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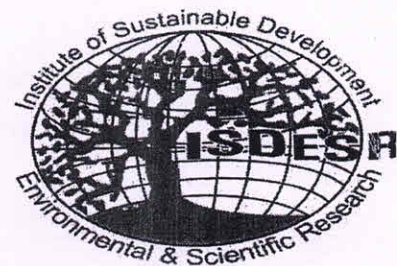
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INDUCED BREEDING OF FRESH WATER FISH CATLA CATLA BY THE APPLICATION OF CARP PITUITARY EXTRACT AND OVAPRIM, AT FISH BREEDING CENTER JAIKWADI, PAITHAN, DIST. AURANGABAD (M.S.) INDIA.

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Abstract

In present study, June - August 2009 and June-August 2010. (breeding season) spawning response of ovaprim compared with pituitary extract in *Catla catla*, at fish breeding center at Jaikwadi, Paithan Dist. Aurangabad (M.S) India. Total ten trial doses of ovaprim were and ten trial doses of Carp Pituitary Extract (CPE) were used for induced breeding in Indian major carps *i.e* *Catla catla*. The overall fertilization 77.12% was found with CPE treatment. and 94.20% with ovaprim treatment. The overall hatching 68.25% with CPE treatment and 92.08% with Ovaprim.

Key words: Induced breeding , *Catla Catla*, Synthetic hormone, Carp pituitary extract, Fertilization rate, hatchling rate etc.

INTRODUCTION

Fish serves as an important source of human diet as they provide proteins, fats and especially vitamins A and D. A special feature of fish is content of vitamins - B, which is absent in plant food. Fish is a good source of calcium. Polyunsaturated fatty acids (PUFA) belonging to linolenic acid series (18:3) are normally present in fish. Fish oil is essential for the prevention of coronary heart diseases. Balanced ratios of ω 3 linolenic acid (18:3) and ω 2 linoleic acid (18:2) in fish flesh are found to be useful for maintaining a healthy heart. The most important fatty acid for human diet is linoleic acid (18:2n6) and linolenic acid (18:3n3), because they cannot be endogenously synthesized (George *et al.*, 2000).

In order to provide food to the ever increasing population, agricultural production alone may not be sufficient to fulfill the demand of the country. As fish food is cheap, rich in protein can possibly be used as an alternative. For this the production of fish on a commercial basis has to be increased, for which all efforts both from research point of view as well as government needed support. The fish eating population is about 56% with a per capita consumption of 9.5 kg/annum. (Jagtap, 2002).

Due to heavy population growth, severely facing the problem of malnutrition and health hazards in common people. For the increasing demand of nutritious food and to get rid off malnutrition, scientists are busy to explore the aquatic resource to the maximum to tide over the problem of people. In the present investigation comparative study of ovaprim and CPE was carried out and finding out an effective substitute for CPE.

MATERIAL AND METHODS

The experiments were carried out during June - August 2009 and June-August 2010. (breeding season) at fish breeding center Jayakwadi, Paithan, Dist. Aurangabad in Maharashtra state 55 km away from Aurangabad. Brooders were collected from the stocking pond of fish breeding center at

Jayakwadi. Healthy males and females were selected: by the external morphological characteristics males and females were identified for the experiments.

After selection of males and females of *Catla catla* in the ratio 2:1 (male: female) were brought to circular hatchery. Pituitary extract was injected intramuscularly in the dorsolateral region. The first dose 0.2 - 0.4 ml/kg body weight was administered to the females for promoting maturation and second dose 0.6 - 0.8 ml/kg body weight was administered to the females at the same time, the first dose 0.2 - 0.4 ml/kg body weight was administered to the males. At the same time for inducing spawning a single dose of ovaprim 0.4-0.6 ml/kg body weight were administered to the both males and females.

Injected brooders were released in a breeding pool. Experimental brooders were observed for 72 hrs after injection; at interval of 4 to 6 hrs the fishes gave responses as a behavioral changes, regarding, maturation, ovulation and spawning.

Preparation of pituitary extract:

The pituitary glands were collected from Indian major carp in the month of June to August. To procure the pituitary, the top of the skull was removed with the help of a knife. Collected pituitaries were homogenized in 0.6% salt solution or distilled water. The solution was centrifuged and the clear supernatants were used for injection.

