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Induced breeding, embryonic and larval development of Koi carp (*Cyprinus carpio*) in Khulna, Bangladesh

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Abstract - The aim of the present study was to perform induced breeding and to observe embryonic and larval development stages in different seasons for mature male and female Koi carp, *Cyprinus carpio*, was done by administering the synthetic hormone ovaprim, in a single dose for males and a double dose for females at 0.4-1.0 ml/kg body weight via an intramuscular injection. Spawning occurred between 5-6 hrs after the 2nd injection in summer (April) at a temperature of 26-29 ^{o}C and 17-20 ^{0}C in winter (December). Using doses of 0.4ml/kg, 0.7ml/kg and 1.0 ml/kg of ovaprim, the fertilization rate of eggs were obtained at 42.31%, 54.55% and 61.82 % in summer and 26.92%, 38.00% and 39.23 % in winter. Hatching rates were 42.78%, 44.60% and 55.00 % in the summer season and 30.22%, 31.01% and 31.67 % in the winter season with the same doses of ovaprim. The fertilized eggs were adhesive, transparent, and spherical with diameters ranging between 0.8 mm and 1.0 mm and yellowish white in color. Due to a positive response to the hormone Ovaprim, considerable fertilization and hatching rate, short embryonic period and larval development, it is possible to conduct a successful breeding program of this species commercially.

Keywords: Fertilization, ovaprim, embryonic development.

Introduction

Ornamental fish keeping is one of the most popular hobbies in the world today. The growing interest in aquarium fishes has resulted in a steady increase in aquarium fish trade globally. This trade has a turnover rate of 5 billion $USD and an annual growth rate of 8 percent and offer a wide of scope for development. The top exporting country is Singapore followed by Honkong, Malaysia, Thailand, Philippines, Srilanka, Taiwan, Indonesia and India. The largest importer of Ornamental fish is the USA followed by Europe and Japan. The emerging markets are China and South Africa. Over US $ 500 million worth of ornamental fish are imported into the USA each year (FAO, 2000).

In Bangladesh the trade of ornamental fish is confined to its own territory until now. But domestic production of ornamental fish can save behind money and apparently can be considered as a very potential mean of export earnings. Induced breeding of endemic major carps has been established as a dependable source of fish seeds since the mid 1960's (Ali, 1967) in hatcheries for production in fry or fingerlings which contributes

significantly to the overall aquaculture production of Bangladesh. Usually small fishes, less than 20 cm long, kept in aquariums and prized for their beauty and rarity are called the ornamental fish. In order to sustain the growth it is absolutely necessary to shift the focus from capture to culture based development. Most of the fish species grown for their ornamental importance can be bred in India successfully. Organized trade in ornamental fish depends on assured and adequate supply of demand, which is possible only by mass breeding (FAO, 2000).

Koi carp (*C. carpio*) is a member of the Cyprinidae family and the order Cypriniformes. Besides it is also known as *Cyprinus carpio, Carpio vulgaris, Cyprinus angulatus* etc. (Linnaeus, 1758). It is an ornamental mutation of the common carp (*Cyprinus carpio*), a native from Asia, especially China and Japan. It may look like a big goldfish, distinguish it for it's barbels at the sides of the mouth and for it's size. It is one of the most popular and favorite ornamental fishes amongst all ornamental fish species and it has high market value for its excellent color. The color and scale pattern (squamation) of the species is highly variable. A variety of colors and color patterns have since been developed; common colors include white, black, red, yellow, blue, and cream. They grow up to 100 cm total length with an elongate body measuring 3 to 4 times less in height than total length. In their natural habitat, koi carp live up to 15-24 years (Kuroki, 1981). Males are known to live longer than females. In this study induced breeding techniques, breeding behavior and ontogenic events of koi carp are discussed, since these aspects may be significance to the farming community venturing into the breeding and culture of ornamental fish species. This species exhibits gonochorism, external fertilization, with the spawning frequency that varies throughout their range (Balon, 1990) and are considered as batch spawners (Kalilola *et al.*, 1993).

