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Bringing blue revolution to the desert

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Abstract

One species of Indian major carps (Labeo rohita) and one species of exotic carp (Cyprinus carpio) were breed by induced breeding method between 19.3.1998 and 8.4.1999 using CIFE D-81 unit which has two type of setting – Indoor and Outdoor. The indoor setting has breeding and hatching unit, while outdoor setting has an underground, overhead tank and a filtration – sedimentation unit. Chlorine, sand and zooplankton free water was used for breeding exercise and natural condition was simulated (showering, water circulation) in the breeding tank after introduction of brooders using the synthetic hormone "OVAPRIM" @ of 0.5 ml/kg wt. The time of giving injection was varied in different exercises but the results were found to be the same. The eggs in case of Labeo rohita were collected in hapa and introduced in the egg containers (2 mm mesh size cotton netting) hanging in hatchery buckets. In case of C. carpio polythene strips having attached eggs were introduced in nursery tank. High dissolved oxygen in nursery tank and hatchery bucket was maintained. The oxygen content in nursery tank was further increased using multiple circular water jet provided at the top circumference of tank. Vertical air lift system in the tank was used for removing metabolites from time to time. The small quantity of water about 5000 to 6000 liters used in whole operation was passed through the filtration and sedimentation unit. The water quality improved considerably after passing through this unit as dissolved oxygen increased from 1.5 ml/l to 4.2 ml/l, free CO_2 decreased from 9.0 ppm to Nil, bicarbonates decreased from 102.0 ppm to 96.0 ppm, nitrate reduced from 16.853 to 7.673 µg/l while phosphate decreased from 0.2567 µgmPO_4^-P/lt to 0.1244 µgmPO_4^-P/lt. This water was found to be suitable for breeding operation and was reused conserving this important commodity of desert.

Keywords: Carps, induced breeding, ovaprim, CIFE- D81 unit, water quality.

Introduction

Fishes, one of the forms of evolutionary higher life to appear in water, is among the earliest vertebrates going back to Devonian period, some 400 million years ago. Today the evolutionary histories of fishes have reached its climax and these are now masters of the world of water. Today some 20,000 species of fishes are known to inhabit water bodies of the world and out of these about 2200 species have been reported from India, which constitutes nearly 11% of the world fish germplasm. The aqua source of India include 2.02 million sq. km. area of Exclusive Economic Zone (EEZ) of surrounding sea, 29000 km length of river, about 1,13,000 kms of canals about 1.75 million ha. of water spread in the form of reservoirs, about 1 million ha. in the form of tanks and ponds and about 0.6 million ha. of stagnant derelict swampy water spread areas.

Tremendous potential exists in India to augment fish production from freshwater aquaculture resources, which are spread across the length and breadth of the country. Fisheries and aquaculture play a key role in providing food and nutritional security to the people, besides providing opportunities for livelihood. India is bestowed with huge untapped aquaculture resources which have to be productively utilized for achieving the set targets. Fisheries contribute significantly to food, nutrition, economic and employment securities, and fortunately are one of the fastest growing agricultural sub-sectors during the last three decades. Currently, fisheries contribute 4.6 per cent of the agricultural GDP, provide employment security to about 11 million people and annually earn foreign exchange worth Rs. 7,300 crore – about one-fifth of the value of the National agricultural export.

Rajasthan possess a large area of inland water bodies which offer potential for development of both intensive and extensive system of culture based fisheries. From the available fresh water resources in the state 3.30 lakh ha. of inland water sheets in the form of reservoir (1.2 lakh ha.) tanks, and ponds (1.8 lakh ha.) and rivers (0.30 lakh ha.) have been identified for capture cum culture fishery management. Out of this about 5700 ha culturable water is available in six districts of western Rajasthan in the form of 176 tanks and reservoirs. The fish production of western Rajasthan is as low as 1000 – 1400kg/ha/year.

