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INDUCED BREEDING OF SILVER CARP *Hypophthalmichthys molitrix* (Valenciennes) UNDER THE EGYPTIAN ENVIRONMENTAL CONDITIONS

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ABSTRACT

The reproductive performance of 181 female silver carps *Hypopthalmichthys molitrix* (Valenciennes) was followed up during 8 months (March-October) in two successive years, 2004 and 2005 under the Egyptian environmental conditions. To study their reproductive performance, ovulation was stimulated in females with two doses of a carp pituitary powder (Argent, Philippines, Manila): a preparatory dose (0.3 mg/kg) followed by a resolving dose of 4.0 mg/kg body weight after 9 h, whilst males were stimulated with one single dose of 3 mg /kg. Females were stripped approximately 7-8 h after the second injection and eggs were immediately fertilized "dry method". Fertilized eggs were incubated in conical shaped fiberglass tanks supplied with aerated fresh flow-through water. An attempt was also carried out to outline the age at maturity in 214 females during 2005. The monthly trend of spawning traits in terms of percentage of ovulated females, egg weight in grams and as a percentage of female's body weight, number of seeds per kg/fish, percentage of fertilized eggs, larval survival rate and percentage of deformed larvae were estimated.

Highest female response to induced breeding and better larval quality were obtainable from April to June. Females of silver carp attained sexual maturity at a body weight of less than 2 kg and an age of two years old or less. The lowest percentage of spawned fish was recorded in March (66.7and 50.0% in 2004 and 2005, respectively). During other experimental months percentage of ovulated females was in the range of 83.3 to 100%. The latency time was approximately 7.5 h at a temperature of 25.5°C. Stripped eggs as a percentage of female body weight (*ESI*) ranged from 6.87 to 15.75%. Relative fecundity of females was in the range of 61,070 and 156,440 eggs/kg. Hatching occurred after approximately 20 hours at 25.5 °C water temperature. Fertilization and survival rates ranged from 85.91 to 97.86% and 72.72 to 80.72%, respectively. The lowest percentages of deformed larvae (11-12%) were observed during April-May compared with either that obtained during the early season (March: 15-20%) or at the late spawning season (July-October: 16-18%).

Keywords: silver carp, hypophysation, sexual maturity, induced breeding, fertilization rate, fecundity, *GSI* index, larval survival rate, percentage of deformed larvae.

INTRODUCTION

The silver carp *Hypopthalmichthys molitrix* (Valenciennes) is probably the most important and the most widely warmwater cultured fish species in the world. The global aquaculture of silver carp has expanded steadily over the past decade, rising from 1.9 million tons in 1993 to 4,1 million tons in 2003 (FAO, Fishery Statistics, 2003). Since the 1950s, after a breakthrough in artificial breeding, the culture of silver carp has spread tremendously in the world due to its unique economic and culture traits.

When grown in cages for 4-6 months, it could attain a weight of approximately 0.7 and 1.0 kg, without supplemental feeding, in Egypt (Personal observation, 2004) and Philippines (Castro *et al.*, 1985), respectively. Silver carp is a native species of China and Russia, where it has been raised in Chinese ponds since historic times. Sexual maturity of Chinese carps varies greatly with environmental conditions and the spawning cycle is profoundly affected by climate (Billard, 1999; FAO, 2003). In China and in other temperate climate areas silver carps are annual spawners. Natural spawning only occurs in riverine habitats providing large volumes of swiftly moving water and rising water may play a role in triggering seasonal spawning, but temperature appears to be the most important factor (Ling, 1980). As such, their spawning cycle breaks down in the tropics (Bardach *et al.*, 1972). The latter authors raised the question of the possibility that the Chinese carps may become perennial spawners under certain environmental conditions. Indeed, Fermin (1991) reported that Chinese carps mature and re-mature the whole year round under cage conditions in Philippines. Peter *et al.* (1988) reported that silver carp spawned early in the season remature again later in the same season.

During the growing seasons of 2002 and 2003 a pronounced expansion in the demand of herbivorous carp fry took place in Egypt. This was due to a recent success in cage culture of silver and big head carps in the river Nile branches. In the growing season of 2003, the price of 1000 silver carp fry increased from almost L.E 25 to L.E. 300 in one year. Therefore, many fish hatcheries attempted to take the opportunity and to hatch herbivorous carps in the following breeding season. An immense demand for brooder fish was evident at that time, and some were sold for approximately L.E 200 a piece, eight or ten times the original market price (Personal observations). However, the available information on hatching and fry rearing of silver carp in Egypt is fairly limited and probably contradicting. The immense activity in artificial propagation of large numbers of silver carps for commercial purposes during the growing seasons of 2004 and 2005 allowed the authors to carry out the present investigation. The principal goal of the present study was to characterize and evaluate the reproductive performance of silver carp females reared under Egyptian environmental conditions.

MATERIALS AND METHODS

Fish and Hormone Treatments in 2004:

The present study was carried out in a private fish hatchery located at Waddy El- Natron, approximately 100 km from Cairo. The study was conducted in two successive spawning seasons, 2004 and 2005. Data of the first year (2004) was collected from 118 silver carp females averaging 4.55 ±0.13 kg body weight, ranging from (2.5 to 8.0 kg). These fish were originally obtained from several fish farms that situated in different areas of Egypt, namely Sharkia, Fayom, and Alexandria governorates, few months before the commencing of the present study. Upon arrival to the hatchery, they were kept in 5 earthen ponds with a total area of 8000 m^2, approximately 1.5 m average depth, at a stocking density that allows approximately 30 m^2 /fish. The earthen fish ponds were regularly fertilized with organic manure along with low doses of single super-phosphate (15 kg per hectare) at fortnightly intervals. Approximately 13% of the water in these ponds was exchanged daily with artisan well water and with the overflow water of a tilapia hatchery. Water quality was around the following criteria: Oxygen, not less than 3.5 mg/l; pH, 7.6; Total ammonia nitrogen, not more than 0.07 mg/l; Total alkalinity, 420 mg/l as calcium carbonate; Total hardness, 380 mg/l as calcium carbonate. Temperature, however, varies during the course of study reaching a maximum of 33°C in few days in July and August, while the lowest was recorded in late March (20°C).