Considering the enormous importance of koi carp this study was conducted to optimize the dose of ovaprim and to observe the impact of seasonal variation on induced breeding and fecundity. Since the fish is becoming increasingly important not only from the standpoint of the ornamental fish trade, and as an experimental test animal, but also because of its ability to adapt itself successfully to environmental conditions (Helen and Battle, 1939), this study was carried out to highlight some aspects of the early life history of koi carp (embryonic and larval stages) in context of Bangladeshi environment.

Materials and methods

Profile of the study area

These experiments were conducted in Fish Physiology Laboratory of Fisheries and Marine Resource Technology (FMRT) Discipline of Khulna University. All the preparations needed for induced breeding of this fish were done in the laboratory. The aquarium set up, water supply facilities, working space etc. were assured before the breeding program.

Rearing and spawning tank

For induced breeding a glass aquarium was used for both rearing and spawning of fish. Two large size glass aquariums (4×1.5×1.5ft) were made

and including 8 small size glass aquariums (1.5×1.0×1.0ft) of the laboratory. The aquariums were supported with a filter, stones, continuous air-pump etc. The broods were conditioned in the large tank and after injecting the brood set, these were consequently separated in the small tank. After fertilizing, the fertilized eggs were also kept in the small tank.

Experimental design

There were two broad parts of the activities in this experiment. The first part of the activity was concerned with the collection of brood fish. The second part of the experiment was conducted at the FMRT Discipline Laboratory, where induced inbreeding was performed along with observations of embryonic and larval stage development. The total experiment was repeated for two times in two different seasons. The first experiment was repeated for two times, in summer from February 6, 2009 to April 16, 2009 and in winter from August 4, 2009 to December 25, 2009.

Brood fish selection, collection and conditioning

Mature healthy koi carp brood (70-200 gm), two males and one female were selected by sexual dimorphism for breeding experiments. Female is usually easier to identify, as the belly of a mature female is generally plump, whereas male remains streamlined and more torpedo shaped. When males are ready for spawning, they develop breeding tubercles on the head and pectoral fins, principally along the bones of the fin rays. Eighteen pairs (24 male and 12 female) brood fishes were collected from the local aquarium fish market and from the aquarium fish market of Dhaka. The brood fishes were taken to the laboratory within 20 minutes and kept in the aquarium (4×1.5×1.5ft). The brood carrying packs were submerged into the aquarium water for 10 minutes, and after this time these were unpacked and released into a well aerated aquarium (Dissolved Oxygen: 4.5-6.5 mg/l, pH: 7.2-7.8; temperature: 28 -29 ^{0}C in summer and 15-20 ^{0}C in winter (Table 1)).

Induced breeding

The selected broods collected from the conditioning aquarium were kept in a separate aquarium. A Continuous air flow was provided in the tanks by use of a aerator. The sex ratio of the spawners was kept at 2:1 for male and female. The breeding experiment was performed by administering the synthetic hormone Ovaprim.

Hormone dose optimization

For induced breeding Ovaprim was injected to the spawners. Female were given two doses and males a single dose of injection. Male was injected at the time of second dose given to the female. The doses used for the sexes have been placed in Table (2).

Stripping and fertilization

At first females were stripped and eggs were collected in a ceramic plate. In the mean time respective males were stripped and collected milt was mixed well with previously collected eggs. For better result of fertilization, saline water was used and mixed well with a clean and sterile feather. The inseminated eggs were then transferred in to incubation tank providing with continuous air flow.

Table 1. Water quality parameters and room temperature during conditioning of brood fish of Koi carp from Khulna, Bangladesh.

Parameters	Summer (1st trial)	Winter (2nd trial)
Water pH	7.8	7.2
Salinity	0.75 ppt	0.5 ppt
Water temperature	28-29 ^{0}C	15-20 ^{0}C
Room temperature	32-34 ^{0}C	18-22 ^{0}C
Hardness of water	110 mg/l	100 mg/l
Dissolve Oxygen (DO)	6.5 mg/l	4.5 mg/l
Free CO_2	0 mg/l	0 mg/l
Acidity	0 mg/l	0 mg/l
Alkalinity	300 mg/l	290 mg/l

ppt**Parts per million (mg/l)*

Table 2. Doses of ovaprim used in the induced breeding of male and female Koi carp from Khulna, Bangladesh.