The most serious constraint in the expansion of fresh water fish culture in India is the inadequate production of basic input in the seeds of important cultivated carps such as *Catla catla, Cirrhinus mrigala, Labeo rohita, Hypophthalmichthys molitrix* (Silver carp) and *Ctenopharyngodon idella* (grass carp). These breeds only in rivers or "bundh" type tanks especially where fluvatile conditions are stimulated during monsoon. The

bottleneck in seed production was partly solved by initiation of hypophysation or induced breeding techniques.

The breakthrough achieved by Chaudhuri and Alikunhi in induced breeding of Indian major carps with hypophysical extract has contributed to a large extent for the spread of fish culture in India[1]. However the technique was not adapted by the farmers in our country due to intricate procedure of collection, maintenance and preparation of pituitary doses[2-4]. One of the most significant findings of last decade in this field is the isolation and characterisation of Salmon GnRH (gonadotrophin). It has been shown to be more effective than mammalian[5]. Among its various analogues D-Arg[6], Pro[9]-Net has been found to be highly effective and this particular analogue along with domperidone is being used in ovaprim. This analogue is 17 times more potent than LHRHa due to its greater affinity for the binding sites in pituitary[6,7]. The ovaprim contain both Salmon GnRH and domperidone dissolved in calibrated quantities in non-toxic organic solvent. It is considered safe for handling and is known to produce no or little adverse effect on the recipient fish[8].

The development of techniques for collection of seeds in the course of time caused the simultaneous changes in the structure of hatcheries. The fixed cloth hatching hapa of 1950 got improved leading to Zoung jar hatchery, Vertical flow jar hatchery, Glass jar hatchery, Aluminum and plastic bin hatchery, and Galvanised and PVC hatchery in the course of time[9-13]. These hatcheries, although effective in terms of result, had certain limitations as they were unable to control the environmental factors like temperature and dissolve oxygen which are important for hatching of eggs[14,15]. To overcome these limitations Diwedi et al in 1988 devised a controlled hatchery unit CIFE D-81 which was independent of external environment and it ensured high efficiency both in breeding and hatching[16].

Material and methods

Induced breeding of three species of Indian major carps (*Labeo rohita*) and one species of exotic carp (*Cyprinus carpio*) was conducted between 19.3.1998 and 8.4.1999. For the conduction of breeding exercises the CIFE D-81 unit was used which has two type of setting – Indoor and Outdoor. The indoor setting has breeding and hatching unit, while outdoor setting has an underground, overhead tank and a filtration – sedimentation unit. Chlorine, sand and zooplankton free water was used for breeding exercise and natural condition was simulated (showering, water circulation) in the breeding tank after introduction of brooders using the synthetic hormone "OVAPRIM" @ of 0.5 ml/kg wt. The time of giving injection was varied in different exercises but the results were found to be the same. The eggs in case of *Labeo rohita* were collected in hapa and introduced in the egg containers (2 mm mesh size cotton netting) hanging in hatchery buckets. In case of *C. carpio* polythene strips having attached eggs were introduced in nursery tank. High dissolved oxygen in nursery tank and hatchery bucket was maintained using small air compressor. The oxygen content in nursery tank was further increased using multiple circular water jet provided at the top circumference of tank. Vertical air lift system in the tank was used for removing metabolites from time to time.

CIFE D81 Hatchery Unit: This unit is based on the principal of vertical jar hatchery and the hatchery buckets are made up of low density polyethlene (LDPE). The unit has two type of setting: i. Indoor setting. ii. Outdoor setting.

Indoor setting: The indoor setting have Breeding unit and Hatching unit: i. Breeding unit: The breeding unit consists of two plasticraft tanks each measuring 6' (1.83m) in dia meter and 3' (0.91mt) in height with water holding capacity of 2456 lt. Both breeding tanks are fed by overhead shower and have bottom water circulation arrangement. ii. Hatching unit: The hatching unit consists of two plasticraft nursery tanks each (spawn rearing tank) connected by three hatchery bucket. Each nursery tank (spawn rearing tank) measures 8' (2.44mt) in dia and 3' (0.91mt) in height with water holding capacity of 4304 lt.