Brood fish were carefully netted for the first time on the 23rd of March 2004 and were sexed and examined for ripeness. Sex identification was made on the basis of the external characters. Mature males were distinguished from the females by the presence of denticulation on the dorsal surface of the pectoral fin which is rough to touch, and by the comparatively flat abdomen. They usually ooze milt at a slight pressure on their abdomen. Female ripeness was assessed by the enlargement and relaxation of the back part of the abdomen, the palpability of the outline of the gonads and the swollen pinkish genital opening. The presence or absence of pre-anal ridge is also taken into consideration as a sign of maturity for selection of female breeders (Horváth *et al.*, 1984; Brzuska, 1999). The first experimental batch was taken on the latter date (23rd of March 2004) when three females manifesting signs of maturity were selected, lightly anaesthetized (according to Jeffrey *et al.*, 2005 with a 100-mg/l concentration of MS-222, tricaine methanesulfonate, Argent Company, Philippines, Manila) in a 150 liter fiberglass tank. Fish were then weighed, marked with a colored thread and the appropriate first carp pituitary dose (powdered carp pituitary, Argent Company, Philippines, Manila) was administrated. Female spawners were treated with two injections of dry pituitary powder according to (Horváth *et al.*, 1984). The preliminary dose was 0.3 mg/kg body weight, whilst the decisive injection (4 mg /kg) was adjusted 9 h later. The latter dose, however, was increased to 5 mg /kg for females weighing 5 kg or more and to 5.5 mg /kg for females weighing 7 kg or more. The same procedure was applied on an equal number of males, although they were receiving different pituitary dose (3 mg hypophysis /kg) at the time of the first injections. Injections of males at that time reduce the hatchery work as males were caught only once before being used, while maximum spermiation is normally maintained for 24 h (Saad & Billard, 1987). The injected fish were immediately placed into concrete circular tanks, 3 m in diameter (spawning tanks). Female and male fish, however, were allocated in different spawning tanks.

The proper pituitary doses were prepared by mixing given amounts of 0.70% physiological saline solution with a precisely weighed pituitary powder. The resultant solution was then drawn up into a syringe with a hypodermic needle. The proper amount of the solution was injected deeply into the muscle below, but ahead of the top dorsal fin and above the lateral line region. The brood fish were then transferred back into the spawning tanks, and during the next hours they were left undisturbed with the tanks covered with shading nets until stripping time. The spawning tanks were continuously provided with flowing freshwater at a constant flow rate that allowed approximately 6 l/min per fish that maintained an oxygen level close to saturation. This water was derived from an artisan well, although it was firstly pumped to a header tank provided with a strong aeration before being distributed to the holding facilities. Water quality was fairly good as indicated by several water analyses. Results of water quality analyses were as follows: Temperature, 25.5°C; pH, 7.4; Oxygen, not less than 5.0 mg/l; Total ammonia nitrogen, <0.05 mg/l. The outflow waters of the spawning tanks were received from the water outlets of the spawning tanks on a fine dark cloth to check the presence of eggs or mucus materials that marks the beginning of estrous among the treated fish.

When the brood fish were in estrus and began to spawn, they were immediately captured and stripped. For striping, brooders were anaesthetized and placed on a thick layer of sponge on the spawning table, and their abdomen, vent and anal fin were thoroughly dried with clean towels. The ripe eggs were released from the ovaries into clean, dry and previously weighed plastic bowls by a gentle massage with a slight pressure on the lower sides of the females from front to back until the ovaries were empty or when a drop of blood appeared. Particular care was taken to avoid any water contact with eggs during the former procedure. Stripped eggs were immediately weighed and a sample of approximately one gram of eggs was then taken and the number of eggs was carefully counted to obtain an estimate for the number of eggs per kg. Fertilization was accomplished using the "dry" method, *i.e.* the milt was added directly to the plastic bowl by stripping the male brooder(s) (Kucharczyk *et al.*, 1997). Eggs and milt were then gently stirred by a goose feather for a minute to ensure that they were thoroughly mixed. Fertilization was then accomplished when few liters of clean water was added gently and mixed. Few minutes later, eggs were rinsed several times and transferred into the egg incubators at a rate of approximately 600 ml of partially swollen eggs per incubator (175 liter). The amount of eggs placed in each jar allowed a total number of hatched larvae in the order of approximately 100,000 per incubating jar. The water flow rate of the incubating jars was carefully adjusted to rotate them gently for the first 10 incubation hours and then was increased to meet the higher oxygen demand of eggs during the final developing stages. Dead eggs were siphoned off several times a day to keep all jars clean and to avoid bacterial and fungal infections. For this later reason, protective doses of formalin (150-200 ppm) were applied during the first 36 hours of this stage. Eggs usually hatch in about 18-20 hours at water temperature of 25.5 °C. The percentage of fertilization was calculated after 12 h incubation period, whilst that of the live embryos and deformed larvae were calculated after 48 h. Sampling of these sexual products was carried out according to Rothbard (1981) and the results were recorded.