OVAPRIM:

Ovaprim is a synthetic drug (spawning hormone for fish) manufactured by M/s Syndel Laboratories Ltd., Canada, containing 20 µg sGnRH (salmon gonadotropin releasing hormone) and 10 mg domperidone in 1 ml solution. It was used for final maturation spawning of *Catla catla* and effectiveness is compared with CPE.

Counting of egg:

Total number of eggs laid can be calculated by using following formula

$$\text{Total no. of eggs Laid (approx)} = \frac{\text{Average number of eggs}}{\text{each sample beaker}} \times \text{Number of beakers of eggs}$$

Percentage of Fertilization:

The fertilization rate was calculated through random sampling by examining 2-3 samples from each breeding tank by using following formula

$$\text{Fertilization rate (\%)} = \frac{\text{Average no. of fertilized Eggs in a sample}}{\text{Average no. of eggs in a sample}} \times 100$$

Percentage of Hatchling:

Percentage of hatchling were calculated by following formula

$$\text{Hatchling \%} = \frac{\text{Total no. of spawn}}{\text{Total no of fertilized eggs}} \times 100$$

Table no. 1:- Spawning response of female *Catla catla* with Pituitary extract. (Year 2009)

Mon	No. of female treated	Total wt of female (kg)	Average no. of eggs obtained	Dose of Pituitary extract ml/kg body weight		Average no. of fertilized eggs	Total no. of hatchling	Average no. eggs Kg ⁻¹ (Fecundity)	Average no. fertilized eggs Kg ⁻¹	Average no. Hatchling eggs Kg ⁻¹	Fertilization rate (%)	Hatchling rate (%)
				I st	II nd							
Jun	4	12.0	9800	0.	0.	6800	40000	7906	5666	33333	69.38	58.82

e 09			00	2-0.4	6-0.8	00	0	9	6			
July 09	4	16.5	1300000	0.2-0.4	0.6-0.8	1000000	690000	78787	60606	41818	76.92	69
July 09	4	16.5	1200000	0.2-0.4	0.6-0.8	930000	635000	72727	56363	38484	77.5	68.27
Aug .09	4	17.5	1300000	0.2-0.4	0.6-0.8	970000	630000	74285	55428	36000	74.61	64.94
Aug .09	4	21.5	1700000	0.2-0.4	0.6-0.8	1450000	1135000	81666	67441	52790	85.29	78.82

Table no. 2:- Spawning response of female *Catla catla* with Ovaprim (Year 2009)

Months	No. of female treated	Total wt of female (kg)	Average no. of eggs obtained	Dose of ovaprim ml/kg body weight	Average no. of fertilized eggs	Total no. of hatching	Average no. eggs Kg ⁻¹ Fecundity)	Average no. fertilized eggs Kg ⁻¹	Average no. Hatching eggs Kg ⁻¹	Fertilization rate (%)	Hatching rate (%)
June 09	4	19.0	2000000	0.4 - 0.6	1900000	1790000	121052	92682	87317	92.00	89.31
July 09	4	12.0	1450000	0.4 - 0.6	1370000	1230000	95294	114166	102500	94.48	89.78
July 09	4	16.0	1760000	0.4 - 0.6	1690000	1560000	120833	105625	97500	96.02	92.30
Aug. 09	4	17.0	1620000	0.4 - 0.6	1500000	1350000	140000	88235	79411	92.59	90
Aug. 09	4	20.5	2300000	0.4 - 0.6	2116000	1890000	110000	111368	99473	95.00	94.21

Month	No. of female treated	Total weight of female	Average no. of eggs obtained	Dose of Pitutary extract ml/kg	Average no. of fertilized eggs	Total no. of hatching	Average no. eggs Kg ⁻¹ Fecundity)	Average no. fertilized eggs Kg ⁻¹	Average no. Hatching eggs Kg ⁻¹	Fertilization rate (%)	Hatching rate (%)
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		(kg)		body weight								
				I st	II nd							
June 10	4	10.5	7800 00	0. 2- 0. 4	0. 6- 0. 8	5700 00	38000 0	7428 5	5428 5	36190	73.07	66.66
July 10	4	7.0	5350 00	0. 2- 0. 4	0. 6- 0. 8	4200 00	30000 0	7642 8	6000 0	42857	78.50	70.42
July 10	4	13.0	9000 00	0. 2- 0. 4	0. 6- 0. 8	7200 00	51000 0	6923 0	5538 4	39230	77.64	68.18
Aug .10	4	10.5	8300 00	0. 2- 0. 4	0. 6- 0. 8	6500 00	43000 0	7904 7	6190 4	40952	78.31	66.15
Aug .10	4	12.0	8500 00	0. 2- 0. 4	0. 6- 0. 8	6600 00	45000 0	7083 3	5500 0	37500	80	71.83