The hatchery buckets are made up of low density polythelene (LDPE) and are cylindrical at top becoming funnel shaped towards bottom. The hatchery bucket has length of 77 cm, upper diameter of 46 cm and lower dia of 33 cm. Each of the buckets is connected to an inlet by PVC pipe of 2.0 cm dia. The outlet of the hatchery bucket is 2.0 cm and it open into the nursery tank (spawn rearing tanks) through PVC pipe fitting. All hatchery buckets are supported on iron tripod so as to keep them at a height above nursery tank (spawn rearing tank) level.

Inside the hatchery bucket, egg container (capacity 20 lt. height 52.0 cm) made of 2 mm mesh size calico cotton netting are hanged so as to keep a space of 7.5 cm above from the bottom. This is to ensure free movement of spawn after hatching. The egg container can hold 2.0–2.5 lakh eggs depending on the egg size of fish species. The rate of water flow in the hatchery bucket is regulated with the help of a bibcock at inlet. For proper oxygenation small air compressor were used so as to keep the dissolved oxygen level high.

The nursery tank (spawn rearing tank) receives the spawn from hatchery bucket through PVC pipe fitting and they have the provision of water jets at the upper edge to increase oxygen concentration. Vertical air lift system in these ponds is used to remove metabolites from time to time.

Outdoor Setting: The outdoor setting consists of an underground storage tank of 25,284 lt. water holding capacity. Water from tank is pumped in overhead tank (capacity 3000 lt). The overhead tank water, free from Chlorine, sand and zooplankton is used for breeding purposes. The effluent water from breeding and hatchery unit is recycled using a filtration and sedimentation unit of 10,900 lt capacity. This unit is a

continuous flow type – slow sand filtration and sedimentation device made of three subsections.

The water from hatchery unit flow by gravity into the first section atop the sand and gravel beds. The water get filtered after passing detention through these bed and it slowly rise in the 2nd section allowing enough detention time for settable particles. In this section air is pumped through air compressor and the supernatant liquid from this section pass into 3rd section through a connecting tube which open at the bottom of last section. As water rise in this last section all the flock formed in 2nd section get settled at the bottom and clear water comes on the surface. The improved quality water flowed by gravity to the brooder tank to compensate the evaporation losses.

Breeding Operation: i. Collection and Transportation of Brooder: Healthy brooders of *Cyprinus carpio* and *Labeo rohita* were collected time to time from nearby ponds and lakes. These were then transported to laboratory in canvas hapa fitted in open jeep. The collections of brooders were done in the morning hours. ii. Acclimatization and Segregation: After bringing to laboratory, male and female animals were segregated and kept in separate tanks, prepared beforehand by proper aeration. These animals were fed rice bran and oil cake mixed in 1:1 proportion once a day. iii. Breeding day: The breeding operation was planned according to the breeding season of the fish and the breeding tank was well prepared by filling fresh water, (Chlorine and zooplankton free) removing foreign matter and doing aeration. The temperature of water was controlled by heating (by circulating hot-water heated by immersion rod) or cooling (by adding ice) as required so as to keep it in the optimum range of 22–26°C. The limnological parameters of breeding tank were closely monitored taking sample from time to time. For breeding operation of *Cyprinus caprio* polyethene strip measuring 6.0cmx60.0cm tied to galvanized pipe were introduced in the breeding tank before releasing the fishes. Both black and white type of polyethene strips were used but no marked difference was observed in the results.

Healthy brooders were picked from the tank in which they were kept and weighed on single pan balance. They were given ovaprim injection at the rate of 0.5 ml per kg weight at the caudal peduncle and were released in breeding tank. The time of giving injection was varied for different breeding operation, giving it at morning, noon, afternoon, evening and night hours. The results at the end were found to be same thus breaking the notion that breeding is successful only when ovaprim injection is given during night hours.

