

# Breeding and Larval Rearing of Asian Catfish, *Clarias batrachus* (Linnaeus, 1758) on Live and Artificial Feed

Prem Prakash Srivastava<sup>1\*</sup>, Sudhir Raizada<sup>1</sup>, Rajesh Dayal<sup>1</sup>, Shipra Chowdhary<sup>1</sup>, Wazir Singh Lakra<sup>1,2</sup>, Akhilesh Kumar Yadav<sup>3</sup>, Priya Sharma<sup>1</sup> and Jyoti Gupta<sup>1</sup>

<sup>1</sup>National Bureau of Fish Genetic Resources, Canal Ring Road, Telibagh, Lucknow – 226 002, UP, India

<sup>2</sup>Central Institute of Fisheries Education, Panch Marg, Off Yari Road, Versova, Mumbai - 400 061, India

<sup>3</sup>Aquaculture Research and Training Unit, National Bureau of Fish Genetic Resources, Chinhat Faizabad Raod, Lucknow – 227 105, UP, India

## Abstract

A study was conducted to observe the breeding and larval rearing of Asian Catfish, *Clarias batrachus* fed with live and/or artificial feed for 21 days in an indoor hatchery. The brooders of *C. batrachus* (Av. wt of female 160 ± 10.5 g; Av. wt of male 120 ± 6.75 g) were procured from outside ponds and stocked in a pond near the experiment site 2-months prior to spawning. The fishes were successfully induced bred using ovaprim @ 1.0–2.0 ml/kg body weight (bw) to females and 0.5–1.0 ml/kg bw to males. Fertilization, hatching and survival percentages at spawn stage were respectively recorded 70.6 - 72.8, 60.7 - 55.3 and 54.3 - 56.2. After yolk-sac absorption, fry of three age groups 7, 14 and 21 days were subjected to feed trial using *Artemia nauplii* followed by laboratory made feed for 21 days. Weekly sampling indicated that higher age groups constantly maintained higher lengths and weights with highest survival in the age group of 14-days old fry and SGR in 7-days old. The quality of hatchery water was recorded for temperature 29 ± 1°C, pH 7.2 ± 0.2, DO 7.1 ± 0.3 mgL<sup>-1</sup> and total alkalinity 132 ± 4.0 mgL<sup>-1</sup> respectively.

**Keywords:** *Clarias batrachus*; Larval rearing; Feed

## Introduction

The Asian catfish, *Clarias batrachus* is one of the most popular food fish of India, Myanmar, Bangladesh, Srilanka and Malaysia [1]. The hardy nature and tolerance to adverse environmental conditions particularly low oxygen levels in water enable its intensive culture with high production rate [2]. However, the major constraint for wide spread aquaculture of the species is non availability of seed both from hatcheries and natural resources due to depletion of natural stocks. Several attempts have been made in past to induced bred *C. batrachus* using various hormones [3-5] however, the commercially available synthetic hormonal preparation Ovaprim<sup>®</sup> has been found most efficient inducing agent for the captive breeding of the species. High level survival of larvae to seed size is another challenge for the hatchery operators as no standardized protocol of feeding to larvae has so far been evolved. Therefore, the objective of the present study was to evaluate breeding and rearing of the early stages of *Clarias batrachus* on live and artificial feed.

## Materials and Methods

### Procurement of fishes

The adults of *C. batrachus* (avg. weight; female, 160 ± 10.5g, males, 120 ± 6.75g) were procured from Unnao and Mohanlal Ganj blocks of Lucknow, UP, India. Both males and females were reared in an earthen pond of size (100 m<sup>2</sup>) for two months and fed with laboratory made diet (Table 1).

### Induced breeding and rearing of larvae

After two months of rearing, the sexes were identified and fully gravid male and female fishes were used for spawning. Gravid females were identified having round and bulging abdomen and button-shape genital papilla. Males on the other hand look slender and have an elongated and pointed papilla. A sex-ratio was 8:10 male:female and 6:10 male:female for undertaking stripping operation. Both the sexes were given injection of Ovaprim (Manufacturer Syndel, Canada) at the rate of 1.0 to 2.0 ml kg<sup>-1</sup> body weight (bw) to females and 0.5 to 1.0 ml kg<sup>-1</sup> bw to males. The testis of male fishes were dissected out after a latency

period of 16-18 hours, macerated with the help of pestle and mortar after pooling all the testes from 8 and/ or 6 testes for getting milt and immediately diluted with 0.9% saline (NaCl solution). The eggs from all the females were stripped out immediately after the preparation of milt suspension in a dry tray and mixed with the milt suspension. A stan-

