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RESEARCH ARTICLE

# Captive Spawning of the Striped Murrel, *Channa striatus* (Bloch) Using sGnRH, in Gangetic Plains of India

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**Abstract** Captive breeding of striped murrel, *Channa striatus* was carried out by synthetic salmon gonadotropin releasing hormone (sGnRH), ovaprim in three sets of experiments to observe its breeding performance. In the first experiment (A), the fish was induced with ovaprim and kept in FRP tanks in the indoor conditions. In the second experiment, equal number of fishes after ovaprim injection were released in a pond (B-1) as well as kept in a hapa installed (B-2) in the same pond. In third system (C), the fishes, after injecting ovaprim, were released in the pond for mass production of seed. The fishes kept in the FRP tank (A) and hapa (B-2) did not breed even after 15 days, whereas, 50 and 62.5 % of the fishes released in the pond in B-1 and C experiments spawned naturally. The fertilization percentage in B-1 and C ranged between 76–81 % and 85–91 % whereas, hatching was recorded between 82–90 % and 88–92 % respectively. The larvae were reared both on live feed and artificial diets for 30 days and showed a survival of 48–56 % and 52–60 % respectively in case of B-1 and C experiments. The present mode of breeding this species in pond conditions and collection of eggs and rearing in indoor condition could provide a suitable platform for raising the seeds of this valuable prioritized fish for aquaculture and for conservation.

**Keywords** Striped murrel · *Channa striatus* · Induced spawning · sGnRH

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## Introduction

Striped murrel, *Channa striatus*, is a highly priced air-breathing freshwater fish of the Indian sub-continent and south-east Asia. They are in great demand as food fish due to their appealing flavour [1], few muscular spines, medicinal importance [2–6] and air-breathing nature [4] that facilitate high density culture and easy transport in live condition to the markets. Murrel adapts well to hypoxic water-bodies and hence can be cultured in high stocking densities. The striped murrel (*C. striatus*) culture is widely popular in Thailand and on limited scale in India, Philippines and Taiwan [4, 7–9] due to non-availability of seed.

Striped murrel breeds in ponds and rivers a little prior to or with the onset of monsoons [7, 10, 12] and their spawning season extends throughout the monsoons [13]. It is a batch spawner and breeds two to three times in a season, the breeding season extends from February–March to October–November and lay small number of floating egg at a time in weed infested marginal areas [13]. Both parents guard the eggs and fry [11].

Parmeshwaran and Murugesan [14] attempted captive breeding of striped murrel in India when they injected carp pituitary glands to both male and female with varying success. Marimuthu et al. [15, 16]. successfully carried out

spawning of striped murrel in a net enclosure fixed in a pond and in cement tanks using Ovatide (sGnRH) at a dose rate of 0.5 ml $kg^{-1}$ and 0.4–0.6 ml $kg^{-1}$ body weight of female but he did not give details of breeding success. The aim of the present study was to use ovaprim (sGnRH and Domperidone) as an inducing agent for testing the breeding performance of striped murrel in different type of systems and to assess its efficiency concerning latency period, spawning response, fertilisation rate, incubation period and hatching time. Since, larval rearing is an integral part of seed production, the performance of larvae on live and formulated diet (FD) were also evaluated in the present study.

## Material and Methods

### Procurement of Test Fish

Wild striped murrel, *C. striatus* (Avg. weight 388–407 g) were collected from different sites of Unnao and Barabanki districts of Uttar Pradesh, India and were stocked in ponds at NBFGR farm facility 3 months prior to spawning experiments. They were fed with laboratory made feed ad libitum (Table 1).

### Feed Preparation and Feeding

The feed was formulated using goat intestine, wheat flour, soybean meal, vitamin and mineral mixture, mixed in a ratio of 33:12:4:1 w/w with a gross energy of 3537.8 kcal/kg (Table 1). The feed was distributed daily once in the pond at fixed points ad libitum.

**Table 1** Feed composition of larval feed acclimatization of brooders of *Channa striatus*

| Ingredients | Percentage | Approximate ratio (w/w) |
|---|---|---|
| Goat intestine | 66.0 | 33 |
| Wheat flour | 24.0 | 12 |
| Soybean meal | 8.0 | 4 |
| Vitamin and mineral mix[a] | 2.0 | 1 |
| Composition | | |
| Protein | 39.01 | |
| Carbohydrate | 23.22 | |
| Fat | 12.10 | |
| Ash | 11.23 | |
| Fibre | 4.55 | |
| Gross energy (kcal/100 g) | 353.78 | |