Table no. 3:- Spawning response of female *Catla catla* with Pituitary extract. (Year 2010)

Table no. 4:- Spawning response of female *Catla catla* with Ovaprim (Year 2010)

Months	No. of female treated	Total weight of female (kg)	Average no. of eggs obtained	Dose of ovaprim ml/kg body weight	Average no. of fertilized eggs	Total no. of hatching	Average no. eggs Kg-1 Fecundity)	Average no. fertilized eggs Kg-1	Average no. Hatching eggs Kg-1	Fertilization rate (%)	Hatching rate (%)
June 10	4	18.0	1700 000	0.4 - 0.6	1595 000	14400 00	9756 0	8861 1	80000	93.82	90.28
July 10	4	17.5	2015 000	0.4 - 0.6	1914 000	18000 00	9444 4	1093 71.	10285 7	94.98	94.04
July 10	4	19.5	2200 000	0.4 - 0.6	2112 000	19640 00	1225 00	1083 07	10071 7	93.06	92.99
Aug. 10	4	20.0	2450 000	0.4 - 0.6	2280 000	21700 00	1151 42	1140 00	10850 0	96	95.17
Aug. 10	4	20.5	2210 000	0.4 - 0.6	2080 000	19300 00	1128 20	1014 63	94146	94.11	92.78

Table no. 5: Overall effect of Pituitary extract and ovaprim on spawning, *Catla catla*. (2009-2010)

Pituitary extract		Ovaprim	
Parameters	Results	Parameters	Results
No. of females treated	40	No. of females treated	40
Total weight of females	137	Total weight of females	180
Total no. of eggs obtained	1037500	Total no. of eggs obtained	1970500
Total no. of fertilized eggs	805000	Total no. of fertilized eggs	1855700
Total no. of hatchlings	556000	Total no. of hatchlings	1712400
Average no. eggs per kg.	75636	Average no. eggs per kg.	112964
Average no. of fertilized eggs per kg.	58307	Average no. of fertilized eggs per kg.	103383
Average no. of hatchlings per kg.	39915	Average no. of hatchlings per kg.	95242
Overall fertilization %	77.12	Overall fertilization %	94.20
Overall hatchlings %	68.25	Overall hatchlings %	92.08

RESULTS

In the present study we have practiced intramuscular injections in each trial during June-August 2009 to June- August 2010 applying appropriate doses of the hormones. Aggressiveness in the brooders was noticed after 4-6 hrs, of the second dose (0.6 - 0.8 ml/kg body weight) of pituitary extract to female and first dose (0.2 - 0.4 ml/kg body weight) of pituitary extract to male. Whereas the single dose of ovaprim were administrated to both male and female *C. catla*.

The ratio of the male:female (2:1) were selected for each trial. In the present study, 80 males and 40 females which were healthy and disease free brooders selected for the experiments. The average results during study period i.e June-August 2009 and June-August 2010 have been tabulated (Table nos. 1, 2, 3 & 4).

Spawning response due to pituitary extract (2009):

In the month of June 2009 the number of treated females fish were four, total weight of fish was recorded 12 kg. The minimum response was recorded the average number of eggs obtained

980000, average number of fertilized eggs were 680000, average number of hatchlings were 400000, average number of eggs per kg body weight of the fish was 79069, average number of fertilized eggs per kg body weight of the fish was 56666, average number of hatchlings per kg body weight of the fish was 33333, fertilization rate (%) was 69.38% and hatchlings rate (%) was 58.82% of *C. catla* presented (Table no. 1).

While in the month of August 2009 the number of treated females fish were four, total weight of fish recorded was 21.5 kg. maximum response was recorded such as the average number of eggs obtained 1700000, average number of fertilized eggs were 1450000 average number of hatchlings were 1135000, average number of eggs per kg body weight of the fish was 81666, average number of fertilized eggs per kg body weight of fish was 67441, average number of hatchling per kg body weight of the fish was 52790, fertilization rate (%) was 85.29% and hatchling rate (%) was 78.82 % of *C. catla* presented (Table no. 1).