After releasing the brooders in breeding tanks, natural conditions were simulated in it by circulating water at the bottom, showering from above and covering the breeding tank with black polyethene. Constant aeration was done to keep high level of dissolved oxygen. iv. Egg laying: The time of egg laying was quite variable depending on factor like water temperature. But at optimum temperature range egg laying occurred in 8 – 10 hours. In case of *Cyprinus carpio* eggs got attached to polyethene strips while in case of *Labeo rohita* they remained afloat due to non adhesive nature. After complete egg laying brooders were taken out and eggs were transferred to nursery tank (spawn rearing tank), in case of *Cyprinus caprio* by lifting the attached polyethene strips and to the egg container hanged in hatchery bucket in case of *Labeo rohita.* The hatchery bucket egg container was constantly aerated.

After hatching the spawn from hatchery bucket were transferred to nursery tank (spawn rearing tank). These spawns were kept in the nursery tank (spawn rearing tank) for about 10 days providing them egg yolk as food after 3 days of hatching. After one month these were transferred to outside tank for better growth. Some of them after early growth were stocked in Rawti pond.

Results and discussion

The results obtained from 14 exercises of Induced breeding conducted between 19.03.1998 and 08.04.1999 is summarized in the Table-1.

***Cyprinus carpio*:** In all, eight exercise of induced breeding on this species of carp was conducted between 19.03.1998 to 08.04.1999. The injection of ovaprim was given in morning, afternoon and evening hours, varying it in a different exercise. The ratio of female: male (by weight) ranged from 1: 0.3125 to 1:1.5263. Non availability of brooders was the cause of deviation from the usual ratio 1: 2 used by other workers. Fertilisation rate was found to be higher (60–90%) in the temperature range of 22.0°C to 25.5°C. The spawning was found to vary from the 7 hours 30 minutes to 65 hours at different temperatures. At 16°C the spawning occurred after 65 hours but the hatching failed to occur. The increase in temperature to 22°C caused the spawning time to decrease by 27 hours, while the hatching took 77hours. The minimum time required for spawning and hatching was observed at 25.5°C, being 7 hours 30 minutes and 31.0 hours respectively. The optimum range for effective results (spawning and hatching) was found to be 22°C–25.5°C and total number of spawns produced by virtue of 8 attempts was 12.9847lakh averaging 1.625lakh per exercise.

***Labeo rohita*:** Three exercise conducted between 23.07.1998 to 18.08.1998 using female: male ratio (by weight) between 1 : 0.6326 to 1:1.17444 yielded 3.0933 lakh spawns. The suitable temperature for spawning was found to be 25.0°C when the spawning occurred in 7 hours 45 minutes. The satisfactory results were obtained when the high temperature of water was cooled by adding ice. Fertilisation rate was more than 70% while time taken for hatching was about 38 – 40 hours.

The optimum range of water temperature for breeding of *Cyprinus carpio* and *Labeo rohita* was found to be 24.5 to 26.5°C.

Table-1A: Induced fish breeding between 19.03.1998 and 08.04.1999.

Fish species	Date of Breeding	Time of Ovaprim Injection	Temp. °C Temp. Increase/ Decrease	Brooder weight in Kg.					
				Male			Female		
				Before breeding	After breeding	Weight loss	Before breeding	After breeding	Weight loss
Cyprinus carpio	19.03.98	09.00am	24.5	2.5	2.2	0.3	8.00	7.6	0.4
Cyprinus carpio	24.12.98	06.00am	24.0 20.0 ↑	2.5	2.1	0.4	7.8	7.4	0.4
Cyprinus carpio	10.01.99	03.30pm	16.0	4.0	3.8	0.2	3.0	2.850	0.150
Cyprinus carpio	10.01.99	03.30pm	22.0 16.0 ↑	6.25	6.0	0.25	5.5	5.0	0.5
Cyprinus carpio	02.02.99	06.30pm	22.0	4.5	4.3	0.2	3.6	3.2	0.4
Cyprinus carpio	02.03.99	06.30pm	23.5	2.88	2.67	0.21	2.35	2.0	0.35
Cyprinus carpio	12.03.99	03.00pm	24.0	3.160	2.940	0.22	3.42	2.940	0.480
Cyprinus carpio	08.04.99	08.30pm	28.5 ↓ 25.5	2.9	2.650	0.250	1.9	1.62	0.280
Labeo rohita	23.07.98	08.30pm	28.0 ↓ 26.5	1.550	1.510	0.040	2.450	2.270	0.180
Labeo rohita	14.08.98	09.15am	30.0 ↓ 25.0	1.570	1.550	0.020	0.900	0.800	0.100
Labeo rohita	18.08.98	10.00am	29.0 ↓ 25.5	1.600	1.550	0.050	1.000	0.900	0.100

