

## Induced Spawning of Bighead Carp *Aristichthys Nobilis* by using Ovaprim-C at Fish Hatchery, Islamabad, Pakistan

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### Abstract

Effects of intramuscular injection of Ovaprim-C, given in single dose, for spawning in bighead carp, *Aristichthys nobilis*, were investigated. Fish were spawned successfully following a single dose injection 0.6ml / kg to female and 0.1 ml / kg to male. Ova and milt were stripped simultaneously and the mixture was stirred for 15-30 seconds for fertilization. Hatching occurred within 18-20 hours after fertilization. Total number of eggs, fertilization and hatching rates of eggs were recorded. Experiment was conducted in circular spawning tank of 2m diameter.

**Keywords:** Induced spawning, Ovaprim-C, Fishes, Bighead carp, *Aristichthys nobilis*

### Introduction

The carp culture is rapidly expanding in Pakistan, but the non-availability of quality fish seed is one of the major constraints in the development of this industry. Major breakthrough achieved by Chaudhry and Alikuknki. (1957), in induced breeding of Indian major carps using pituitary extract has greatly contributed to the rapid development of carp culture. Although technique of hypophysation is practiced successfully but there are certain problems, which have prevented it being taken up widely by fish farmers.

In recent years, human chorionic gonadotropin (HCG) has received some attention as a substitute for pituitary but met with little success, except in the breeding of silver carp, (Chondar, 1990). Luteinizing releasing hormone (LH-RH), a mammalian hypothalamic peptide, has the capacity to release gonadotropin from pituitary gland (Porter *et al.*, 1980).

The Chinese report on the success of mammalian based LH-RH analogue (D- Ala<sup>6</sup>, Pro<sup>9</sup>, Net) for induced breeding of carps created world wide interest on the

use of LH-RH for breeding various species of fish (Anonymous, 1977). A major breakthrough in fish breeding research was the finding that dopamine acts as an inhibitory factor for synthesis of gonadotropin (Peter *et al.*, 1986) when LH-RH alone was used without Pituitary Gland. Spawning failure clearly indicates that dopamine blocks the action of LH-RH on the secretion of gonadotropin. Thus blocking of dopamine action with some antagonists like pimozide, potentiate the action of LH-RH resulting in successful (Chang and Peter, 1983).

Among the various analogues of salmon releasing hormones D-Arg<sup>6</sup> Try<sup>7</sup> Leu<sup>8</sup> Pro<sup>9</sup>, Net has been found to be highly effective and this particular analogue is used in ovaprim. This high effectiveness of salmon releasing hormone is due to its higher affinity for binding sites in the pituitary. (Habibi *et al.*, 1989). Syndel Laboratories Ltd, a Canada Company developed Ovaprim, that contains an analogue of salmon gonadotropin releasing hormone (D-Arg<sup>6</sup>, Pro<sup>9</sup>, Net) and dopamine antagonist, (Nandeeshas, *et al.*, 1990).

There has been considerable research in India on spawning of carps with ovaprim. Kaul and Rishi (1986) reported the successful spawning of mrigal with ovaprim. There are interesting observations reported by other workers in review of (Nandeeshas *et al.* 1990). During (1989), trials were conducted in nine states of India to understand the efficacy of this drug in different agro climate regions, and studied the effect of Ovaprim in comparison with pituitary. Khan *et al.* (1992), reported the successful spawning of rohu and mrigal with Ovaprim (LH-RH analogue) at Fish Hatchery Islamabad. The present report is the outcome of breeding trials carried out at Fish Hatchery and Research Center (FHRC) Islamabad on Bighead carp, *Aristichthys nobilis* during 1992 to 1993.

The Bighead Carp, *Aristichthys nobilis*, like the silver carp, is a filter feeder (Chaudhari *et al.*, 1966; Hopher and Pruinin, 1981) and has omnivorous habit, taking large phytoplankton, algae. It gives preference, to zooplankton in its diet (Lazerava, *et al.*, 1977) which may include rotifers, copepods and cladocera. This species which fits in well in a fish polyculture system, is an extremely fast grower, being able to reach a biomass of 1.5 kg within five months during the summer in countries such as Israel (Hopher and Pruinin, 1981).

The Fingerling of bighead carp were imported from China in 1989. Fingerlings were kept at FHRC Islamabad where they were raised to maturity within three years. These fishes were initially kept in earthen pond for a period of three years, apart from regular manuring, pelleted fish feed was also added. During period of three years, the bighead carps grew from an initial average weight of 40-150gm to a final mass of 2.5 to 6.0 kg in rearing pond, where no artificial feed was provided.

## Materials and Methods

A total of 14 bighead craps brooders weighing from 3.5-5.4 kg were used for this study conducted from May 1992 to June 1997. Fish were selected from stocks reared in 2.5-hectare reservoir at Islamabad Hatchery.

## Selection and Handling

Females with soft, distended belly and pink - red genital papilla and males, which released milt when subjected to gentle pressure on the abdomen, were selected. Fishes were transferred into cemented holding tanks of fish hatchery and anestheized with 100-120 ppm 2-phenoxyethanol solution in a 50 litres fiberglass tank half filled with tap water.

## Hormone Injection

Sex ratios of one female to two males were used per spawning trial. Fishes were injected Ovaprim, intramuscular in a single dose to both sexes following protocol of Nandeeshas *et al.* (1990). Males were injected 0.1-ml/kg Ovaprim, for female dose was 0.6 ml / kg 1).

