitized by using KMnO₄ @10ppm and also sanitize the required material (Aerator, air-stone, filter, hand net, and other used materials). Before the start of the experiment, fish were acclimated for two days under control conditions.

Experimental setup for captive breeding. Portable glass tank pabda hatchery, I use six brooder tanks (4'×2'×1'), one breeding pool (6'×2'×1') and one incubation pool (4'×2'×1'). from September to January 2023

Broodstock management. There have used six glass chambers with a dimension of (4'×2'×1') were used for the rearing of brooder fish and the average size of the

fish was 22.5 cm in length and a body weight of around 60.35 g. After two days of procurement, the trial meal was fed to the brooders. The fish were fed on chicken liver and egg yolk to bring maturity and gonadal development. The boiled chicken liver was cut into small pieces and the boiled egg yolk was mixed with floating feed. The chicken liver and egg yolk mix in a 1:1 ratio and given @ 3% of total body mass twice a day at 6:00 AM and 5:30 PM.

After one hour of feeding, regular leftover food and waste were sucked out using a siphon. Every four days, the filter medium was cleaned before feeding. Three times a week, only 50 - 100% of the water was changed to get rid of the collected faces (Chakraborty *et al.*, 2010; Chakrabarti *et al.*, 2009).



Fig. 1 (A) Captive rearing of brood fish (B) Experimental diets.

Gonadosomatic index (GSI): Total body length and weight was measured before dissection and separation of the gonads. Then shup scalps and bone cutter, an incision was opened on dual ventral side of the abdomen. The testes were carefully removed and measured.

$$\text{GSI} = (\text{Weight of gonads}) / (\text{Total weight of fish}) \times 100$$

Fecundity: The Fecundity is measured by using the formula

$$\text{Fecundity} = (\text{Total ovary weight} \times \text{No. of sample eggs}) / \text{Weight of total eggs}$$

Breeding procedure: Segregated the perfect mature males and females from the rearing brooder tank. Then set the breeding tank by taking males and females in a 2:1 ratio and use the synthetic hormone Hatchme for breeding which contains SGnRH, Domperidone, and Propylene Glycol by diluting with 54-95% sterile water. The hormonal dose was minimized and standardized after 12 trials. The breeding setup was done before hormonal injection (Intraperitoneal). Initially, the breeding cage was placed inside the breeding tank. It not only helps to separate the brooders after breeding but also increases the egg efficiency. Following that, measurements were taken and documented without upsetting the breeding pair, and the water's temperature and dissolved oxygen content were checked. The fish spawned naturally in a glass breeding chamber under captive conditions, without sacrificing males and without stripping females. After spawning the brownish shrinking semi-adhesive eggs come out from the cage and settle down into the bottommost of the tank. After the complete breeding, the eggs and the brooder were separated easily and the fertilized eggs were collected and distributed in the hatching tray inside the glass hatching chamber (4'×2'×1') under an aeration system.

The ovulation, fertilization and hatching rate were calculated as below:

$$\text{Ovulation rate (\%)} = (\text{Total no. of eggs spawning} / \text{Total no. of fecundity}) \times 100$$

$$\text{Fertilization rate (\%)} = (\text{Total no. of fertilized eggs} / \text{Total no. of eggs}) \times 100$$

$$\text{Hatching rate (\%)} = (\text{Total no. of eggs hatched} / \text{Total no. of fertilized eggs}) \times 100$$



Fig. 2. Injection process.



Fig. 3. Glass breeding chamber.

Physico-chemical parameters analysis for captive breeding. The water temperature was recorded daily. The water samples were tested to study the various physico-chemical parameters for broodstock rearing,

breeding-cum-egg laying and incubation-cum-hatching. DO, pH, Ammonia, Nitrite, Nitrate, and Total Hardness were tested by using test kits for “Bionex” company. The water temperature was recorded with the help of a mercury thermometer.

RESULT AND DISCUSSION

Gonado Somatic Index (GSI). The GSI (Gonado Somatic Index) of *O. bimaculatus* was found out of 2 months from August and September 2023. The colour of the mature gonad is slightly brownish. The female specimen attending an average weight of 55 gm and gonad weight of an average of 9 gm was measured. The GSI (Gonado Somatic Index) for the gravid females ranged from 17.62 – 15.10 gm with an average of 16.36 gm.