Treatment	Sex	Dose: Ovaprim (ml/kg body weight)		Time interval between the doses (hrs)
		1st Dose	2nd Dose	
1	Female	0.4	0.8	6
	Male	-	0.4	
2	Female	0.7	1.5	6
	Male	-	0.7	
3	Female	1.0	2.0	5-6
	Male	-	1.0	

Determination of fecundity

The fecundity of females was observed immediately after stripping. The fecundity was determined by the following formula:

$$\text{Fecundity} = \frac{\text{No of egg in the sample}}{\text{weight of sample}} \times \text{Total egg weight}$$

Determination of fertilization rate

After a certain period (1-2hrs) the eggs were examined to observe the fertilization rate. The fertilized eggs were easily separated from the unfertilized eggs by the presence of a transparent shell with a gray or black spot within the egg shell, while the unfertilized eggs were opaque.

The fertilization rate was determined using the following formula:

$$\text{Fertilization rate} = \frac{\text{No. of fertilized eggs}}{\text{Total no. of eggs}} \times 100$$

Determination of hatching rate

Hatching was completed after 48 ± 2 hours of fertilization in the summer and 70 ± 2 hours in the winter season. In determining hatching rate the samples were collected from the hatching tank and the total numbers of fertilized eggs in the sample and along with the hatchlings were counted by visual observations. The hatching rate was determined using the following formula:

$$\text{Hatching rate} = \frac{\text{No. of hatchlings}}{\text{Total no of fertilized eggs}} \text{X } 100$$

Embryonic and Larval stages observation

Samples of eggs were taken prior to fertilization at every 30 minute interval were taken for further studies. In the present study, the developmental stages were divided into embryonic, larval and post larval development. The embryonic stage occurs inside the chorion and ends in hatching. The larval stage was characterized by nutritive contribution of the yolk sac and the stage ends when the larva becomes capable of exogenous feeding. The post larval stage begins immediately upon absorption of the yolk sac and was characterized by autonomous feeding. After, the yolk sac absorption, the larvae were fed boiled egg yolk twice daily along with zooplankton (Artemia). Developmental time from post fertilization was rounded to the nearest minute until the morula stage and then to the nearest hour. The age of the larvae was denoted an hour after activation.

Descriptions of the developing stages were made by examining live specimens under an Electron microscope and microphotographs of the developmental stages of eggs and larvae were also taken. For a clear an observation, individuals were temporarily stained with methylene blue. The specimens were measured by placing them over a slide having 1.0 mm graph paper at the bottom. Five to ten specimens were used to describe each stage.

Results

In the present study, spawning behavior was observed between 1-8 hrs after hormonal treatment and breeding occurred. The fertilized eggs of koi carp were adhesive, demersal and spherical in nature. The egg envelope is thick, transparent and sticky (Kovac, 2000). The eggs were deposited singly and were highly adhesive throughout the incubation period. Due to the adhesive nature of the egg, considerable debris adhered to the capsule of the egg. The yellowish white egg capsule was transparent, while the yolk was pale yellow or green and granular. The eggs became translucent as development progressed. The diameter of the fertilized egg capsule ranged between 0.8 to1.0 mm while the yolk sphere ranged from 0.6 to 0.8 mm.

Fecundity

Fecundity was estimated soon after the stripping of fish. In the present study 12 female fish (70-160 gm) were examined and the fecundity was 15000 ± 2000 to 70,000 ± 10,000 kg^{-1} total body weight in summer but 1500 ± 200 to 7000 ± 1000 in winter.