Spawning season of 2005

To confirm the results obtained in 2004 and to study the attainment of maturity of silver carps under Egyptian conditions, one ton of growing silver carps, consisting of approximately 400 fish were bought in August 2004 from a private cage farm located in the Rashid branch of the river Nile. These fish were raised in these cages for 16 months (since May 2003) under a high stocking density of approximately 5 fish /m^3. At that time, none of these fish showed signs of maturity, although some specimens exceeded 2.5 kg body weight. The new fish were kept in earthen ponds at a stocking density that allows approximately 20 m^2/fish until the commencing of the following breeding season, *i.e.*, 2005, at which time they were examined for maturity during the period lasted from 27th March till 25th October 2005. During this season, a total number of 214 female fish weighing (3.41 ±0.12) kg in average (ranging from 1.75 to 5.00 kg) were examined for sexual maturity in May 2005, when they were 2 years old. Evaluation of maturity was made according to the signs of ripeness mentioned above. Besides, 115 male fish of the same group were checked for maturity at the same time. Males that ooze milt at slight pressure on their abdomens were considered mature. The percentage of apparently sexually mature females was then recorded, and 63 of them were induced to spawn during the breeding season of 2005, according to the procedure adopted in 2004 to obtain the experimental parameters mentioned above.

Statistical analyses

Statistical characteristics of the present data are given in Tables 1 and 2. Data are expressed as means ± SE. The data were analyzed using analysis of variance, followed by a Duncan's multiple-range test for multiple comparisons. Prior to the ANOVA arcsine transformation was carried out on all value expressed as percentages. The following traits were assessed: percentage of brooders responded to the hormonal treatment, weight of eggs in grams, weight of eggs as a percentage of females' body weight (Egg-Somatic Index), absolute fecundity, relative fecundity, percentage of fertilized eggs, percentage of living embryos and percentage of deformed larvae.

RESULTS

The results of the present study for the breeding seasons of 2004 and 2005 are presented in Table 1 and Table 2, respectively. The data are shown as means (first line) ± SE (second line).

Attainment of maturity

Attainment of sexual maturity in female silver carps were assessed in 214 female fish weighing 3.41 ±0.12 kg in average, ranging from 1.75 to 5.00 kg. The assessment was performed according to the number of females showing signs of ripeness during May 2005 when they were 2 years old, and females with such signs were considered matured. Accordingly, a total of 203 females were considered mature representing a percentage of 94.9% of the examined females. A total 63 of the females from the same cohort were induced to spawn during the breeding season of 2005, while 125 of the evaluated females were sold to another private hatchery in Sharkeia governorate, at which they were induced to spawn. No complain about their reproductive performance was recorded. All the examined males (115 fish) were mature as 100% of them ooze milt when a slight pressure was applied on their abdomens, although many of them were less than 2 kg in weight.

Table (1): Reproductive performance of female silver carps induced to breed in the breeding season of 2004, (means in the first line and ± SE, second line).

Month	No. of Injected fish	% Response	Fish Body Wt. kg	GSI	No. Egg /kg fish x10^3	% Fertilization	% Survival	% Deformed larvae
March	3	66.67 ab ±8.34	5.25 ±1.25	6.87 a ±0.42	61.07 a ±3.72	95.58 bcd ±0.43	79.28 c ±0.73	15.27 bcd ±0.97
April	19	94.74 bc ±1.54	4.18 ±0.31	10.65 abc ±0.91	102.19 abc ±8.76	97.86 d ±0.33	79.47 c ±0.78	11.23 a ±0.82
May	22	90.91 bc ±4.55	4.34 ±0.27	14.25 bc ±1.16	141.09 cd ±11.46	97.45 d ±0.59	80.72 c ±2.58	11.98 ab ±0.99
June	18	100.00 c ±0.00	4.78 ±0.38	15.75 c ±1.62	156.44 d ±16.11	94.83 bcd 0.60	79.45 c 1.46	14.77 abc 0.77
July	15	86.67 bc ±6.67	4.69 ±0.43	10.37 abc ±1.83	92.90 abc ±16.36	86.45 a ±3.72	74.86 abc ±2.35	17.87 c ±1.20
Augst.	15	93.33 bc ±3.34	4.82 ±0.44	8.86 ab ±0.83	80.44 ab ±7.55	85.91 a ±2.80	71.37 a ±1.77	17.81 c ±1.20
Sept.	13	100.00 c ±0.00	3.87 ±0.32	10.31 abc ±0.73	92.47 abc ±6.52	90.91 abc ±1.74	75.67 abc ±0.28	18.44 c ±0.62
Oct.	13	92.31 bc ±3.85	4.46 ±0.35	11.16 abc ±0.78	100.37 abc ±7.00	92.13 bcd 1.34	76.56 bc ±2.39	16.61 bc ±1.40

Means with the same letters are not statistically different (P>0.05).

Table (2): Reproductive performance of female silver carps induced to breed in the breeding season of 2005, (means in the first line and ± SE, second line).