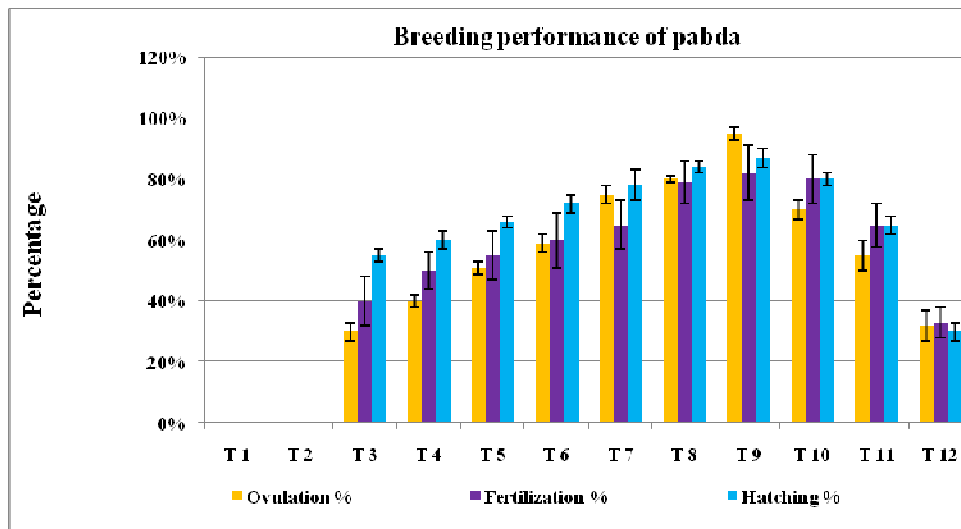
Fecundity. Pre-spawning fecundity of *O. bimaculatus* was found to be 372.4 ± 2.85 eggs/g body weight of the female. The lowest number of ova produced was 15521 in a female having a length of 17.5 cm and weight of 42 g. A maximum of 26267 ova was produced by a female having a length of 23.5 cm and a weight of 70 g.

Standardize the hormonal doses and study the induced natural breeding of Pabda. To standardize the hormonal dose of *O. bimaculatus* found at the end of the 12-time breeding experiment in Trial 1 to Trial 12 are 1.6, 1.5, 1.4, 1.3, 1.2, 1.1, 1.0, 0.9, 0.8, 0.7, 0.6 and 0.5 ml/kg for females and 0.80, 0.75, 0.70, 0.65, 0.60, 0.55, 0.50, 0.45, 0.40, 0.35, 0.30 and 0.25 ml/kg for the males. I dilute 57% to 95% water with the hormone during the total breeding experiment. The highest result and standard dose (Trial 9) @0.8 ml/kg for females and 0.4 ml/kg for males with the maximum ovulation %, fertilization %, and hatching % are $95 \pm 2\%$, $82 \pm 9\%$, and $87 \pm 3\%$ respectively. The lowest result was found in T1 and T2, in which ovulation %, fertilization %, and hatching % are 0% (Table 1). The sacrifice-bred using ovaprim at a rate of 1.0 ml/kg body weight for males and 1-1.5 ml/kg body weight for females (Chakrabarti *et al.*, 2010). The male and female ratio is 1:1 (Mahapatra, 2023). The hatching period of 24–25 hours (Bhowmik *et al.*, 2000).

Table 1: Data on induced natural spawning of pabda.

No. of trials	Total body weight of brooders (g)		No. of Brooders		Dose of hormone (ml/kg)		Time To Oval action(h)	Oval action (%)	Fertilization (%)	Hatching Time (h)	Hatching (%)	Physico-chemical condition	
	F	M	F	M	F	M						WT (°C)	DO(ppm)
T1	200	295	3	6	1.6	0.80	-	-	-	-	-	28±2	5±1
T2	210	310	3	6	1.5	0.75	-	-	-	-	-	29±3	6±5
T3	200	300	3	6	1.4	0.70	12±2	30±3	40±8	19±3	55±2	28±4	5±1
T4	205	295	3	6	1.3	0.65	11.5±2	40±2	50±6	19±3	60±3	28±3	5±1
T5	208	302	3	6	1.2	0.60	11±2.5	51±2	55±8	18±3	66±2	29±8	6±1
T6	200	312	3	6	1.1	0.55	11±2	59±3	60±9	17±3	72±3	30±2	7±5
T7	189	298	3	6	1.0	0.50	10.5±3	75±3	65±8	18±2	78±5	28±3	6±1
T8	190	315	3	6	0.9	0.45	10±3	80±1	79±7	17±3	84±2	29±9	7±1
T9	210	325	3	6	0.8	0.40	10±2	95±2	82±9	18±2	87±3	29±1	6±1
T10	200	310	3	6	0.7	0.35	11±2	70±3	80±8	18±3	80±2	29±7	6±5
T11	206	320	3	6	0.6	0.30	11±2.5	55±5	65±7	17±3	65±3	30±3	7±1
T12	200	315	3	6	0.5	0.25	11±2	32±5	33±5	17±4	30±3	29±5	7±1

