Oodev Injection Frequency and Time Period in Advancing Gonad Rematuration of Snakehead (Channa striata Blkr) in Hapa System

Kaspul Anwar¹, Untung Bijaksana², Herliwati², Ahmadi²*  

¹Postgraduate Program of Fishery Science, ²Faculty of Marine and Fisheries, Universitas Lambung Mangkurat, Banjarbaru 70714 Indonesia  
*Corresponding author: ahmadi@unlam.ac.id

Abstract—This study provides meaningfully scientific information on the attempt to accelerate the gonad rematuration of snakehead (Channa striata) to support the quality seed production for both commercial and restocking purposes in South Kalimantan, Indonesia. A total of 48 snakehead fish broods (296-312 mm total length and 234-298 g weight) were subjected to different treatment levels using oodev dose of 0.5 ml kg⁻¹ fish weight to determine the best frequency and time period for gonad rematuration after spawning. Treatment-A: 3 times/9 days, B: 2 times/6 days, C: 1 time/3 days, and D: no oodev injection (control). The 12 hapas were used comprising 4 individuals per hapa (0.4×0.4×1 m) under the controlled condition. After all, ten snakehead brood samples were dissected to find out the gonad maturity level of fish. The treatment-A showed the best performance in term of mean fecundity (1960±450.00 granules/individuals), egg diameter (1.10±0.15 mm), and gonad somatic index/GSI (3.41±0.90 %) among other treatments. Dealing with hepatosomatic index (HSI), the treatment-C was significantly higher than treatment-A and D, but not differ from treatment-B. The mean HSI was varied from 0.89±0.03 % to 1.53±0.23 %. Temperature, pH and DO during sampling period are considered comfortable for snakehead fish broods.

Keywords— Channasriata, gonad rematuration, fecundity, egg diameter, GSI, HSI.

I. INTRODUCTION

In the world, the snakeheads comprise two extant genera, namely Channa (36 species native to Asia) and Parachanna (four African species), and most recently, barcoding snakehead is revisited to solve perpertuated taxanomi confusions (Conte-Grand et al., 2017). Snakehead (Channa striata Blkr) of family Channidae, is considered as valuable food fish not only in Indonesia (Widodo et al., 2013; Irhamsyah et al., 2017), but also other countries such as Thailand (Khomsab and Wannasri, 2017), Philippines (Jumawan and Seronay, 2017), Vietnam (Quyen et al., 2016), Malaysia (Song et al., 2013), Cambodia (Sinh, 2014), Sri Lanka (Wijeyaratne, 1994), Nigeria (Ama-Abasi and Ogar, 2013), Bangladesh (Islam et al., 2013), India (Kashyap et al., 2014), Pakistan (Najero et al., 2015) and China (Gu et al., 2015) due to delicious, high-quality meat fish and availability throughout the year. Snakehead can be commercially cultured in fish farming, earthen ponds, or hapa system (Kumar et al., 2011; Quyen et al., 2016), and culture strategies of them are currently being developed (Xie et al., 2002; Xie et al., 2017; He et al., 2015; Istitiano and Diana, 2016). They inhabit freshwater such as swamps, rivers, streams, lakes, reservoirs, irrigation canals and paddy fields (Saikia et al., 2012; Muthmännah, 2013; Kashyap et al., 2014; Sakhare, 2015; Singh and Serajuddin, 2017). Snakehead locally known as “Haruan”, is able to tolerate to adverse environments due to its hardiness and air-breathing capabilities assisted with a suprabranchial chamber, an air-breathing organ (Chandra and Banerjee, 2004). They are carnivorous feeders that consume plankton, aquatic insect, mollusks, fish or frogs. Snakeheads from river or swamps are caught by using hooks, gillnet, castnet or fish pot (Song et al., 2013; Irhamsyah et al., 2017). Overfishing, pollution, habitat disruption, disease, and growing human intervention on wetlands is very likely threat to this species (Balkhīs et al., 2011; Uthayakumar et al., 2014; Rao et al., 2015).