Table-1B: Induced fish breeding between 19.03.1998 and 08.04.1999.

Fish species	Time / Date of Egg Laying	Spawning time (Hrs.)	No. of Eggs Laid (Lakh)	Fertilization (%)	Hatching time (Hrs.)	Hatching (%)	No. of Spawn Production (Lakh)
Cyprinus carpio	05.15pm 19.03.98	08.15	1.76	80	36.00	70	0.9856
Cyprinus carpio	12 noon 25.12.98	18.00	3.56	90	70.00	80	2.56
Cyprinus carpio	08.30am 13.01.99	65.00	1.25	5	--	--	--
Cyprinus carpio	05.30am 12.01.99	38.00	5.6	90	77.00	60	3.024
Cyprinus carpio	06.30pm 03.02.99	24.00	3.8	80	65.00	66.66	2.02
Cyprinus carpio	06.00am 03.03.99	11.30	3.22	70	38.00	80	1.8
Cyprinus carpio	02.00am 13.03.99	11.00	2.5	60	37.30	80	1.2
Cyprinus carpio	04.00pm 08.04.99	07.30	2.7	65	31.00	82	1.4391
Labeo rohita	06.00am 24.07.98	09.30	2.1	90	40.00	69	1.3041
Labeo rohita	05.00pm 14.08.98	07.45	0.720	70	38.00	73.08	0.3683
Labeo rohita	06.00pm 18.08.98	08.00	2.0	95	40.00	75	1.425

Water temperature in the breeding and hatching tank varied from 22°C to 25.5°C while it ranged from 21.0°C to 21.4°C in filtration and sedimentation tanks sections. The pH remained alkaline, 7.1 to 7.5 except in the subsection I[st] of filtration sedimentation unit. Free carbon dioxide was observed in the breeding and hatching tank after breeding and hatching averaging 4.0 ppm and 8.0 ppm respectively. It was also observed in the I[st] subsection of filtration and sedimentation unit being 9.0 ppm. The carbonates which was present before breeding and hatching was found to be absent after spawning and hatching of eggs. Bicarbonates were found to have increased after breeding and hatching. The high values of bicarbonates were kept in check by changing the water Bicarbonate level which was high in subsection I[st] of filtration and sedimentation unit decreased as the water passed from II to the III subsection. Nitrates were found to have increase from 4.647µg/l to 16.835µg/l after breeding and from 4.887µg/l to 15.272 µg/l after hatching. Phosphate also showed similar rise from 0.096µg/l to 0.2477µg/l after breeding and from 0.098µg/l to 0.2468µg/l after hatching. Dissolved oxygen which play important role in all life activity was found to have decreased after spawning and hatching even with a constant supply of compressed air. Low dissolved oxygen observed in the subsection I[st] of filtration and sedimentation unit, increased as the water reached the III subsection (4.2ml/l) thus improving the water quality (Table-2). The acidic pH observed in the I[st] subsection become alkaline and nitrate, phosphate quantity got reduced at the end after passing from II and III subsection of filtration and sedimentation unit.