Fertilization rate and hatching rate

The fertilization rate of the eggs was 42.31%, 54.55% and 61.82% in the summer and 26.92%, 38.00% and 39.23 % respectively in the winter to 0.4, 0.7 and 1.0 ml/kg body weight of the synthetic hormone (Table 3). The hatching rate of fertilized eggs was obtained at 42.78%, 44.60% and 55.00 % in the summer season and 30.22%, 31.01% and 31.67 % in the winter season respectively (Table 3).

Table 3. Fertilization and hatching rate of Koi carp eggs with respect to different hormonal treatment.

Hormone Dosage (Ovaprim)	Season	Average fertilization rate (%)	Average Hatching rate (%)
T-1	Summer	42.31	42.78
(0.5 ml/kg body weight)	Winter	26.92	30.22
T-2	Summer	54.55	44.60
(0.7 ml/kg body weight)	Winter	38.00	31.01
T-3	Summer	61.82	55.00
(1.0 ml/kg body weight)	Winter	39.23	31.67

T**Treatment*

Embryonic development of Koi Carp (Fig. 1)

Fertilized egg: The fertilized eggs of koi carp were adhesive, demersal and spherical in nature. The eggs were deposited singly and were highly adhesive throughout the incubation period. The eggs became translucent as development progressed. The diameter of the fertilized egg capsule ranged between 0.8 mm and 1.0 mm while the yolk sphere ranged from 0.6-0.7mm.

Formation of embryo: The fertilized eggs presented a spot (blastodisc) on one pole which was distinguished through macroscopic observations. The blastodisc was divided into two distinct cells by vertical cleavage within 1 hour and 20 minutes after fertilization and after 1 hour and 50 minutes a second cleavage was observed forming four cells. The 16-cells stage was obtained after 2 hours and 50 minutes of fertilization. Subsequent cleavage increased cell number and reached morula stage within 5 hours and 10 minutes of fertilized. A cap like structure was seen over the animal pole, which gradually increased in size. Yolk invasion was over started after 7 hours of fertilization and completed after 19 hours of fertilization. The head and tail ends of the embryo became distinguishable during yolk plug stage. Yolk invasion was over and the blast pore was almost closed.

Differentiation of embryo: The notochord was clearly seen at 52 hours after fertilization and at that time 22 somites were seen and lens formation was started and heart formation was almost completed. Blood circulation