Ingredients	Percentage
Fish meal	60.0
Wheat flour	25.0
Soybean meal	13.0
Vitamin & Min. Mix*	02.00
<b>Composition</b>	
Protein	40.05
Carbohydrate	22.58
Fat	10.20
Ash	12.48
Fibre	04.50
Gross Energy (k J/g)	15.33

\*Vitamin and Mineral Composition (Per 100 g): Manufacturer: Sunder Chemical Pvt. Ltd., Chennai, India for Agrivet Farm Care Division of Glaxo SmithKline Pharmaceuticals Ltd (Batch No. SC7450).

Vitamin A, 70000 IU; D3, 7000 IU; E, 25 mg; Nicotinamide, 100 mg; Cobalt, 15 mg; Copper, 120 mg; Iodine, 32.5 mg; Iron, 150 mg; Magnesium, 600 mg; Manganese, 150 mg; Potassium, 10 mg; Selenium, 1 mg; Sodium, 0.59 mg; Sulphur, 0.72%; Zinc, 960 mg; Calcium, 25.50%, Phosphorus 12.75%)

**Table 1:** Feed composition used for feeding to brooders of *Clarias batrachus*

\*Corresponding author: : Prem Prakash Srivastava, National Bureau of Fish Genetic Resources, Canal Ring Road, Telibagh, Lucknow – 226 002, UP, India, E-mail: [ppsrivastava63@gmail.com](mailto:ppsrivastava63@gmail.com)

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lard method of dry stripping was followed subsequently. The fertilized eggs were incubated in plastic round sieve trays kept in a tank having provision of flow-through system of water (0.2-0.5 l/min). The eggs hatched out within 24-28 hours and yolk sac was absorbed in 4 days. The larvae were then stocked in the plastic pools (capacity 300 L) and given live *Artemia* as feed for subsequent feed trials. The experiment was carried out in three replicates.

### Age dependent larval rearing trials

Five hundred larvae of age groups 7, 14 and 21 days old were procured from the plastic pools used for rearing test fishes and reared for 21 days in the plastic pools (capacity 300 L) in a completely randomized triplicate design. The length and weight of 25 larvae was measured by a measuring scale and electronic balance prior to stocking and after every 7-days of rearing. The larvae start to take up food after 4<sup>th</sup> day. The larvae were fed exclusively *Artemia nauplii* for 7 days followed both with *artemia* and formulated diet to satiation in all the treatments till end of the experiment. Rearing pools were cleaned every second day and about half of the water was replaced with fresh bore-well water to reduce the nitrogenous waste accumulated. At the end of twenty-one days, all surviving larvae were collected and counted for each pool. The final total length and weight of twenty five larvae were recorded and the total biomass was also recorded for the individual pools. The percent

survival and Specific Growth Rate (SGR) were calculated as: Survival (%) = (Number of larvae stocked – Number of dead larvae) / Number of larvae stocked × 100; Specific growth rate = (ln final weight – ln initial weight) / Days of experiment × 100.

### Procurement of *Artemia nauplii* and preparation of larval feed

The *Artemia* cysts were hatched in salt water (salinity 20 ppt) in the indoor hatchery and freshly hatched *Artemia nauplii* were fed in uniform quantity to the larvae in all the tanks three times a day. The formulated diet was prepared by mixing ingredients listed in Table 3 in dough form, cooked in a pressure cooker and after cooling hand-grated to fine particles (size 150-200 μ) and given to satiation level observing the left over feed in all the tanks.

### Physico-chemical parameters of water

The water quality indices of all the experimental pools for temperature, pH, DO and total alkalinity were monitored on every alternate day during the feeding trial following the standard methods [6].