[a] vitamin and mineral composition (per 100 g): vitamin A 70,000 IU, vitamin $D_3$ 7,000 IU, vitamin 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 96 mg, calcium (%) 25.50, phosphorus (%) 12.5. From Agrivet Farm Care Division, GlaxoSmithKline Pharmaceuticals Limited (Mfg. by Sunder chemicals Pvt. Ltd., Chennai). Lot/Batch No. SC7450 July, 2006

### Experimental Design

At the end of 3 months of acclimation the brood fishes were collected from the pond and used for induced breeding experiments. The fishes were kept for induced breeding in three sets of experiments. In Experiment A, 4 females (total weight 1.2 kg) along with 6 males were kept in a FRP tanks of 1,200 l (size 1.8 × 1.0 × 0.7 m). The tank was provided with aeration and commonly found macrophytes, *Eichhornia crassipes* and *Hydrilla verticellata* as they provide shelter and nesting in natural conditions. In Experiment B, two sets of breeding arrangements were made. In the first set of arrangement (Experiment B-1a), 4 females (total weight 1.3 kg) along with 6 males (total weight 1.7 kg) were kept in a hapa (size 3 × 3 × 1.5 m) fitted in an earthen pond and was provided with a cover of *Eichhornia crassipes* and *Hydrilla verticellata*. In the second arrangement (Experiment B-2), 4 females (total weight 1.5 kg) along with 6 males (total weight 2.0 kg) were kept in the same earthen pond (size 9.1 × 6 × 1.3 m) that was fitted with the above hapa. The pond was also provided with sufficient cover of macrophytes. In experiment C, 8 females (total weight 2.7 kg) and 12 male fishes (total weight 4.0 kg) were kept in an earthen pond for mass breeding. This pond was also provided with the cover of macrophytes.

### Induced Breeding

Both male and female fishes were injected (intramuscular) with sGnRH, ovaprim (Syndel, Canada) at the rate of 0.5–0.8 ml $kg^{-1}$ body wt in case of female fishes and 0.2–0.4 ml $kg^{-1}$ body wt in male fishes respectively during June 2010 and 2011. The hormone injected fishes were released respectively in different systems as stated above for spawning and observed daily for breeding behaviour and parental care.

### Collection of Egg Samples

The fishes were observed regularly for breeding till 15 days. As this fish forms nest on the margin of the pond amongst macrophytes for laying eggs and observe parental care, such areas were carefully observed for egg mass collection. Whenever eggs were found in the nest, approximately 500 eggs were collected with the help of a hand net and kept in a plastic trough for recording the fertilization and hatching rates. The size of the egg and newly hatched larvae was recorded with the help of a microscope.

### Rearing of Egg Samples and Larvae

The eggs kept in the plastic troughs were allowed to hatch in the bore-well water. The troughs water was continuously aerated with an aquarium pump. The time of first and last hatching was recorded. The size of just hatched larvae along with size of yolk-sac was measured with the help of a microscope. The larvae were reared in troughs till yolk-sac is completely absorbed in 4–5 days. Survival was recorded when yolk-sac of all the larvae were completed absorbed.

### Rearing of Larvae

The yolk-sac absorbed larvae were reared in plastic pool of 300 l (diameter 80 cm and height 60 cm). Plastic pools were filled with 100 l pre-settled bore-well water and stocked with 100 larvae in duplicate set from each batch of spawning. Each tank was provided continuous aeration from a portable blower. The larvae were fed initially for 5 days with freshly hatched brine shrimp nauplii (BSN) followed by BSN and FD with gradual reduction in BSN and from tenth day onwards feed exclusively on FD (Table 2). The proximate composition of feed was estimated by standard laboratory methods following AOAC [25]. Feed was given twice daily in the morning and evening ad libitum. FD was grounded to small particle size (50–100 $\mu$) by passing through a fine meshed sieve so that larvae could easily feed on them. Around 50 % of the tank water was changed every day at the time of siphoning of the debris. The survival of the larvae was counted after 30 days of rearing.

### Water Quality

The water quality parameters were recorded for water temp., pH, dissolved oxygen, total alkalinity and total dissolved solids (TDS) following standard methods [17]. The water quality maintained during the experiment period were temp., 26 ± 2 °C, pH 7.6 ± 0.2, alkalinity 136 ± 6.0 mg $l^{-1}$, EC 450 ± 48 µm h $sec^{-1}$, TDS 225 ± 18 mg $l^{-1}$ and DO 7.8 ± 0.2 mg $l^{-1}$.

## Results

### Induced Spawning

Breeding did not occur in Exp. A and Exp. B-2 even after keeping the brooders for 15 days in the same system. However, 50 and 62.5 % (Table 3) of the females spawned in Exp B-1 and Exp-C and formed nest at pond margins amongst *Eichhornia* and *Hydrilla* weeds. The breeding pairs were observed from courtship behaviour 1–2 days in advance till spawning was completed with roaming, nudging and splashing water together. However, the breeding behaviour at the actual time of spawning could not be observed as the fish spawned during night all the time. The floating egg mass was observed at the time of examination of pond margins. The first breeding was observed on day-2 of giving injection and second on day-4 in Exp B-1. In case of Exp-C, the first breeding was observed on day-2 (2 fishes spawned), second on day-5, third on day-8 and fourth on day-10.