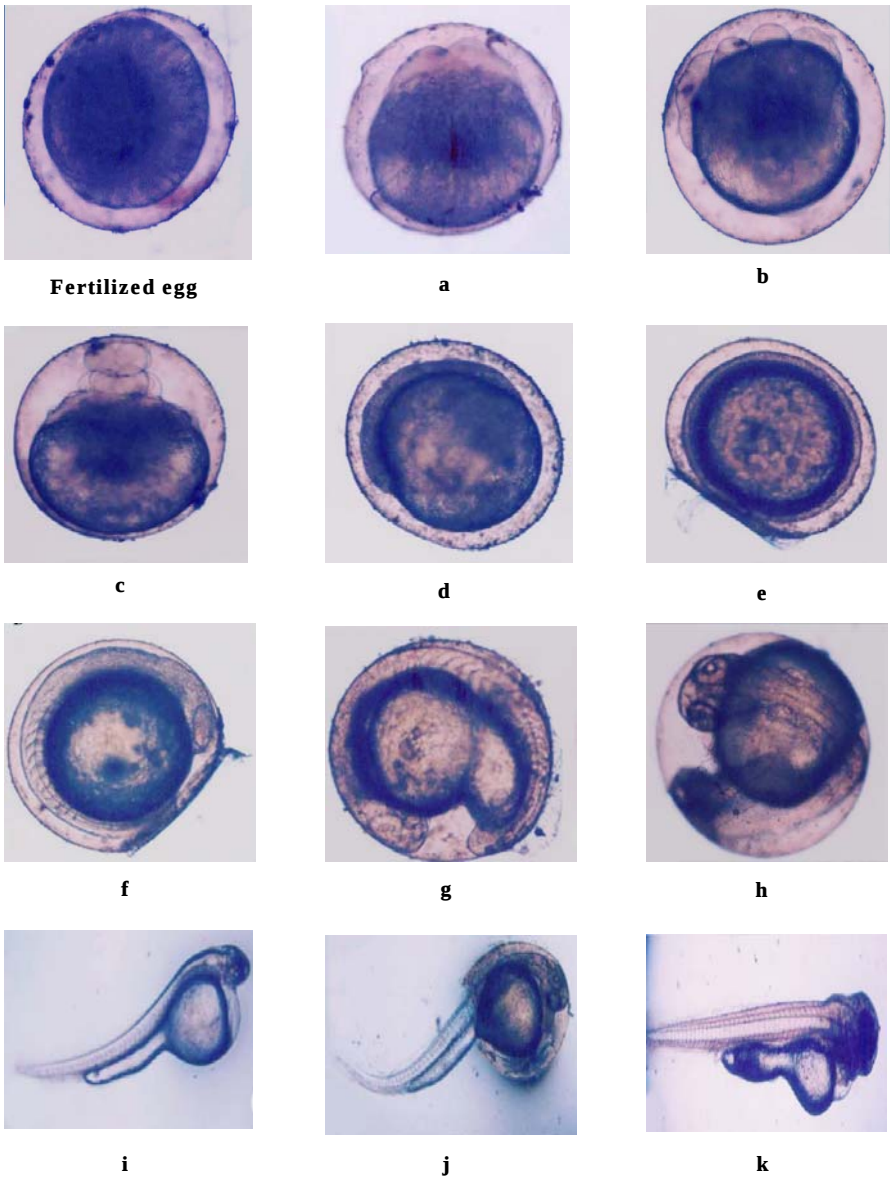

Figure 1. Photographs representing Koi carp during embryonic and larval development stages. Fertilized egg (a) Formation of two blastomeres (b) Eight cell stage (c) Morula stage (d) Eight hours old embryo (e) Eighteen hours old embryo (f) Twenty hours old embryo (g) Thirty hours old embryo (h) 39 hours old embryo (i) Just hatched larva (j) Seventy three hours old embryo (k) 4 day old larva.

commenced over the yolk into the rudimentary heart lying anterior to the yolk sac and 89-93 heartbeats per minute were recorded at this stage. After 71.20 hours of fertilization the caudal region was detached from the yolk mass and became free. Two otoliths were seen in the otic vesicle and 130-140 heartbeats per minute were observed. In the final stage of embryonic development, the growing embryo occupied the entire previtelline space; a frequent twitching movement of the tail whipping against the egg capsule was exhibited.

Larval development

Hatchling: The larva free itself through violent whipping movements of the tail which eventually rupture the egg capsule. Hatching occurred at 75-80 hrs after fertilization and the hatchlings were transparent characterized by the presence of an almost round yolk sac. Newly hatched larvae were slender, straight and transparent, gradually tapering towards the tail. The hatchlings ranged between 2.0 and 2.5 mm in length and tried to hide in any refuge they find. At this stage of development they had no swim bladder, mouth or vent. They breathe by absorbing oxygen through the fine blood capillaries that surround the yolk sac, which were still attached to the gut. The head of the hatchlings were seen above the yolk sac, and the brain was clearly visible.

11-13 hours old larva: After 11-13 hrs of hatching the fin folds were seen continuously around the tail. The vent and gill rudiments were formed. The gut was straight to slightly curve in the anterior portion. The air bladder was shallow, and seen behind the pectoral region, which developed into two chambers in the post larval stage.

2 day old larva: The length of the larvae was 2.7-2.9 mm. The larvae were transparent; fin fold; fin fold originates and heavy ovoid yolk sac. The larvae increased 3.0±0.05 mm in size.

3 day old larva: After 3 days the hatchlings showed free movement. Larvae were able to successfully adhere to the aquarium walls or any fragments of plants. The heart developed chambers, although it is still almost vertical in position at the anterior end of the yolk sac.

4 day old larva: after 4 days of hatching the larvae started exogenous feeding.