## Ovulation and spawning

Spawning behavior occurred at about 9-10 hours after the single dose of Ovaprim-C. This was indicated by the intermittent splashing on the water surface as males chased the females. The activity lasted for about 30-60 minutes after which fish were netted out for stripping. A ripe female, which upon slight pressure on the abdomen, released some eggs from the urinogenital splashing on the water surface as males chased the females. The activity lasted for about 30-60 minutes after fish were netted out and stripped. A ripe female, which upon slight pressure on the abdomen, released some eggs from the

urenogenital pore when held ventrally. Females were stripped and eggs were collected in dry plastic bowl. In case of partial spawning, fish were returned to the spawning tank to ovulate further. One or two males were stripped simultaneously with a female to fertilize eggs following the semi-dry fertilization method (Chaudhuri and Singh, 1984). Milt was mixed with the eggs using a bird feather and stirred for 15-30 seconds prior to washing. Three one-gram egg samples were weighed out and counted. The total number of eggs spawned was calculated by multiplying the average number of eggs from the three one gram samples with the total weight of eggs sampled

### Incubation

Breeding test were conducted in circular spawning tanks with 2-meter diameter. Underground water with total hardness of not less than 300ppm was used as hatching medium (Gonzal *et al.*, 1987). Percent fertilization rate expressed as the ratio of

fertilized eggs to total eggs counted ( $n = 100$ ) from each of three sampling of known volume (eggs + water) was determined 6-7 hours post incubation period (early blastula stage). Hatching occurred after 18-24 hours at water temperature of 24.0-26.5°C, hatching rate was then determined. Newly hatched larvae remained in the circular spawning tank for three days until yolk was absorbed.

### Results

Data on ovulation, fertilization and hatching in bighead carp, *Aristihthys nobilis* following intramuscular injection of Ovaprim-C are present in Table 1. Fish given single injection of Ovaprim-C were successfully induced to spawn (Table II). Fourteen females were injected with Ovaprim-C; ovulation of fish in these treatments is 100%. Total number of eggs per/kg was 54,690, fertilization and hatching percentage was 78.36% respectively (Table 1).

**Table 1: Effect of Ovaprim-C on Bighead Carp.**

Parameters	Ovaprim Treatment
No of females treated	14
Total weight of females	56.5kg
Total No of eggs	3.09Million
Total No of fertilized eggs	2.39Million
Total No of hatchling	1.41 Million
Overall fertilization Percentage	78.36%
Overall hatching Percentage	58.99%
Average No of eggs. Kg.	54690
Average No of fertilized eggs/kg	42300
Average No of hatchling/ kg.	24955

Table II: Spawning response of female Bighead carp, *Aristichthys nobilis*

Month	Temperature °C.	No of females	Total weight of females	Dose of Ovaprim (ml/kg)	No of eggs (in Million)	Fertilization rate (in Million)	No of hatching (in Million)
May, 1992	24.5	2	9.2	0.6	0.64	0.60	0.2
May, 1992	24.6	3	9.6	0.6	0.50	0.35	0.23
June, 1992	24.5	1	5.0	0.6	0.30	0.15	0.1
May, 1993	24.0	2	9.7	0.6	0.60	0.54	0.38
May, 1993	24.0	2	7.0	0.6	0.45	0.37	0.27
May, 1993	24.0	2	7.7	0.6	0.25	0.2	0.17
May, 1993	24.0	2	8.3	0.6	0.35	0.18	0.06

## Discussion

Results of this study show that successful induction of spawning in bighead carp *Aristichthys nobilis* is achieved by using a single dose of Ovaprim-C. certain drugs and different analogues of LH-RH are being tested for breeding fishes with varying success (Harvey and Hoar, 1979). However, it was only when the dopamine inhibitory activity in the synthesis of gonadotropin was demonstrated (Peter et al., 1986), that the reason behind the spawning failures became clear. Investigations have now clearly shown the potentiated actions of analogues when they are combined with dopamine antagonists like pimozide or doperidon (Billard et al., 1983; Chang and Peter, 1983; Change et al., 1984; Lin et al., 1986, 1988; Peter et al., 1986; Sokolowska et al., 1984). Based on the extensive research on Chinese carp, Peter et al (1988), defined a new method of breeding called the "Linpe" method in which LH-

RH analogue is combined with a dopamine antagonist. It is a major breakthrough in the history of aquaculture. Ovaprim is a new drug developed essentially on this new method combining releasing hormone with dopamine antagonist. Earlier studies conducted in India (Nandeasha et al. 1990 Khan et al., 1992) have clearly demonstrated superiority of ovaprim in induced spawning of major carps.

Bighead carp does not appear to be a difficult fish to spawn under local conditions. Dose of Ovaprim-C used was 0.6 mg/kg for female. This was based on report of (Nandeasha et al., 1990 Peter et al., 1986) have reviewed dopamine activity in various fish species and indicate that it may vary considerably between species. The positive response of both male and female bighead carp to a single simultaneous injection of Ovaprim-C is very significant from the point of view of

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commercial carp seed production, as it saves a considerable amount of time and avoids excessive handling of brood fish. Present experiments showed that Bighead carp mature within three years and preferably spawn during mid May to June. Table IV fertilization rate was 78.6% while hatching percentage was 88.9% favorable temperature for incubation was 24-26.5°C (Table II).

Generally, the number of eggs spawned by bighead carp in the present study was lower than previously reported. This may be due to the low nutritional status of the brood fish as brood stocks depended completely on the natural productivity of the reservoir and were not given artificial feed throughout the rearing period.

Ovaprim-C being ready to use form, will simplify the breeding operation and hence can be easily adopted in carp seed production. Further trial are needed to standardise the dosage for adoption on a commercial scale.

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