Discussion: Trials conducted with ovaprim, on different species of carps administered in single dose to either sex simultaneously showed excellent results in induced spawning. It has been used for breeding commercially important species like *Catla catla, Labeo rohita, Labeo bata, Cirrhinus mrigala, Hypophthalmichthys molitrix, Ctenopharyngodon ideila, Arichthys nobilis, Labeo fimbriatus, Puntis javonicus* and *Heteropneusies fossilis* under different agroclimatic conditions of Karnataka, Kerala, Tamil Nadu, Andhra Pradesh, Madhya Pradesh, Uttar Pradesh, Bihar, West Bengal, Assam and Meghalaya[17-22]. Khan et al Naik and Mirza, Lakra et al and Pandey and Singh successfully bred the Indian major carps using ovaprim[22-26]. The present attempt of breeding *Cirrhinus mrigala, Labeo rohita* and *Cyprinus carpio* with ovaprim was also found to be successful.

Although the injection of ovaprim cut short the events leading to spawning, but the effect of environmental and hydro biological conditions on physiological condition of fish cannot be overlooked. In the present study it was also observed that these conditions greatly affected the ultimate result. Some of these factors are being discussed here.

The water temperature during induced breeding of *Cyprinus carpio* ranged from 16.0°C to 25.5°C. The spawning time varied with the water temperature being high at low water temperature. Little and Dowson reported the spawning time of most of carp within 8– 6 hours[27]. Nandeesha et al reported the range 10–14 hours while Pandey et al reported the spawning time to be over 24 hours[28]. The variability in spawning time is directly governed by water temperature and it was found during the present study that the spawning time of *Cyprinus carpio* increases with decrease in water temperature, below 22.0°C. No spawning was reported in all the three species whenever the water temperature rose more than 31.0°C. The optimum range observed for *Cirrhinus mrigala* and *Labeo rohita* was 24.5°C – 26.5°C. Khan and Chaudhuri observed that Indian major carp breed in range of 24.0°C to 31.0°C and they fail to spawn if the temperature is increased beyond 31.0°C[29,30]. Diwedi et al during the breeding of Indian major carp in the controlled environment observed that temperature range of 26.0 – 28.5°C gives best results[31].

Table-2: Limnological parameters studied during breeding of *Cyprinus carpio.*

Parameters	Breeding Tank (Mean + SE)		Hatchery Tank (Mean + SE)		Filtration and Sedimentation Unit (Mean + SE)		
	Before Breeding	After Breeding	Before Hatching	After Hatching	Subsection I	Subsection II	Subsection III
Temperature (°C)	22.68 + 0.4	22.0 + 0.3	22.5 + 0.3	22.0 + 0.4	21.0 + 0.5	21.4 + 0.5	21.3 + 0.4
pH	7.5 + 0.1	7.2 + 0.1	7.5 + 0.1	7.1 + 0.2	6.8 + 0.1	7.4 + 0.3	7.4 + 0.2
DO (ml/l)	5.04 + 0.40	2.35 + 0.30	4.14 + 0.15	3.97 + 0.2	1.5 + 0.2	3.46 + 0.4	4.2 + 0.3
Free CO_2 (ppm)	Nil	4.0 + 0.1	Nil	8.0 + 0.3	9.0 + 0.3	Nil	Nil
Carbonates (ppm)	10.0 + 0.5	Nil	12.0 + 0.6	Nil	Nil	17.0 + 0.7	15.0 + 0.7
Bicarbonates (ppm)	94.0 + 4.5	102.0 + 5.0	95 + 5.0	100 + 4.5	102 + 4.5	98.0 + 5.5	96.0 + 4.5
Nitrates (µg/l)	4.647 + 0.1	16.835 + 0.2	4.887 + 0.1	15.272 + 0.3	16.853 + 0.3	8.543 + 0.2	7.673 + 0.3
Phosphates (µg/l)	0.096 + 0.003	0.2477 + 0.011	0.098+0.002	0.2567+0.003	0.2567+0.003	0.1982 + .002	0.1244 + 0.001