Spawning response due to ovaprim (2009):

During June, 2009 the number of treated female fish were four, total weight of fish recorded was 19.0 kg. the minimum response was observed such as the average number of eggs obtained 2000000, average number of fertilized eggs were 1900000, average number of hatchlings were 1790000, average number of eggs per kg body weight of fish was 121052, average number of fertilized eggs per kg body weight of fish was 92682, average number of hatchling per kg body weight of fish was 87317, fertilization rate (%) was 92.00% and hatchling rate (%) was 89.31% of *C. catla* presented (Table no. 2).

In the month August, 2009 the number of treated females fish were four, total weight of fish recorded was 20.5 kg. the maximum response was noticed such as the average number of eggs obtained 2300000, average number of fertilized eggs were 2160000, average number of hatchling were 1890000, average number of eggs per kg body weight of fish was 110000, average number of fertilized eggs per kg body weight of fish was 111368, average number of hatchling per kg body weight of fish was 99473, fertilization rate (%) was 95.00% and hatchling rate (%) was 94.21% of *C. catla* presented (Table no. 2). Fertilization and hatchling rate (%) which were observed due to pituitary extract and ovaprim are presented in (Fig no. 1& 2).

Spawning response due to pituitary extract (2010):

During June, 2010 the number of treated female fish were four, total weight of fish recorded was 10.5 kg. minimum spawning response was found such the average number of eggs obtained were 780000, average number of fertilized eggs were 570000, average number of hatchling were 380000, average number of eggs per kg body weight of fish was 74285, average number of fertilized eggs per kg body weight of fish was 54285, average number of hatchling per kg body weight of fish was 36190, fertilization rate (%) was 73.07% and hatchling rate (%) was 66.66 % of *C. catla* presented (Table no. 3).

In the month August, 2010 the number of treated female fish were four, total weight of fish recorded 12.0 kg. maximum response was found such as the average number of eggs obtained were 850000 average number of fertilized eggs were 660000 average number of hatchling were 450000, average number of eggs per kg body weight of fish was 70833 average number of fertilized eggs per kg body weight of fish was 55000, average number of hatchlings per kg body weight of fish was 37500, fertilization rate (%) was 80.00% and hatchling rate (%) was 71.83 % of *C. catla* presented (Table no. 3).

Spawning response due to ovaprim (2010):

During the month of June 2010 the numbers of treated female fish were four, total weight of fish recorded was 18.0 kg. minimum spawning response was found such as the average number of eggs obtained were 1700000, average number of fertilized eggs were 1595000, average number of hatchling were 1440000, average number of eggs per kg body weight of fish was 97560, average number of fertilized eggs per kg body weight of fish was 88611, average number of hatchling per kg body weight of fish was 80000, fertilization rate (%) was 93.82% and hatchling rate (%) was 90.28 % of *C. catla* presented (Table no. 4) and fertilization and hatchling rate (%) which were observed due to pituitary extract and ovaprim are presented in (Fig no. 3 & 4).

During month August, 2010 the number of treated female fish were four, total weight of fish recorded 20.5 kg. the maximum response was observed such as the average number of eggs obtained 2210000, average number of fertilized eggs were 2080000, average number of hatchling were 1930000, average number of eggs per kg body weight of fish was 112820, average number of fertilized eggs per kg body weight of fish was 101463, average number of hatchling per kg per

kg body weight of fish was 94146, fertilization rate (%) was 94.11% and hatchling rate (%) was 92.78% of *C. catla* presented (Table no. 4).

The overall effect of pituitary extract such as average number of eggs per kg body weight of fish was 75636, average number of fertilized eggs per kg body weight of fish 58307, average number of hatchling per kg body weight of fish 39915 overall fertilization rate (%) 77.12% hatchling rate (%) 68.25% of *C. catla* presented in (Table no. 5).

