



Optimisation of Hormone Dosage for Breeding of *Bangana devdevi*

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ABSTRACT

Bangana devdevi (Hora) is an indigenous medium-sized and benthopelagic minor carp, widely distributed in the Chindwin headwaters of Manipur, Northeast India. Induced breeding of *B. devdevi* was done using Gonopro-FH in different doses @ 0.4ml, 0.5ml and 0.6ml/kg body weight in a set of three experimental groups T1, T2 and T3, respectively. All the males were given half the doses of females. Spawning commenced 6–10 hr after injection and was completed within 4–5 hr. Fertilized eggs were hatched out after 11–16 hr of fertilization at temperatures of 26.4–27.5°C. Statistical analysis was carried out to determine the relation between the hormone dosage with egg output, fertilisation rate and hatching rate. The highest number of fertilisation (94.25%) and hatching rate (89.03%) were found in fish with Gonopro-FH @ 0.5ml/kg body weight female and significantly higher ($P < 0.05$) than T1 and T3. The present study may be beneficial for species conservation and management strategies in rivers and tributaries as well as mass seed production of *B. devdevi*.

Key Words: Induced reproduction, Gonopro-FH, Sexual maturity, Spawning.

INTRODUCTION

Bangana devdevi, Hora, 1936 (formerly *Labeo devdevi*) is commonly known as ngaton or khabak (in Manipuri) depending on their size where fingerlings are known as ngaton and advanced fingerlings/matured fish as khabak. It is one of the most popular indigenous minor carps in the north eastern states of India particularly in Manipur. It is distributed in Myanmar and Thailand (Talwar and Jhingran, 1991). The main characteristic feature of this species is the deep transverse groove across top of snout. The body is oblong and compressed in adults. Its flesh is well flavoured and highly esteemed as food in north-eastern region of India. It reaches a length of 30 cm and usually caught during post monsoon season with cast net and dip net from rivers. It is categorised as least concern according to CITES (2013). This species feeds on insect larvae, algae, zooplankton

and detritus. Sexual dimorphism is prominent during breeding season. Male have rough pectoral fin whereas the dorsal side of pectoral fin is smooth in female. The female genital aperture is reddish and the belly is swollen and soft. In case of male milt oozes out when gentle pressure is applied on belly. The maturity of the fish attains in 2+ yrs and male mature earlier than females.

B. devdevi is an economically important as there is high demand of the fish in the local market of Manipur. It reaches almost triple the prices of that of Indian Major Carps (IMC). A little study has been done about the reproductive biology and there is no study on breeding of *B. devdevi* till today. Since no proper breeding protocol has been established for this species, there is a need of developing induced breeding protocol for the production of seeds to increase the population and also to meet its increasing demand in the market as

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this fish does not breed naturally in captivity. Since this fish is a new potential candidate species for aquaculture, there is no report on induced breeding protocol for this species. The present research aims to standardise the optimum effective dosage of a synthetic hormone, Gonopro-FH for induced breeding of *B.devdevi* to find out the effectiveness of spawning, fertilization and hatchability of the fish in captivity.

MATERIALS AND METHODS

Collection of brood fish for induced breeding

100 numbers of individual fish were collected from Iril river, Thoubal river and Imphal river. The fish were acclimatized at Central farm pond, Central Agricultural University, Imphal located at Lamphelpat, Imphal West where the experiment was conducted. The fish were collected and transported in well aerated Hundi. The fish were reared in brooder pond and were fed with a mixture of rice bran and mustard oil cake at the ratio 1:1 @ 3% body weight per day. 40 males with average length of 17cm and 20 females of average length 18cm were kept for around 1 month in separate tanks before the induced breeding start. Sexual dimorphism of *B. devdevi* is distinct during breeding season, the pectoral fin of male will be rough when touch whereas for female will be smooth. When the fish is over 2 yr, they will be matured enough for breeding however males mature earlier than females.

Experimental design

The breeding experiment was conducted during the month of June 2018. The brooders were collected from the brooder pond by netting and transferred into FRP tanks for acclimatization for 5–6 hours before giving injection with hormone Gonopro-FH marketed by APC Nutrients Pvt. Ltd. Each ml of Gonopro-FH contain synthetic gonadotropin releasing hormone analogue (SALMO-Gn-RhA – 20mcg viz., in 0.1 mL 2mcg of SALMO-Gn-RhA) is present. Five different experimental group viz. T1, T2, T3 and Control were given four different doses of hormone Gonopro-FH.

Selection of brood fish

12 fish (4 females and 8 males) were put in each experimental group in the ratio of 1:2 (female:male) and each group have 4 replicates. Milt oozing males and fully matured female having 65±2g were used for this experiment. The matured females were distended abdomen and readily oozed eggs when pressed.