Abbreviation: T1 h- hour, g- gram, F – Female; M – Male; WT- Water temperature; DO-Dissolved oxygen. Value is present as Mean ±SE.



Abbreviation: Ovulation%, Fertilization%, and Hatching% in the 12 trials (T1 – T12) of *O. bimaculatus*. Value is present as Mean ±SE.

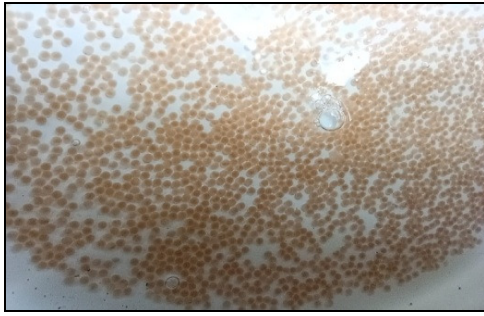


Fig. 4. fertilized eggs.

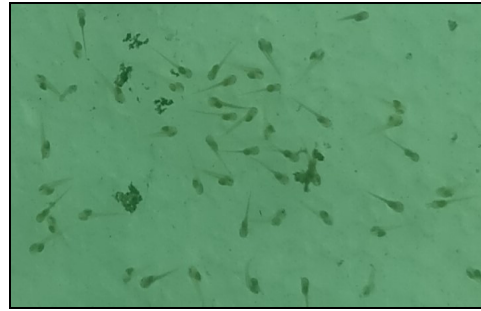


Fig. 5. Hatchling.

Water quality parameters analysed. Several physico-chemical characteristics were examined utilizing a test kit that was used in the broodstock raising,

breeding/egg-laying, and incubation/hatching stages (Table 2).

Table 2: Physio-chemical parameters in different tanks.

Parameters	Broodstock rearing	Breeding-cum-egg laying	Incubation-cum-hatching
Temperature (°C)	28±2	28.5±1.5	29.5±0.5
pH	8±0.5	8.25±0.25	8.25±0.25
Dissolved Oxygen (ppm)	7±1	6.5±1.5	7.5±0.5
Total hardness (ppm)	140±10	150±15	125±6
Ammonia (ppm)	0.020±0.010	0.060±0.040	0.006±0.002
Nitrite (ppm)	0.75±0.25	0.075±0.025	0.06±0.02
Nitrate (ppm)	7.5±2.5	10±5	5±0.25

Abbreviation: Value is present as Mean ±SE.

CONCLUSIONS

The current research, titled “Hormonal Manipulation in the Breeding of Pabda, *Ompok bimaculatus* Under Captive Conditions,” aims to address this need by enabling large-scale seed production. This study pioneers a method that avoids the need for sacrificing males or stripping females, thereby allowing natural spawning to occur through hormone injections. The hormonal doses have been carefully optimized to minimize intervention while maximizing efficiency, with dosages set at 0.8 ml per kg of body weight for females and 0.4 ml per kg for males.

This optimized hormonal approach not only enhances fish production but also holds promise for sustainability by potentially reducing the impact on fish health compared to more invasive traditional methods. My findings offer valuable insights for researchers and farmers focusing on the aquaculture of Pabda fish, ensuring that the growing demand is met responsibly and efficiently.

FUTURE SCOPE

The study on “Hormonal manipulation in breeding of Pabda, *Ompok bimaculatus* under captive condition” will help another interested researcher and farmer who work in pabda, *Ompok bimaculatus*. Through this research, I aim to contribute to sustainable aquaculture practices that could help prevent the extinction of this valuable species while fulfilling the market's needs.

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