Post larval development

8 day old larva: After 7 days a lemon-yellow color was seen in the post-larvae and they attained 4.0-4.5 mm length. By this time the yolk was completely absorbed and the larvae began wandering in search of food.

17 day old larva*:* After 15 days of hatching a distinguishable metamorphosis occurred from the post larva to the fry stage. Most of the fry were lemon yellow in color whereas others had a black and orange coloration. At this stage the larvae increased 6.1-6.6 mm in size.

37 day old larva: After 35 days of hatching the fingerling stage was noticed. At this stage the fingerlings increased between 18.0-23.3 mm in length, distinguished 8 branched rays in the dorsal fin and 7-9 in the caudal fin. Fins were well developed with 17-18 pectoral fin rays, 17-21 pelvic fin rays and 5-6 anal fin rays. They were entirely covered by scales and appeared similar to an adult (Table 4).

Table 4. Summary of embryonic and larval development of koi carp in Fish Physiology Laboratory of Fisheries and Marine Resource Technology (FMRT) Discipline of Khulna University, Bangladesh.

Embryonic development		Larval development		
Time after fertilization (hrs)	Progress in embryonic development	Larval Age (days)	Length (mm)	Characters
0.50	Blastodic formations	2	2.7-2.9	Transparent; fin fold originates and heavy ovoid yolk sac.
1.20	2 celled stage	4	4.3-5.0	Yolk sac absorbed; starte exogenous feeding; eye pigmented
1.50	4 celled stage	7	5.1-5.6	Yellow pigments on the body and a silvery band on the lateral sides
2.20	8 celled stage	17	6.1-6.6	Fin rays differentiated; pigmentation gets dark
2.50	16 celled stage, two tires	22	11.3-13.6	Pigmentation started; various colour combinations noticed
5.10	Morula	37	18-23.3	Fin rays complete; started feeding artificial feed, color more conspicuous.
8.20	Germinal ring formed			
12.30	Half, yolk invasion completed			
19.00	Yolk invasion completed.			
26.00	Cephalic region broadened with distinct fore brain			
32.30	14 somites; cephalic region broadened			
37.40	16-18 somites; optic lens starts differentiating			
45.30	embryonic fin fold formed; gut differentiation started			
52.00	22-25 somites, lens formed in the eye; heart formed; blood circulation commenced, embryo showed slight movements; 89-93 heartbeats / minute.			
67.15	The embryo encircles the whole of the yolk; vigorous twitching movements seen.			
71.20	Lens fully formed, pectoral fin buds seen; 130-140 Heart beats/minute; 2 otoliths in otic vesicle.			
75.20	Hatching started			
80.30	Hatching completed.			

Discussion

In this study the results obtained indicate the fecundity of *C. carpio* did not differ significantly with season. However, significant changes in fecundity occurred with respect to fish size. In this case, generally the larger fish gave a greater number i.e. when considering egg production. The relationship between fecundity (no. of egg production) and the body weight is thus proportional. The fecundity found from 1500 ± 200 to 7000 ± 1000 in each female (15000 ± 2000 to 70,000 ± 10,000 kg^{-1} total body weight). Freeman (1987) reported that the female *C. carpio* deposits eggs - approximately 100,000 per kilogram of body weight. The slightly lower amount in egg number found in this study may probably be due to a smaller brood size. 3 different doses of ovaprim hormone (0.4, 0.7 and 1.0 ml/kg in 1st dose and 0.8, 1.5 and 2.0 ml/kg body weight in 2nd dose) were used, with the best response to reproduction obtained from the dosage of ovaprim of 1.0 ml/ kg and the lowest result was obtained in the case of 0.4/kg body weight of fish.