Hormone injection

The males and females in each group were injected according to their required doses of hormone using 1ml graduated syringe intramuscularly at an angle of 45° between dorsal fin and lateral line. The fish were divided into five treatment groups. Three groups were injected with 0.4, 0.5, 0.6ml Gonopro-FH per Kg body weight of fish. Another group was injected with CPE (Carp Pituitary Extract) collected from silver carp. The acetone dried pituitary gland (PGs) of locally available silver carp were placed on blotting paper to take out excess acetone followed by weighing to pool the desired amount of PGs. The PGs were ground using a tissue homogeniser and diluted with desired amount of distilled water. The females were injected with an initial dose of 2mg/Kg body weight of fish and after 6hr final dose was injected with 4mg/Kg body weight of fish as a positive control. The male was given a single dose of 4mg/Kg body weight at the time of final injection. The control group was not given any injection. Immediately after injecting the hormone, the brooders were randomly distributed into sixteen hapa installed in the farm pond. Courtship behaviour of fish began to show after 10hr of injection and partial spawning started to occur within 1-2 hr. The fecundity of each female was determined by random sampling of eggs in a 10ml graduated measuring cylinder from the total eggs released by the female. The number of total eggs in 1ml were counted and multiplied with total volume of eggs released. The fertilization rate of egg was determined by randomly taking sample of approximately 100 eggs in a glass petridis. Fertilized eggs were having intact nucleus inside the clear egg cells. After spawning the spent

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Table I. Induced breeding of *Bangana devdevi* injected with Gonopro-FH, carp pituitary extract (CPE) as positive control and negative control without any injection.

Experimental group	Hormone dose for female (ml/Kg)	Hormone dose for male (ml/Kg)	Latency period (hr)	No. of egg released (,000)	Fertilization (%)	Hatching (%)
T1	0.4	0.2	10	10.400±0.13 ^a	90±0.40 ^a	78.93±0.26 ^a
T2	0.5	0.25	8	12.325±0.05 ^b	94.25±0.63 ^b	89.03±0.14 ^b
T3	0.6	0.3	6	12.800±0.04 ^c	86.75±0.63 ^c	80.35±0.12 ^c
T4 (CPE)	1 st dose	2mg/Kg BW	--	12.200±0.09 ^b	93.75±0.85 ^b	88.88±0.09 ^b
	2 nd dose	4mg/Kg BW	4mg/Kg BW			
Control	0		0	0	0	0

Different superscripts indicate significant differences ($p < 0.05$).

fish were removed from each respective breeding hapas. The fish were observed every hour after 5hr of injection to check spawning.

Calculation

Fertilization rate (%) = [(Number of fertilized eggs)/(total number of eggs counted)] × 100

Hatching rate (%) = [(Number of eggs hatched)/(total number of eggs in the batch)] × 100

Statistical Analysis

Statistical analysis was worked out by using SPSS version 16.0 for Windows. One-Way ANOVA was used to analyze the variance to determine the relation between the hormone dosage with different parameters like fertilization rate, egg output and hatching rate.

RESULTS AND DISCUSSION

The results of optimisation of hormone for the induced breeding of *B. devdevi* using different three doses of Gonopro-FH is shown in Table I. The highest egg output was found in T3 however the fertilization rate and hatching rate were found to be lowest among the three doses. In T2 the egg output was slightly lower than the T3 but the

fertilisation rate and hatching rate was significantly higher than the T3. Among all the three doses T1 produced the lowest egg output, fertilisation rate and hatching rate. However, there were no significant difference in egg output, fertilization and hatching rates in T2 and T4 at ($P < 0.05$). The control group doesn't produced any egg as they were not given any hormone. Spawning commenced 6–10 hr after injection and was completed within 4–5 hr. The fertilized eggs were bluish white in colour, demersal and translucent. Unfertilized eggs were paler and opaque. Fertilized eggs were hatched out after 11–16 hr of fertilization at temperatures of 26.4–27.5°C. The hatchlings were transparent and measured 3.20–3.80 mm of total length with a large oval head, a well defined yolk sac and a short tail.

Induced breeding is a technique where organism is stimulated by particular hormone or other synthetic hormone or by providing condition, introduced to breed in captive condition. The stimulation promotes timely release of sperms and eggs from ripe gonads. Induced breeding in captivity of many indigenous fish species were conducted successfully using different hormone. The brooders having the age 2+ years and body

weight as 100-250g attained full maturity during the month of July- August in captivity at 18-22°C water temperature. It breeds during the south west monsoon season in natural condition. The current results showed that successful spawning of *B. devdevi* occurred at the dose of 0.4ml/Kg to 0.6ml/Kg and the dose of hormone affected significantly the percentage of fertilization, egg output, hatching rate respectively. From the current study, 0.5ml/kg body weight of female and 0.25ml/kg body weight for male showed good results for induced spawning and hatching. Similar observations were also reported by Bedajit *et al* (2010) in *Anabas testudineus* by using ovatide as inducing agent. It may be used as a standard dose for induced breeding of *B.devdevi* and can be effectively used for the conservation of this species. It can be suggested to the farmers for commercial seed production of *B.devdevi* in captivity also. In this study, the latency period of *B.devdevi* was found to be 6-10 hours. Bashuda *et al* (2017) observed the latency period of *Bangana dero* as 7-10 hr. Purkayastha *et al.*(2012) found that the latency period of *Ompok pabda* as 9–10 hr when given ovatide at 0.6 mLKg⁻¹ body weight in female.

CONCLUSION

The objective of the present study was fulfilled. Gonopro-FH administered at 0.5ml/Kg body weight produces the highest spawning rate, egg production and hatching rate in *B.devdevi*. The positive response of both males and females to a single dose of Gonopro-FH is significant for commercial seed production. This breeding protocol does not require a high investment, so it can be adopted by small farmers for seed production as well as for species

restoration and conservation. The present study also indicated that optimum dose of Gonopro-FH for normal spawning of *B.devdevi* is 0.5ml/Kg. Since present study was limited to only a few trials, more researches may be necessary to optimize the dose for better production of *B.devdevi* however, considering the present seed demand of *B.devdevi* in Manipur the present information may be effectively utilized for mass-scale seed production.

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