**Table 2** Feed composition of larval feed of *Channa striatus*

| Ingredients | Percentage | Approximate ratio (w/w) |
|---|---|---|
| Hen's egg with yolk | 25.0 | 5 |
| Lactogen powder | 35.0 | 7 |
| Fishmeal powder | 35.0 | 7 |
| Vitamin and mineral mix[a] | 5.0 | 1 |
| Composition | | |
| Protein | 48.34 | |
| Carbohydrate | 20.18 | |
| Fat | 9.76 | |
| Ash | 5.45 | |
| Fibre | Nil | |
| Gross energy (kcal/100 g) | 388.27 | |

[a] vitamin and mineral composition (per 100 g): vitamin A 70,000 IU, vitamin $D_3$ 7,000 IU, vitamin 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 96 mg, calcium (%) 25.50, phosphorus (%) 12.5. From Agrivet Farm Care Division, Glaxo-SmithKline Pharmaceuticals Limited (Mfg. by Sunder chemicals Pvt. Ltd., Chennai). Lot/Batch No. SC7450 July, 2006

### Egg Collection and Incubation

The eggs were found near the pond corners at the places which were encircled by weeds. They could be easily located by observing the guarding parents mostly male fish who happens to roam all around the egg mass most of the time in a typical manner. The eggs of one female were found only in one egg mass. They were non-adhesive, floating, pale yellow in colour with a diameter of 1.8 ± 0.3 mm. When eggs were kept in the plastic troughs for hatching, they assembled on the margins of the trough. The rate of fertilization was recorded 76–81 % (Table 3) in case of Exp. B-1 and Exp. C respectively. In indoor conditions, the eggs hatched out in 24 ± 2 h in all the cases at a temperature of 26 ± 2 °C. The hatching rate was found between 82 and 90 % in Exp. B-1 and 88–92 % in Exp. C (Table 3). The newly hatched larvae had lengths of 3.10 ± 0.4 mm. The yolk-sac was absorbed within 4–5 days.

**Table 3** Breeding performances in different set of conditions in *Channa striatus*

| Exp. no. | Total wt of female in kg (nos.) | Dose of ovaprim (ml/kg) | Total wt of male in kg (nos.) | Dose of ovaprim (ml/kg) | Breeding result | Fertilization (%) | Hatching (%) | Survival at fry stage (%) |
|---|---|---|---|---|---|---|---|---|
| A | 1.2 (4) | 0.5–0.6 | 1.6 (6) | 0.2–0.3 | No breeding | – | – | – |
| B-1 | 1.5 (4) | 0.5–0.6 | 2.0 (6) | 0.2–0.3 | 2 females spawned | 76–81 | 82–90 | 48–56 |
| B-2 | 1.3 (4) | 0.5–0.6 | 1.7 (6) | 0.2–0.3 | No breeding | – | – | – |
| C | 2.7 (8) | 0.5–0.8 | 4.0 (12) | 0.2–0.4 | 5 females spawned | 85–91 | 88–92 | 52–60 |

Larval Rearing

The larvae initially showed high degree of schooling behavior which gradually reduces as they become older. They were found easily consuming both BSN and FD (Feed-2) immediately after their application. The survival percentage was 48–56 % and 52–60 % in Exp. B1 and Exp-C respectively after 30 days of rearing.

## Discussion

Only a few workers have attempted captive breeding of striped murrels with varying success. Alikunhi [11] and Parameshwaran and Murugesan [14] attempted breeding of this species by hypophysation using pituitary gland. Haniffa et al. [18], Selvaraj and Francis [19] induced bred this fish using sGnRH and HCG respectively. Haniffa et al. [20] also made a comparative study on induced breeding this fish using pituitary gland, HCG, LHRH-a + Pimozide and ovaprim. Marimuthu et al. [15] used Ovatide (sGnRH anologue) for breeding in hapa fitted inside the pond. Presently, a comparative study was made on breeding systems using ovaprim as an agent for induced breeding. No breeding in FRP tank and hapa indicated that this fish does not breed in small container/enclosures, perhaps may be due to formation of nest. However this observation is contrary to Marimuthu et al. [15], who reported breeding of this species in hapa without mentioning the breeding success. The breeding success of 50 % in Exp. B-1 and 62.5 % in Exp. C obtained with ovaprim (sGnRH anologue) with doses of 0.5–0.8 ml $kg^{-1}$ in the present case in pond condition revealed that the ideal natural condition for fish breeding is in pond as it needs to construct nest for laying eggs and the later was observed in every breeding. Haniffa et al. [20] also made similar observation with ovaprim in pond conditions. Since the breeding occurred on different days ranging from 2nd to 11th days in these experiments, it is likely that all the fishes selected for spawning might not be in the right stage of final maturity and those close to the final stage of maturity were triggered with introduction of ovaprim and took comparatively longer time for final maturity and that is why the breeding occurred till 11th day after giving ovaprim injection. This way, ovaprim not only induces immediate spawning of fish as occurred in carps but it may also trigger maturity advancement in this species which is a batch spawner.