The fertilization rate and hatching rate are also affected by the seasonal variation that means with variation in temperature. Findings of the present study indicate that both the hatching rate and the fertilization rate are increased in an ambient temperature of 26-29 ^{0}C that is the normal temperature of water in our country in summer season and significantly decreased at winter with a decreased temperature of 17-20 ^{0}C (Table 3). But during the experiment in winter season the environmental condition was very rough. There was an extremely low level of Dissolve Oxygen (2.5-3.0 mg/L) and low temperature that caused an unexpected result; no hatching was observed in this case. The incubation period of eggs depends largely on water quality parameters such as salinity and temperature (Kuo *et al.*, 1973; Liao, 1975). In the present study the findings denotes that the incubation period or hatching time is varied significantly with season especially with temperature. In the summer (26-29 ^{0}C) the eggs hatched earlier than the winter (17-20 ^{0}C).

The present study, succeeded in obtaining *C. carpio* larvae by artificial fertilization the time needed for hatching is about 36-48 hours in 28-29 ^{0}C and 76 ± 4 hours at 17-20 ^{0}C which is near to the findings of Watson *et al.* (2004*)*. They found the time required to hatch is 46 to 54 hours at 28-29 ^{0}C and in 5 to 7 days at 20-24 ^{0}C but vary from the findings of Haniffa *et al.* (2006). They found 72-73 hours are needed for hatching of koi carp eggs at a temperature of 26-28 ^{0}C by using a single dose of ovaprim (0.3 ml/kg). That can be due to different physical condition of the brood fish. The breeding behavior observed by the recent study after injecting the brood fish is almost same that are found by Haniffa *et al.* (2006). The diameter of the fertilized egg capsule found in this study ranged between 0.8 mm and 1.00 mm while the yolk sphere ranged from 0.5- 0.7 mm while Haniffa *et al.* (2006) found that the fertilized eggs were adhesive and transparent with diameter ranging between 0.9 to 1.10 mm. This slight variation can be caused by smaller size brood fish than the brood size selected for the study by Haniffa *et al.* (2006). In the present study, 3 different doses of ovaprim have been used (0.4, 0.7 and 1.0 ml/kg) and it has been found that koi carp response at all the 3 doses but the best response found at a higher dose of

1.0 ml/kg. While Das (2003) observed that most carp fishes responded well at a dose ranging between 0.40 to 0.60 ml per kg body weight. Earlier Nandeesha *et al.* (1990) recommended a dose range of 0.40 – 0.70 ml/kg ovaprim for most carp species in India.

In the present study, we succeeded in obtaining the larvae of koi carp (*Cyprinus carpio*) by artificial fertilization. The fertilized eggs were round, transparent, demersal and adhesive. The color of the fertilized eggs was yellowish white. Balon (1995) and Haniffa *et al.* (2006) found more or less similar in case of common carp and koi carp. The diameter of koi carp (*Cyprinus carpio*) eggs after being fertilized increased from 0.8 to 1.0 mm, while according to Haniffa *et al.* (2006) in koi carp the diameter ranged between 0.9 to 1.10 mm. Miah *et al.* (2008) found the diameter of *Labeo bata* eggs after being fertilized was increased from 0.7 to 0.8 mm. The difference of the egg diameter was due to the species and brood size of koi carp. The size of the swollen fertilized eggs may vary widely even within species. The two cells, four cells, eight cells and sixteen cell stage of *C. carpio* were found within 80, 110, 140 and 170 minutes after fertilized at 17-20 0C, respectively. According to Haniffa *et al.* (2006) in koi carp the same series occurred at 60, 90, 110 and 140 minutes after fertilization at 26-28 ^{0}C. In common carp, the same series occurred at 30, 80, 100 and 120 minutes after fertilization at 26 ^{0}C (Balon, 1995). In *L. rohita*, the same series occurred at 35, 45, 70, 95 and 135 minutes after fertilization (Mookerjee, 1945). Morula stage was found in 5.10 minutes after fertilization where Haniffa *et al.* (2006) observed the same stage in koi carp 4 h 40 min after fertilization and Balon (1995) observed the same stage in common carp three hours after fertilization. This variation might be due to temperature and species difference. The gastrula stage was observed in koi carp at 8.20 to 12.30 hours after fertilization of egg at 17-20 ^{0}C in Winter where Haniffa *et al.* (2006) the same stage in koi carp at 7.30 to 11.40 minute after fertilization at 26-28 in summer season. Balon (1995) observed initiation of gastrulation of *C. carpio* occurring 6 hrs and 30 mins after fertilization of the eggs at 26-28 °C.