Month	No. of Injected fish	% Response	Fish Body Wt. kg	GSI	No. Egg /kg fish x103	% Fertilization	% Survival	% Deformed larvae
March	2	50.00 a ±8.34	4.00 ±0.00	8.88 ab ±0.00	79.88 ab ±0.00	90.00 ab ±0.00	73.75 abc ±0.00	20.13 c ±0.00
April	12	91.67 bc ±1.54	3.36 ±0.28	8.32 ab ±0.56	81.89 ab ±5.53	97.22 d ±0.94	77.11 bc ±0.80	11.32 a ±1.58
May	8	100.00 c ±0.00	3.53 ±0.27	11.84 bc ±0.48	114.90 bcd ±4.68	96.83 d ±0.96	79.61 c ±3.56	10.87 a ±1.52
June	12	83.33 abc ±8.34	3.78 ±0.23	10.82 abc ±0.47	106.90 abc ±4.65	96.51 cd ±1.18	78.51 c ±0.78	13.74 abc ±2.23
July	6	100.00 c ±0.00	3.38 ±0.46	9.81 abc ±1.41	92.36 abc ±13.31	93.74 bcd ±1.26	74.75 abc ±1.98	15.46 abc ±1.15
Augst.	6	100.00 c ±0.00	3.08 ±0.38	9.15 ab ±1.18	73.67 ab ±9.49	92.27 bcd ±1.48	72.72 ab ±1.21	17.31 c ±1.86
Sept.	8	100.00c ±0.00	3.19 ±0.33	9.06 ab ±1.13	82.56 ab ±10.26	93.51 bcd ±1.37	74.11 abc ±1.20	16.75 bc ±0.10
Oct.	9	100.00 c ±0.00	3.31 ±0.31	12.55 bc ±0.96	118.66 bcd ±9.02	95.26 bcd ±1.36	76.06 abc ±0.93	16.53 bc ±1.07

Means with the same letters are not statistically different (P>0.05).

Percentage of females spawned after hormonal stimulation

Data of the first year (2004) was collected from 118 records of silver carp females averaging 4.46 ±0.13 kg body weights and from 63 silver carp females with an average body weight of 3.41 ±0.12 kg, for the second year (2005). The lowest percentage of spawned fish ($P<0.05$) was recorded in March, 66.7 and 50.0% for 2004 and 2005, respectively (Fig. 1). During other experimental months, however, these values were not significantly different ($P>0.05$) and were in the range of 86.7 to 100%, and 83.3 to 100% for year 2004 and 2005, respectively. Ovulation took place after approximately 7-8 h (*i.e.*, 178.5 - 204 degree hours at 25.5°C) of the decisive injection.

Weight and quality of obtained eggs

Results of the analysis of variance showed a significant effect of spawning months on the weight of eggs expressed as a percentage of female body weight, *i.e.*, Egg-Somatic Index, *ESI* (Table 1) (Fig.2). Higher values for *ESI* were observed in May and June, 2004 (14.25% and 15.75%, respectively) when compared to those of other months, with the exception of October 2005 when *ESI* of 12.55 was recorded (Table 2)(Fig.2), which was the highest estimate recorded in the latter year. The lowest *ESI* values were determined in March and August 2004 (6.87% and 8.86%), respectively. A positive significant correlation ($P<0.05$) was observed between the weight of eggs in grams and female body weight (+ 0.67). Females' relative fecundity was in the range of 61,070 to 156,440 eggs /kg (Table 1). Highest values ($P<0.05$) were recorded in June, 2004 (Fig.3). A positive significant correlation ($P<0.05$) was observed between female body weight and the number of eggs produced (+ 0.65). The egg quality of females spawned during April-June was much better ($P<0.05$) than that produced in the other months as indicated by the improved percentages of both fertilization and live embryos (Fig. 4 and Fig. 5) during those months. Highest fertilization rates were observed in April and May, both in 2004 and 2005, reaching approximately 97-98% compared to (86%-92%) during August, 2004 and 2005, respectively. Similar trends were determined for survival rates, where the lowest were estimated in August (71% and 72%, for 2004 and 2005, respectively), whilst the highest were observed in May (81% and 80%, approximately 81% versus 71%, for 2004 and 2005, respectively). Similarities among the results obtained in 2004 and in 2005, in the above respects were noticeable (Table 1 and Table 2). The lowest percentages of deformed larvae (11-12%) were observed during April-May (Fig. 6) when compared with either that obtained during the early spawning season (March, 15-20%) or the late season (July-October, 16-18%). The differences among those values were statistically significant ($P<0.05$).

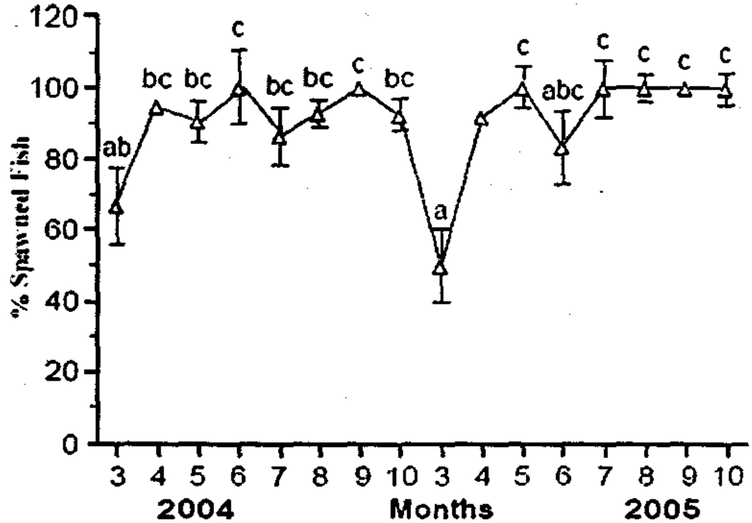

Fig. 1. Percentage of spawned females during 2004 & 2005 breeding seasons. Values with the same litter(s) are not statistically different (P> 0.05).