Dissolved oxygen which was 5.0 ml/l before breeding dropped down considerably after spawning and hatching even when a constant supply of compressed air was maintained. This may be due to the increased consumption by fertilised eggs and the hatchlings. The reduction of dissolved oxygen observed in Ist subsection of filtration and sedimentation unit might be due to increased organic load. The free carbon dioxide observed after breeding and hatching might be due to decomposition of unfertilised eggs. Diwedi et al. also attributed the presence of free carbon dioxide to the fouling caused by unfertilised eggs which are present in large number whenever the fertilisation rate is low. During the breeding of *Cyprinus caprio* (conducted on 10.01.1999) also, high free CO_2, 10 ppm was observed and the fertilisation rate was found to be very low (5%). The free CO_2 observed in the subsection I of filtration and sedimentation unit may be due to the egg shells and excreta coming from breeding and hatching tanks. Nitrate level which was found to have increased after breeding and hatching may be due to the decomposition of excretory products.

Conclusion

The successful results obtained by giving the ovaprim injection to brooders at morning, afternoon, evening and night hours show that time of injection plays no significant role in the spawning of fish. The small quantity of water about 5000 to 6000 liters used in whole operation was found to be suitable for breeding operation and was reused conserving this important commodity of desert.

References

1. Alikunhi K.H. and Chaudhuri H. (1955). Observation on the spawning in Indian carps by hormone injection. *Curr. Sci*, 26(12), 381-382.

2. Nandeesha M.C., Rao K.G., Jayanna R., Parker N.C., Varghese T.J., Keshavanath P. and Shetty H.P. (1990). Induced spawning of Indian major carps through single application of Ovaprim. In the second Asian fisheries forum, (Eds: R. Hirano and M. Hanyu). *Asian Fisheries Society, Manila, Philippines*, 581-585.

3. Harker K. (1992). Breeding carps with ovaprim in India. *Infofish. – Int*., 1, 36-39.

4. Lakra W.S., Mishra A., Dayal R. and Pandey A.K. (1996). Breeding of Indian major carps with the synthetic hormone drug ovaprim in Uttar Pradesh. *J. Adv. Zool*., 17(2), 105-109.

5. Sherwood N., Eiden L., Brownstein M., Spiess J., Rivier J. and Vale W. (1983). Characterization of a teleost gonadotropin-releasing hormone. *Proceedings of the National Academy of Sciences*, 80(9), 2794-2798.

6. Peter R.E., Sokolowska R. and Nahorniak C.S. (1986). Comparision of (D, Arg^6, Trp^7, Leu^8, Pro^9 Net) – Luteninzing harmone – releasing harmones (LHRH-A), in combination with pimozide, in stimulating gonadotropin release and ovulation in the gold fish. *Carrassius auratus*. *Can. J. Zool*., 65, 987-991.

7. Habibi H.R., Marchant T.A., Nahorniak C.S., Van Der Loo H., Peter R.E., Rivier J.E. and Vale W.W. (1989). Functional Relationship between Receptor Binding and Biological Activity for Analogs of Mammalian and Salmon Gonadotropin-Releasing Hormones In the Pituitary of Goldfish (Carasslus Auratus). *Biology of reproduction*, 40(6), 1152-1161.

8. Singh B.N., Das R.C. and Kanugo G. (1998). *Proceedings of the National Workshop on Recent Advances in Harmonal Physiology of Fish and Shell Fish Reproduction*. April 16 – 18. Central Institute of Fresh Water Aquaculture. Bhubaneswar.

9. Huet Marchel (1971). Text Book of fish Culture – Breeding and Cultivation of Fish. Fishing New Books Ltd. Farnham, Surrey, England.

10. Shrigur G.A. (1972). Observations on efficiency of vertical flow jars (glass and transparent polythene) for hatching of major carp eggs. *J. Inland. Fish. Asso*., 2(1-2), 90-101.

11. Bhowmick R.M. (1974). *Summer Institute on Inland Aquaculture*, CIFRI, Barrackpore.

12. Shirgur G.A. (1973). Observations on efficiency of large capacity vertical flow bins for hatching major carp eggs. *J Ind Fish Assoc*, 3, 80-85.