While the overall effect due to ovaprim such as average number of eggs per kg was 112964, average number of fertilized eggs per kg was 103383 average number of hatchling per kg 95242, overall fertilization rate (%) 94.20%, hatchling rate (%) 92.08% of *C. catla* presented in (Table no. 5). The effect of ovaprim on spawning response was noticed such as average number of eggs obtained, average number of fertilized eggs, total number of hatchling, average number of eggs per kg body weight of fish, average number of fertilized eggs per kg body weight of fish, average number of hatchling, per kg body weight of fish, fertilization rate (%), hatchling rate (%) of *C. catla* was higher as compared with pituitary extract in every trial. The fertilization and hatchlings percentage were graphically presented (Fig. nos. 1, 2, 3 & 4). The overall response due ovaprim was found better than response due to pituitary extract.

FERTILIZATION (%) RATES:

In the present study due to the pituitary extract the overall average number of fertilized eggs per kg body weight was 58307, Overall fertilization 77.12% while due to the ovaprim the overall average number of fertilized eggs per kg body weight of fish was 103383 and Overall fertilization rate 94.20%.

Fertilization rate was analyzed by T- test, the average number of fertilized eggs per kg body weight of treated fish with pituitary extract as well as ovaprim t- value 1.72 recorded was showed non- significant difference. However, due to ovaprim treatment 17% better results towards fertilization rate (%) as compared with the pituitary extract treatment had been recorded.

HATCHLING (%) RATES:

In the present study due to the pituitary extract the overall average number of hatchling eggs per kg body weight of fish was obtained 39915 and the overall hatchling rate 68.25% while due to the ovaprim the overall average number of hatchling eggs per kg body weight of fish was obtained 95242 and overall hatchling rates 92.08% (Table no. 5)

Hatchling rate was analyzed by T- test, the average number of fertilized eggs per kg body weight of fish treated with pituitary extract as well as ovaprim t- value 1.27 recorded was showed non- significant difference. However, due to ovaprim treatment 24% better results were noticed towards hatchling rate as compared with the pituitary extract treatment.

DISCUSSION

During the present study, a single intramuscular dose of pituitary extract administered to the male and double dose to the female. Whereas due to a single dose of synthetic hormone ovaprim to both male and female were resulted in successful spawning in Indian major carp, *C. catla*. The results of the hormonal stimulation in the present study are quite similar to the earlier findings using ovaprim-C (Muhammad *et al.*, 2005; Basaran *et al.*, 2008).

Results of the present study indicate that ovaprim had the better results as compared with the pituitary extract. Due to pituitary extract ovulation, fertilization and hatchlings values in *C. catla* were recorded such as, total number of eggs 1037500, fertilization percentage 69.38-78.50 %, hatchlings percentage 58.82-78.82%.

Due to ovaprim ovulation, fertilization percentage and hatchlings percentage in *C. catla* were recorded such as total number of eggs 1970500, fertilization percentage 92 - 96.02 %, hatchlings percentage 89.31 - 94.98 %.

In the present study from June-August 2009 – 2010 (breeding seasons), every trial shows that the number of eggs per Kg body weight are higher in ovaprim treated fishes as compared with the pituitary extract treated fishes. Due to the pituitary extract administration, the number of eggs obtained per kg body weight of fish were 79069 – 81666 and due to the ovaprim administration the number of eggs obtained per kg body weight of fish recorded were 94444 – 140000.

Earlier workers have been reported that ovaprim has better results in induced spawning, fertilization and hatchling rate compared with pituitary extract are more or less similar to the present findings regarding induced breeding (Nandeeshha *et al.*, 1990); (Azad *et al.*, 1991); (Khan *et al.*, 1992); (Alok *et al.* 1993); (Chauhan *et al.*1999); (Reddy *et al.*, 2000); (Ragade, 2000);

(Dhabe, 2002); (Dhawan *et al.*, 2004); (Das, 2004); (Muhammad *et al.*, 2005); (Naeem *et al.*, 2005a, b, c); (Rokade *et al.*, 2006); (Sahoo *et al.*, 2007); (More *et al.*, 2010); (Seyed *et al.*, 2010); (Indira *et al.*, 2012); (Abdulraheem *et al.*, 2012) and (Indira, 2012). (More *et al.*, 2016)

Conclusions

Present result indicates that ovaprim might be considered best substitute over pituitary extract during induced breeding.

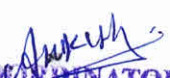
Based on present study it is consequently concluded that the rate of fertilization and hatchling are higher in ovaprim because reduced handlings of brood fish due to the single dose administrated to both the sexes at the same time which decrease post spawning mortality of fish and increase spawning response in ovaprim dose compared with pituitary extract treatment.


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