Striped murrel makes nests for spawning and lay non-adhesive and floating eggs amongst the floating and submerged weeds. The combination of *Eichhornia crassipes* and *Hydrilla verticellata* were found to be the suitable combination of aquatic weeds for making nest by this fish and for providing shelter to the larvae. Marimuthu et al. [15] have also used similar weeds for the natural spawning of striped murrel. Parameshwaran and Murugesan [14], however, used *Pistia* as floating weeds in combination with *Hydrilla verticellata* and Haniffa et al. [20] used *Eichhornia crassipes* alone for spawning of striped murrel.

Collection of eggs of striped murrel is comparatively very easy as they are non-adhesive, free floating and remain in the shape of egg mass at only one place amongst the free floating weeds. Further, they can be identified by observing the rhythmic movements of parents near the nesting place more particularly of male fish which guards the eggs more dominantly. Only problem with collection of eggs is daily careful observance all around the pond particularly on the margins where this species generally spawn. Since the size of eggs is very small and they are pale yellow in colour, they are not visible from a distance. A good binocular may help to watch the eggs from a distance. Once the eggs are located, they can be collected easily with the help of a scoop net of fine mesh cloth as done in the present study.

The fertilization rate was found high in both the experiments (76–81 %) in the present study which was slightly less than that observed by Haniffa et al. [20] in ovaprim injected fishes. Whereas, Pati et al. [21] reported a higher hatching percentage of 92–98 % when WOVA FH was used for induced breeding. Parameshwaran and Murugesan [14], however, mentioned a great difference in the rate of hatching from 28 to 100 % with pituitary gland which was not used in the present case. The rate of fertilization was higher in this fish as it spawns in small batches where chances of mixing of gametes are greater. In the present study hatching took place within 24 ± 2 h at a temperature of 26 ± 2 °C in all the cases which was quite similar to the

recording of Haniffa et al. [20] who obtained this in the range of 21–23 h when he used ovaprim but he did not mention the temperature of media. It is to emphasize that this group of workers observed higher incubation period (36–43 h) when this species was administered pituitary gland, HCG, LHRHa + Pimozide for induced breeding. Banerji [22] reported incubation period of Spotted murrel, *C. punctatus* to be 24 h at temperature 28 °C that also matches with the present study.

The hatching rate of 82–90 and 88–92 % in B-1 and C experiments and survival between 48 to 56 and 52 to 60 % respectively in Exp. B-1 and Exp-C were quite higher and comparable with that obtained by Haniffa et al. [20] for hatching when ovaprim was used for induced breeding. The larval length of yolk-sac absorbed larvae was measured as 3.10 ± 0.4 mm which was more of less similar to the recordings of Parameshwaran and Kamal [23] who recorded it in the range of 2.81–3.22 mm. The combination of both live feed (BSN) and formulated feed strategies with survivals of 48–56 and 52–60 % in Exp-B1 and Exp-C were also found highly encouraging.

The present study shows that captive breeding of *C. striatus* can be taken up on breeding in pond conditions. Such ponds should be small in size where a few numbers of brooders could be induced bred at a time and eggs can be located easily. The eggs could be easily collected as stated above and reared in indoor conditions in plastic troughs, plastic pools and FRP tanks. The yolk-sac larvae can be easily fed on BSN and supplementary diets such as used in the present study for rearing up to fry size. Qin and Fast [24] also reported that in nursery culture of snakehead, *Artemia* nauplii could be used as starter feed in indoor tank culture and formulated feed should be introduced when fish reaches 12 mm in length.

## Conclusion

Thus, it is recommended that the seed of *C. striatus* could be produced in captivity. Synthetic sGnRH is not only precipitating immediate spawning in fully gravid females but also help in enhancement of maturity in fishes which could make delayed spawning in the pond conditions. Rearing of eggs and hatchling is very simple under indoor conditions; however, a great care is required in rearing yolk-sac larvae to fry. Since the breeding protocol does not require higher inputs in this type of breeding arrangements, it can be taken up by small farmers for seed production.

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