### Statistical analysis

Statistical analysis of the data for all experiments were done by one-way ANOVA (Analysis of Variance) and DNMRT (Duncan's New Mul-

	Dose to female (ml/kg Bw)	Dose to male (ml/kg bw)	No. of Male Sacrificed (n)	No. of Female (n)	Male Avg. Weight (g)	Female Avg. Weight (g)	Latency period (hr)	Total number of Eggs stripped out	Fertilized eggs	Fertilization (%)	Number of Hatchlings	Hatching(%)	Survival %
1	1.0	0.5	8	10	120 ± 6.75	160 ± 10.5	16a ± 1	4265 <sup>a</sup> ± 36	3615 <sup>a</sup> ± 22	72.8* ± 1.3	2515 <sup>c</sup> ± 110	60.7 <sup>a</sup>	56.0 <sup>a</sup> ± 2.56
2	2.0	1.0	6	10			18b ± 2	4354 <sup>b</sup> ± 23	3716 <sup>b</sup> ± 69	70.6* ± 1.5	2673 <sup>d</sup> ± 122	55.3 <sup>b</sup>	54.0 <sup>a</sup> ± 3.17

\* Same Superscript denotes no significant difference between them; Different superscript denotes significant difference between them (between a, b and \* & \*\*: p < 0.05; between c, d: p > 0.01). The data are based on the means (± SE) of pools in each group.

Table 2: Reproductive performance of *Clarias batrachus* induced bred with Ovaprim

Ingredients	Percentage
Hen's egg with yolk	24.3
Lactogen powder	35.7
Fishmeal powder	36.0
Vitamin & Min. Mix*	04.00
<b>Composition</b>	
Protein	48.97
Carbohydrate	21.06
Fat	09.43
Ash	5.72
Fibre	nil
Gross Energy (kJ/g)	16.46

\*Vitamin and Mineral Composition (Per 100 g): Manufacturer: Sunder Chemical Pvt. Ltd., Chennai, India for Agrivet Farm Care Division of Glaxo SmithKline Pharmaceuticals Ltd (Batch No. SC7450)

Vitamin A, 70000 IU; D<sub>3</sub>, 7000 IU; E, 25mg; Nicotinamide, 100 mg; Cobalt, 15 mg; Copper, 120 mg; Iodine, 32.5 mg; Iron, 150 mg; Magnesium, 600 mg; Manganese, 150 mg; Potassium, 10 mg; Selenium, 1 mg; Sodium, 0.59 mg; Sulphur, 0.72%; Zinc, 960 mg; Calcium, 25.50%, Phosphorus 12.75%)

Table 3: Feed composition used for feeding to fry of *Clarias batrachus*.

Age (days)	Initial length (mm)	Initial weight (mg)	Final length (mm)	Final weight (mg)	Specific growth rate (SGR)	Survival (%)
	8.34 <sup>c</sup> ± 0.11	6.47 <sup>c</sup> ± 0.12	24.16 <sup>b</sup> ± 0.1	188.02 <sup>b</sup> ± 12.09	8.56 <sup>a</sup> ± 0.80	14.06 <sup>c</sup> ± 1.02
14	14.16 <sup>a</sup> ± 0.32	21.3 <sup>a</sup> ± 1.01	36.37 <sup>a</sup> ± 2.0	379.04 <sup>a</sup> ± 42.11	17.38 <sup>b</sup> ± 2.23	30.21 <sup>b</sup> ± 1.42
21	18.00 <sup>a</sup> ± 0.23	29.7 <sup>a</sup> ± 1.43	33.6 <sup>a</sup> ± 0.08	290.36 <sup>c</sup> ± 11.18	12.96 <sup>c</sup> ± 0.45	55.00 <sup>a</sup> ± 2.76

The data are based on the means (± SE) of 3 replicate pools in each group.

<sup>a, b, c</sup> Mean values bearing different superscripts in a column differ significantly (P < 0.05).

Table 4: Rearing performance of different age groups of *C. batrachus* larvae and changes in total length, weight, SGR and survival rate during three weeks period

tiple Range Test) to determine differences between the means taking at 1 and 5 percent significance levels using SPSS version 16.0. [7].

## Results

### Breeding performance

Table 2 indicated that both low (1.0 ml Kg<sup>-1</sup>) and high doses (2.0 ml Kg<sup>-1</sup>) of Ovaprim to female fishes were effective in the maturation of ova to complete ripeness. Freshly fertilized eggs were hazy brown in colour. The number of eggs were found significantly higher ( $p > 0.05$ ) with higher dose (2.0 ml Kg<sup>-1</sup>) of Ovaprim. On the contrary, the percentage of fertilization and survival of fry after yolk-sac absorption were insignificant ( $p < 0.05$ ) both with low and high doses of Ovaprim. The percentage of fertilization was found  $72.6 \pm 1.3$  &  $70.6 \pm 1.5$ , hatching  $60.7$  &  $55.3$  and survival of fry after yolk-sac absorption was found  $56 \pm 2.56$  &  $54 \pm 3.17$  in case of respective low and high doses of Ovaprim. The quality of hatchery water was found suitable with respect to temperature, pH, total alkalinity and dissolved oxygen levels which were recorded in the range of  $28 \pm 2^\circ\text{C}$ ,  $6.9 - 7.6$ ,  $128 - 136 \text{ mg L}^{-1}$  and  $6.8 - 7.4 \text{ mg L}^{-1}$ , respectively during the entire rearing period.