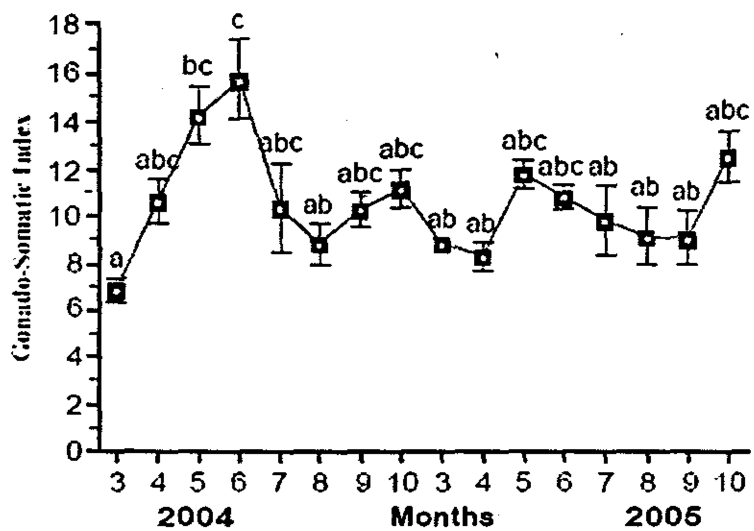

Fig. 2. Weight of eggs as a percentage of female body weight (Egg-Somatic Index) during the experimental periods of 2004 and 2005. Values with the same litter(s) are not statistically different (P> 0.05).

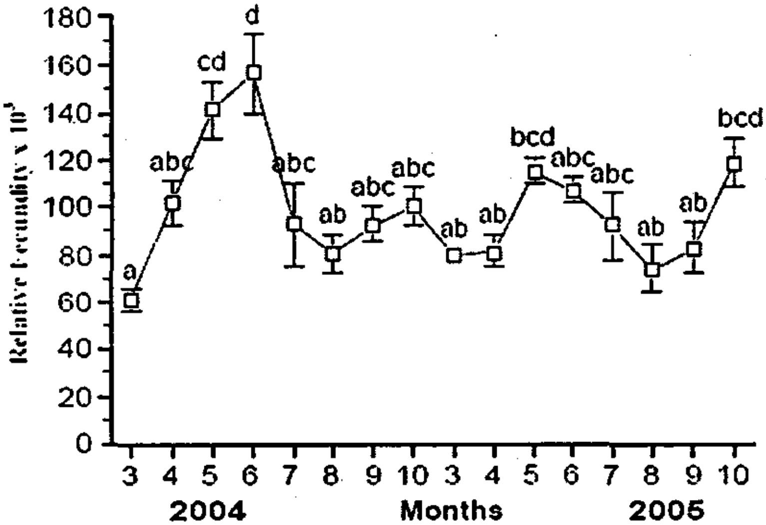

Fig. 3. Females' relative fecundity (No. of eggs /kg fish body weight) during the experimental period. Values with the same litter(s) are not statistically different (P>0.05).

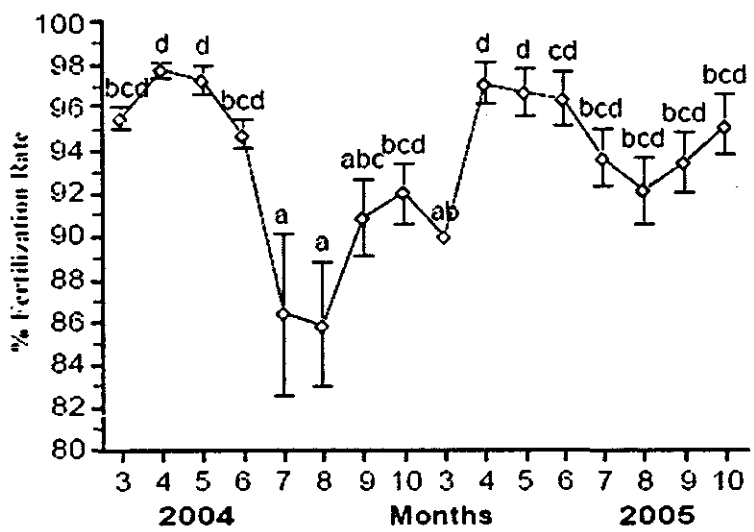

Fig. 4. Fertilization rates during the spawning seasons of 2004 and 2005. Values with the same litter(s) are not statistically different (P>0.05).

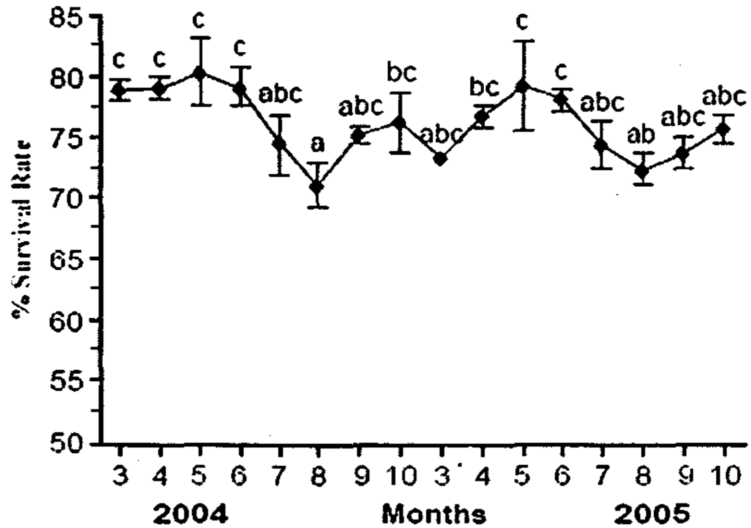

Fig. 5. Survival rates of silver carps' larvae during the spawning seasons of 2004 and 2005. Values with the same litter(s) are not statistically different (P>0.05).