13. Diwedi S.N. (1976). Indigenous vertical jar carp hatchery. *J. Inland Fish. Asso*., 6(1-2), 140.

14. Mukerjee H.K. (1945). Factors influencing the spawning of principal carps of India. *Proc. Nat. Inst. Sci. India*., 11(3)**,** 312-315.

15. Silver S.J., Warren C.E. and Doudoroff P. (1963). Dissolved oxygen requirements of developing steelhead trout and chinook salmon embryos at different water velocities. *Transactions of the American Fisheries Society*, 92(4), 327-343.

16. Dwivedi S.N., Reddy A.K. and Tiwari V.K. (1988). A carp hatchery for India with controlled environment. *J. Inland. Fish. Soc. India,* 20(1), 6-12.

17. Nandeesha M.C., Das S.K., Nathaniel D.E. and Verghese T.J. (1990). Project Report on breeding of carps with ovaprim in India. Special Publ. 4. *Asian Fisheries Society* Indian Branch, Manglore, Karnataka, 1-41.

18. Francis T., Sukumaran N., Devaraj M. and Sundararaj V. (1991). Induced breeding of major carps using Ovaprim. Fishing Chimes, 11(7), 25-27.

19. Azad I.S. and Shimrary D.K. (1991). First success in induced breeding of Indian and exotic carps in Manipur using ovaprim. *Fish. Chimes,* 11(7), 28-29.

20. Pandey A.C. and Singh R.N. (1997). Breeding of *Catla catla* (Bhakur), *Labeo rohita* (rohu) and *Cirrhinus mrigala* (Nain) by ovaprim injection for quality seed production. *J. Adv. Zool*., 18(1), 38-41.

21. Nayak P.K., Mishra T.K., Singh B.N., Pandey A.K. and Das R.C. (2001). Induced maturation and ovulation in Heteropneustes fossilis by using LHRHa, pimozide and ovaprim for production of quality eggs and larvae. *Indian J. Fish*, 48(3), 269-275.

22. Singh B.N., Das R.C., Sarkar M., Kanungo G., Sahoo G.C. and Pandey A.K. (1999). A balanced diet for rearing of singhi, *Heteropneustes fossilis*, broodfish and induced breeding using ovaprim. *Indian J. Fish*.

23. Khan M.N., Janjua M.Y. and Naseem M. (1993). Breeding of carps with ovaprim (LH-RH analogue) at hatchery in Islamabad. *Proc. Pakistan Cong. Zool*., 12, 545-552.

24. Naik I.U. and Mirza Z.S. (1993). Use of ovaprim-C in induced spawning of Indian major carps in Punjab, Pakistan. *Proc. Pakistan Congr. Zool*., 12, 411-416.

25. Pandey A.C. and Singh R.N. (1997). Breeding of *Catla catla, Labeo rohita* and *Cirrhinus mrigala* by ovaprim injection for quality seed production. *J. Adv. Zool*., 18(1), 38-41.

26. Pandey A.C. and Singh R.N. (1997). Ovaprim for mass breeding of Indian major carps. *Indian J. Anim. Sci*., 67 (12), 1133-1134.

27. Little J.M. and Dawson J.A. (1989). Advances in finfish induced breeding – from laboratory to field. *Marine Biotechnology Conference,* Tokyo, Japan.

28. Pandey A.K., Patiyal R.S., Upadhyay J.C., Tyagi M. and Mahanta P.C. (1998). Induced spawning of golden mahasheer (*Tor pulitora*) with ovaprim at state fish farm near Dehradun. *Indian J. Fish*., 45, 457-459.

29. Khan H. (1945). Observations on the spawning behaviour of carps in Punjab. *Proc. Nat. Inst. Sci. India*., 11(3), 315-320.

30. Chaudhari H. (1960). Experiments on induced spawning of Indian carps with pitutary injections. *Indian J. Fish*., **7** (11), 20-49.