### Age dependent growth study for 21 days

The age dependent rearing performance of *C. batrachus* larvae has been presented in Table 4. The trend of initial difference ( $P < 0.05$ ) in total length between three age groups of fry at the time of stocking was maintained till the end of the experiment. The weekly samplings indicated that the higher age groups (14 and 21 days) constantly maintained the trend of superior length increment than that of 7-day old larvae. Larvae maintained significant variations in weight between the three age groups at the start also maintained significant variation in weight in age groups of 14-days and 21-days larvae though final weight was obtained by 14-day larvae. SGR also showed significant ( $P < 0.05$ ) differences in the three age groups with highest in 14-day larvae followed by 21-day and 7-day respectively. The rate of survival was found significantly ( $p < 0.05$ ) higher in higher age group fry with highest in 21-day larvae followed by 14-day and 7-day larvae.

### Physico-chemical parameters of larval rearing tank water

The physico-chemical parameters of larval rearing tank water for temperature, pH, total alkalinity and dissolved oxygen were found in the range of  $28 \pm 2^\circ\text{C}$ ,  $6.9 - 7.6$ ,  $128 - 136 \text{ mg L}^{-1}$  and  $6.8 - 7.4 \text{ mg L}^{-1}$ , respectively during the entire rearing period.

## Discussion

Ovaprim has been successfully employed for induced spawning of fishes in a number of commercially important food as well as ornamental and threatened species [8-15]. The product has been reported to be an efficient inducing agent for oocyte maturation and ovulation in *C. batrachus*. The latency period of 16-18 hours after the injection of Ovaprim (dose 1-2 ml Kg<sup>-1</sup> bw to female and 0.5-1.0 ml Kg<sup>-1</sup> bw to male) to subjected fishes was found suitable for the ovulation of this species. Similar findings were also reported by Sahoo et al. [16] in the same species while using sGnRH $\alpha$  in combination with domperidone (14 to 23 hours). However according to Sahoo et al. [16], the suitable latency period for final maturation of ova is also dose dependent when using sGnRH $\alpha$  and domperidone combinations on spawning performances and deduced that latency period of 14-17 hours and dose of 20  $\mu\text{g}$  sGnRH $\alpha$  along with 10 mg domperidone and 30  $\mu\text{g}$  of sGnRH $\alpha$  & 15 mg domperidone per kg of female was found to be suitable for best

spawning and larval production. Thus our findings are in connivance to the reporting of Sahoo et al. [16] in case of *C. batrachus*.

A suitable feed is the basic requirement for the growth and survival of fish larvae. Though most of the fish larvae relish best on planktonic fauna in the young age, they need nutritionally balanced food in bulk quantity at later stages of their life. This was well demonstrated in the present findings where the larvae of 21-day showed highest survival (55.00%) and moderate SGR (12.96) in comparison to 14-day (30.21%, 17.38) and 7-day (14.06%, 8.56) larvae. The reason being that the 7-day larvae being small in size needed exclusive or more of plankton (*Artemia nauplii*) in comparison to formulated diet which was not available to the satiation level in the larval rearing tanks. On the contrary, the 14-day and 21-day larvae which grow to larger size initially in the stock tanks were able to consume both *Artemia nauplii* and bulk feed in the experimental tanks and hence showed better growth and survival. Mollah et al. [5] & Mollah and Nurullah [17] have also reported more or less similar findings when they recorded a survival of 97.6 -99.6% while feeding tubifid worms to the fry (size 88.1 to 125.2 mg) of *C. batrachus*. Thakur [18] mentioned that *Clarias batrachus* fry attained 3-7 cm size within 20-25 days with 40% survival that also support our findings. Thus the present study demonstrated that the fry of *C. batrachus* may be fed with planktonic fauna up to 14-days and thereafter supplementation with formulated diet can greatly enhance growth as well as survival of larvae.

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