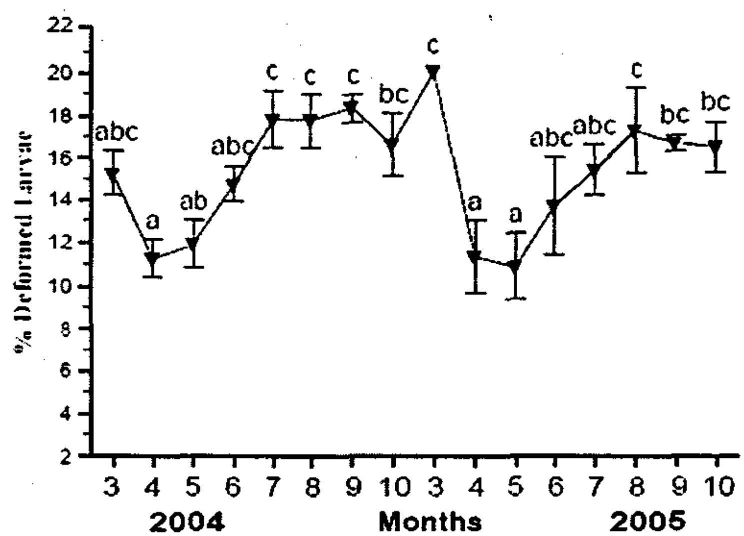

Fig. 6. Percentage of deformed larvae of silver carps induced to spawn during 2004 and 2005 breeding seasons. Averages with the same litters are not statistically different (P>0.05).

DISCUSSION

Under the Egyptian conditions, spawning season of silver carp extends from the end of April to October and reaches its maximum yearly intensity in April-June as indicated by the percentage of spawned female and the quality of their sexual products. The state of maturity in Chinese carps depends, among other things, on the accumulated degree-days from the start of the year in temperate zones, and is thought to be in the range of 1300-1500 (Billard, 1999), which could be achieved in Egypt during spring. The reproductive performance of fish spawned outside the above normal range (April-June) was poor. The poor performance observed in late March may be due to a shortage in the accumulated degree-days. A steady decline in reproductive performance was also experienced after June, probably because of the rise in temperatures in July and onwards above optimum. Nevertheless, although carp spawners at the late season yielded low egg weights with somewhat less quality but still adequate for embryonic development. The present results are in agreement with that critically reviewed for Chinese carps in China (FAO, 1983). According to this report, spawning of Chinese carps is temperature dependent: optimum 25°C, minimum 20°C, maximum 31°C and their natural spawning season extends on average from the end of March to the end of September and reaches its maximum yearly intensity in April and May. A close similarity between temperatures recorded during the course of the present study and that presented in the last report as the lowest temperature was recorded in late March (20 °C), whilst a maximum of 33 °C was observed during some days in July and August. Therefore, resemblance between the present results and that reported for carps in China could be expected. In accordance with these results, spawning season of silver carps under central European climatic conditions is slightly late between May to July, when natural water temperature is in the range of 21-23 °C (Horváth, *et al.*, 1984). In earlier studies, Bardach *et al.* (1972) listed a table showing the differences in spawning activity in eight countries associated with differences in water temperatures and climatic changes, highlighting the differences in spawning seasons of Chinese carps among some different geographical locations.

Results of the present study showed that female silver carps attained sexual maturity when they were two years, although the present findings are inconclusive and it is possible that they could attain maturity at a considerably younger age. Maturity is attainable in small size females, of approximately 1.75 kg body weight or probably less. Differences of age at maturity of Chinese carps among some different geographical locations were reported (Miller, 1995). The latter author showed that such differences even exist among fish grown in north, central and south of China, the natural habitat of silver carps. They attain sexual maturity at 2-3 years in South China, but require 5-6 years to mature further north. In fact, they may require 6-9 years to attain maturity in Rumania (Miller, 1995). The main conclusion is that age of maturity in silver carps depends mainly on rearing temperatures which are closely related to the geographical location they live in. On the other hand, there is a possibility that silver carp can attain sexual maturity at one year old. Billard (1999) noticed that carp in Poland mature at 3-4 years old, 2 years in France, and only one year in Israel. Although maturation in Egypt would be similar to that occurs in Israel, maturation of one year old female silver carp has not been observed in the present study. It has been mentioned earlier that female fish checked for maturity in the present study were obtained from a cage farm, at which they were intially reared for 16 months under a high stocking density of approximately 5 fish /m^3. It is possible that such rearing conditions were not favorable for gonadal maturation. The early attainment of silver carps to maturity in Israel could be due to better husbandry techniques and is possibly achievable in Egypt under good farming conditions. On the other hand, approximately 1 kg males were frequently able to spawn. It is known that maturation of male spawners generally takes 1-2 years less than that of females (Horváth, *et al.*, 1984).