The embryo encircles the entire yolk and vigorous twitching movements seen at 67.15 hours after fertilization. At 71.2 hrs after fertilization, lens were fully formed, pectoral fins were observed and 130-140 heartbeats/minute were recorded with 2 otolithsfound in the otic vesicle. Whereas, Miah *et al.* (2008) observed the heart rudiment, gill rudiment and pectoral fin buds of L. *bata* appeared after 17 hours and 30 minutes, 18 hours and 16 hours of fertilization. Mookerjee (1945) observed the heart rudiment, gill rudiment and pectoral fin buds in 15 hours and 50 minutes, 14 hours and 50 minutes and 13 hours and 30 minutes, respectively in *Labeo rohita*. This variation might be due to species difference and temperature. The larvae attained free movement with the help of fins. According to Haniffa *et al.* (2006), 6-8 hours after hatching those above characteristic are found into koi carp larva. This variation might be due to temperature variation. The larvae of koi carp started feeding after 4 days of hatching when they reached a length of 4.3 to 5.0 mm. The embryonic and larval development of koi carp were studied at an ambient temperature of 18 - 20 °C. The rate of larval development of the larvae varied in other

species. This variation is thought to be temperature dependent, the higher the temperature the quicker was the development. The present work generated some information on the early life history, developmental stages and commencement of first feeding time for larval rearing. This study will help the fishery biologist in understanding the biology and ecology of the fish, which might be of great use to take appropriate steps for the sustainable development of the culture and management technology of koi carp. In view of the above findings and discussion, it may be concluded that the embryonic and larval development of koi carp is essential to know its history, culture of fry and fingerlings for nursery and small scale and commercial aquaculture.

Conclusion

The high fecundity, short embryonic period and simple breeding technique of the fish suggest that koi are suitable species for commercial culture. As there are no commercial approaches of induced breeding and seed production of koi carp by any scientific organizations but there is a great demand of koi carp in our country for its colorful and attractive appearance. The breeding techniques discussed in the present paper can be considered a first (if it is) contribution to further large scale breeding of this species. The brood fish, hormone, and other related material required for breeding is available. Due to a good response to the ovaprim, year round breeding performance, considerable fertilization and hatching rate it is possible to conduct breeding program of this species commercially. The successful induced breeding of ornamental fish may reduce the uncertainty and unavailability of fish seed/fry, may increase the large scale production for export purpose and may be a potential sector for meeting the national demand and help to increase foreign exchange earnings. The embryological development of koi carp provides a better idea on their developmental stages and this information helps the mass production of this tropical ornamental fish in captivity.

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التوليد المستحث والتطور الجنيني واليرقي لسمكة الكارب *Cyprinus carpio* في كاهولنا، بنغلاديش

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المستخلص – الهدف من الدراسة الحالية هو لإجراء توليد مستحث لإناث وذكور سمكة الكارب الاعتيادي لغرض متابعة التطورات الجنينية واليرقية خلال فصول السنة. لتحقيق ذلك حقنت الاسماك بهرمون الاوفابريم الصناعي بواقع جرعة واحدة للذكور وجرعتان للاناث وبتركيز 0.4 الى 1 ملم لكل كغم من وزن الجسم. حدث

التفريخ بعد 5 – 6 ساعات وبدرجة حرارة 26 – 29 مئوية في الصيف 17 – 19 مئوية في الشتاء. لوحظ ان البيوض المخصبة لزجة وبيضاء الى صفراء اللون وكروية الشكل وبقطر 0.8 – 10 ملم. ان الاستجابة الواضحة للهرمون ومعدل التخصيب والتفريغ وقصر الفتره اليرقية يشجع على استخدام هذا الهرمون في التفريخ التجاري لهذه الأسماك.