Response of silver carps to the ovarian stimulation was high, except for that observed at the beginning of the breeding season (late March). Signs of maturity were always less manifested at the latter time when compared with that observed during the peak of the season, particularly a distinct enlargement of abdomen sides. Percentage of female response were comparable to that reported in cyprinid species when similar hypophysation technique was adopted (Brzuska & Grzywaczewski, 1999; Kucharczyk *et al.*, 1999) but higher than that obtained in some other studies (Kucharczyk, 1996, 1997; Haniffa & Sridhar, 2002). The availability of large numbers of brooders, during the course of the present study, allowed selection of mature spawners with distinctive signs of maturity, which certainly has contributed to the high percentage of response recorded in the present study. A higher response was observed in females spawned in 2005 in comparison with that obtained in 2004. Brooders of 2005 were more adapted to farm condition than those utilized in 2004 which could have been less stressed than those of 2004. It should be mentioned, however, that these spawners were less in weight (3.41 ±0.12 kg and 4.46 ±0.13 kg for brooders spawned in 2005 and 2004, respectively). The effect of female body weight on the reproductive performance of silver carp was investigated by several authors (Hussain & Mazid, 1999; Shafer, 2000). There is an agreement that small brooders mature early and tend to have a higher relative fecundity but with smaller eggs than larger fish, and this may be further supported by the present findings. Experience gained from the current study showed that the use of smaller brooder size in carp hatcheries is advantageous in several ways. Smaller fish easy to handle, require less quantities of ovulation stimulators, exhibit higher fecundity (Shafer, 2000) and are subjected to less losses after hypophysation. These advantages are certainly applicable to males, particularly general observations made during the present work, on large fish numbers, showed that the amount of milt produced by males are not correlated with males' body size. In fact, Kucharczyk et al. (1996) obtained a negative correlation between quantity of milt and body weight in perch. Furthermore, it has been observed during the course of the present study that a noticeable percentage of mortality occurred in brooders few days after hypophysation. This was generally associated with the stress of handling, injections of ovulation stimulators, spawning and stripping. The immense struggle of large fish to handling could result in fish injures. It has been noticed that post-spawning mortalities were always higher in larger fish. A total of 14 (11.9%) female brooder died after hypophysation in 2004, but only 5 fish (7.9%) died in 2005. This result can be attributed, at least partially, to the difference in size between brooders in breeding seasons 2004 and 2005. Under normal conditions, there is a delay from the time of the last (decisive) injection until the parent fish enter estrus. This period is called the latency or response time, which is an important factor in planning induced spawning in fish and in organizing hatchery processes and may be directly affecting the degree of egg maturation and egg quality (Xu Pao *et al.*, 2003) related to . In the present study, an average latency period of approximately 7.5 hrs (approximately 191 degree-hours) was recorded. Similarly, Xu Pao *et al.* (2003) recorded a latency period for silver carps of 7-8 h, when kept at a temperature range of 24-25 °C, however, Sarder *et al.* (2002) reported slightly less latency time (6 h) for carps in Bangladesh. On the other hand, Makeyeva *et al.* (1996) obtained a much longer latency time in silver carps (20-28 h) treated with LHRH-a analogue. Xu Pao *et al.* (2003) believe that the response time of various species are similar with some slight differences, when given the same hormone and the same dosage in the same season (*i.e.*, temperature). Drori *et al.* (1994) and Yaron (1995) found that latency was always shorter in fish treated with carp pituitary extract than in fish treated with other hormones and drugs. Considering the large numbers of females employed in the current study, the present value should be considered when carp pituitary is used as an ovulation stimulator under the local conditions.

The effect of spawning months on the weight of eggs both in terms of grams per kg fish and as a percentage of female body weight (Egg-Somatic Index) was remarkable. Female silver carps showed relatively high rates of fertility, either as absolute fecundity (784,830 to 243,830 eggs/fish) or in terms of the relative fecundity (61,070 to 156,440 eggs /kg). Maximum values were recorded during June, whilst the lowest were observed during March and August. Similar values (100,000-150,000 eggs/kg) were reported by Kumar (1992) who estimated the number of eggs /kg body weight for silver carp when treated with carp pituitary. However, Brzuska (2004) showed that females of different origins produce different egg weights, whether in terms of g or as a percentage of female body weights. The highest values of Egg-Somatic Index (13.1 and 13.3%) were observed during May and June, respectively, whereas the lowest (7.9%) was recorded during March. The poor performance observed in fish induced to spawn in March may be due to premature spawning. Hussain & Mazid (1999) advocated that artificial seed production from early matured females was associated with poor growth and lower larval survival rates. The quality of the carp gametes obtained in the present work after administration of carp pituitary was relatively high, but somewhat similar to that reported in previous papers when artificial propagation is applied on this species. Irrespective of the spawning month, fertilization was 85.9% and above. Highest percentage of fertilization (97.5%) was noticed in April whereas the lowest was recorded in August (89.1%). High percentages of fertilization in silver carps were also reported by Xu Pao, *et al.* (2003). The latter authors recorded values of 85 and 90% for fertilization and hatching rates, respectively. Shafer (2000) reported a mild fertilization percentage in silver carp ranged from 77.55 to 86.97%. The relatively higher percentages of fertilization rates recorded in the present study might be correlated to the exceptional good water quality with consistent characteristics in the spawning tanks and in the egg incubating facilities.

The overall survival rate of hatchlings in the current investigation after 48 incubation hours was also high (76.5%) with a range of (71.4–80.7%). Comparable results were obtained in several other studies. Brzuska (1999) reported an average survival rate of 75.5% in silver carp, although the same author (Brzuska, 2003) obtained higher survival rates in the common carps after 36 h incubation period (87.2%). Radunz-Neto, *et al.* (1994) believe that commonly reported mortalities of larval carp range is usually around 10%. The percentage of deformed larvae observed during the optimum spawning months (April - June) did not exceed 12.3% which is consistent with the results of other authors (7-13%, Brzuska, 1999). Increased percentages of deformed larvae were observed outside the above seasonal range, reaching a maximum of 20% in March 2005. The most common malformations observed in the current study involved various vertebral abnormalities which amounted to more than 80% of all the defects recorded, in addition to yolk sac malformations, short and very small bodied larvae. Similar observations were reported for newly hatched larvae of common carp (Jezierska, *et al.*, 2000) irrespective of the quality of the rearing water. Several factors are reported to induce malformation in carp larvae. These include low (Stott and Cross, 1973; Tomasik *et al.*, 1982) and high (Mis *et al.*, 1995) temperatures, low dissolved oxygen (Tomasik *et al.*, 1982; Keckeis *et al.*, 1996), water acidification or alkalization (Kugel *et al.*, 1990), exposure to heavy metals (Jezierska *et al.*, 2000). However, none of these factors is known to be exists in the present study. In contrary, water quality of the egg incubators and early stages of larval rearing were of exceptionally good quality. The water was obtained from an artisan well with a constant water temperature of 25.5 °C and was strongly aerated before being directed to the water flow-through rearing facilities. In fact, most of the other examined larval quality criteria were better than averages which were reported by other investigators, and the good present results could be attributed, at least partially, to the exceptionally good experimental water quality. Furthermore, larval malformation occurred in all trials even during the optimum conditions known for carps. Hence, it seems that the existence of malformation of carp larvae of approximately 10% or slightly higher may be due to intrinsic factors related to gamete quality rather than environmental ones. However, the significantly higher percentage of deformed larvae during March may support the early findings of Stott & Cross (1973) and Tomasik *et al.* (1982) who highlighted the increased percentages of deformed larvae due to low environmental conditions.

Conclusion

Female silver carps could attain sexual maturity at two years old and are ready to spawn under the Egyptian environmental condition from March to October, but with varying reproductive performance. Highest female response to induced breeding and better larval quality are obtainable from April to June. Spawning techniques described in the present study (hypophysation) was successful and provided consistent results, producing in average ovulation in 83 to 100 % of the treated females. Absolute fecundity is in the range of 784,830 to 243,830 eggs/fish and the relative fecundity is 61,070 to 156,440 eggs/kg fish. Percentage of fertilization, and larval survival rates were in the order of 85.9% and 76.5%, respectively. Percentage of deformed larvae was mild averaging 12.3% during optimum spawning months but increased during the coldest month of the breeding season (March) reaching a maximum of 20%.

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استحداث التبويض في أسماك الكارب الفضي (*Hypopthalmichthys molitrix*) تحت ظروف البيئة المصرية

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تم تتبع الأداء التناسلي لمجموعة تتكون من ١٨١ من إناث أسماك الكارب الفضي خلال فترة ثمانية أشهر (مارس–أكتوبر) لعامين متواليين ٢٠٠٤ و ٢٠٠٥ تحت الظروف المصرية. استحدث التبويض في الإناث بحقن جرعتين من مسحوق نخامية الكارب (ارجنت – الفلبين) جرعة تحضيريه (٠,٣ مجم/كجم) وجرعة نهائية مقدارها ٤ مجم/كجم بعد حوالي ٩ ساعات بينما تم معاملة الذكور بجرعة واحدة (٣ مجم/كجم أثناء وقت حقن الجرعه التحضيريه في الإناث. تم الحصول على البيض من الإناث عن طريق العصر بعد حوالي ٨ ساعات من الجرعة الثانية وتم تخصيبها في الحال بالطريقة الجافه. تم تحضين البويضات المخصبه في أقماع من الفيبرجلاس مزودة بمياه مهواه ومتدفقه حتى وقت الفقس. اجريت محاوله لدراسة العمر عند النضج الجنسي في مجموعه من الإناث عددها ٢١٤ سمكه في عام ٢٠٠٥. تم تقدير المعدلات الشهريه لنسب الإناث التي باضت ووزن البيض (جم) وكنسبة من وزن جسم الأمهات وعدد البويضات المنتجة /كجم من وزن الأمهات ونسب البويضات المخصبة ومعدلات بقاء اليرقات ونسبة اليرقات المشوهة.

اتضح أن أعلى نسبة استجابه لاستحداث التبويض وأفضل حاله لليرقات الناتجه تحت الظروف المصريه كانت في الفتره من ابريل إلى يونيو. تصل إناث أسماك الكارب الفضي إلى النضج الجنسي على وزن يقل عن ٢ كجم وعمر سنتين أو أقل. أقل نسبة للتبويض كانت في شهر مارس (٦٦,٦% ، ٥٠% في عامي ٢٠٠٤ و٢٠٠٥ على الترتيب). خلال الأشهر التجريبيه الأخرى تراوحت نسبة الإستجابه في الإناث من ٨٣,٣% إلى ١٠٠%. كان وقت الإستجابه حوالي ٧,٥ ساعه عند درجة حراره ٢٥,٥ مئوية. تراوحت أوزان البيض الناتجة بالنسبه لوزن الأمهات من ٦,٨٧ إلى ١٥,٧٥%. تراوح عدد البويضات الناتجة من الأمهات بين ٦١٠٧٠ إلى ١٥٦٤٤٠ لكل كجم. تم الفقس بعد حوالي ٢٠ ساعة من التحضين على درجة حرارة ٢٥,٥ درجة مئوية. تراوحت نسب الإخصاب والبقاء بين ٨٥,٩–٩٧,٩% و ٧٢,٧–٨٠,٧% ، على الترتيب. لوحظ أن أقل نسبة لليرقات المشوهة (١١–١٢%) كانت في الفترة بين ابريل –مايو بالمقارنه بتلك التي لوحظت في بداية الموسم (مارس ، ١٥–٢٠%) أو في نهايته (يوليو–أكتوبر ، ١٦–١٨%).