Seasonal reproduction is a strategy adapted by fishes to guarantee better chances of their offspring’s survival bytiming reproductive activity with favorable conditions during certain times of the year (Bernal et al., 2015). An understanding of the reproductive biology of fishes in relation to external factors (e.g. photoperiod, temperature, rainfall, salinity, and food supply) in their habitat is useful in the propagation of captive fish broodstock (Bromage et al., 2001). Snakehead in swamp waters spawning in the early or mid-rainy season. It takes longer time for the gonad growth and reproductive process, meanwhile the culture development is highly depend on the availability of
fish seeds that meet the timeliness, quality and quantity. Seeds can be continuously produced if it is supported by the availability of matured broodstock with good egg quality. One of efforts to meet the need of seeds production is by increasing the spawning frequency of Nilem fish broodstock through the acceleration of gonad rematuration period after spawning. The gonad rematuration period usually takes longer time compared to the maturation process. It is clearly found in *Osteochilus hasselti* in which the gonad maturation period takes 10 days (Cholifah, 2016), while its rematuration period takes 17 days (Fitriatin et al., 2018) by using oodev hormone 1 mL kg⁻¹.

Understanding the proper function of follicle stimulating hormone (FSH) of oodev content will reduce the artificial spawning failure of the snakehead, because it is directly involved in the acceleration of the vitelogenesis process (Moore and Ward, 1980). The previous studies confirmed that oodev hormone beneficially supports for gonad maturation and spawning in *Pangasius hypophthalmus* (Ernawati, 1999), *Clarias batrachus* (Zairin et al., 2000), *Hemibagrus nemurus* (Supriyadi, 2005), and *Osteochilus hasselti* (Cholifah, 2016), as well as for gonad rematuration in *P. hypophthalmus* (Agustinus, 2013), *Anabas testudineus* (Sari, 2015), and *O. hasselti* (Fitriatin et al., 2018), but lack of available information on most snakehead species. Therefore, the present study is performed to determine the best frequency and time period (day) for advancing the snakehead gonad maturation through oodev hormone intervention. From all stages of this research, we endeavor to provide a simple technology package of snakehead gonad rematuration to supply the needs of quality seeds for both commercial and restocking purposes.

II. MATERIALS AND METHODS

Study site and Experimental condition

The research was carried out in the Wet Laboratory at the Faculty of Marine and Fisheries, University Lambung Mangkurat for three months. A set of tools, equipment and materials are well-prepared as described in Table 1. About one hundred snakehead broodstocks were collected from local fishers in the monotonous swamp and were kept in the Wet Laboratory. The only forty-eight fish samples (296-312 mm total length and 234-298 g weight) were used in this study, while the remaining fish was kept in a large hapa (1.5×1.5×1.0 m) as stock in case of the death during the trial periods. The hapas were placed in a concrete tank pond (7×4×1.2 m). Tap water was precipitated one week before treatment. Water lettuce (*Pistia stratiotes* L) was given for shelter and supplemental nutrients in the water. Fish were fed with the frog as 4% of body weight per day with the frequency of once a day. The twelve hapas were used in this experiment with the density level of 4 individuals per hapa (0.4×0.3×1.1 m).

Water quality parameters recorded during the study were as follows: temperatures from 26.40 to 27.45 °C, pH from 6.90 to 7.23, DO from 4.59 to 6.10 mg L⁻¹ and NH₃ from 0.15 to 0.23 mg L⁻¹. Temperature, pH and DO were measured using Watercheckker U10 Horiba, while NH₃ was determined by Spectofotometer with Spec-Nessler method.

Experimental Procedures

The experimental design used was Completely Randomized Design with four different treatments (A, B, C, and D) and each treatment was repeated three times. For treatment-A, B and C, the *Ooodev* dose was given to the snakehead broodstocks with the same amount of 0.5 mL kg⁻¹ fish weight but vary with frequency and time period (day). For instance, the treatment-A: 3 times/9 days, B: 2 times/6 days, C: 1 time/3 days, and D: no oodev injection (control) and later to be compared to other treatments. The details of experimental treatment are given in Table 2. At the end of experiments, a total of ten snakehead samples were dissected to find out the gonad maturity level of fish, and reproductive indicators such as egg diameter, fecundity, Somatic Gonad Index (SGI) and Hepato Somatic Index (HSI) were measured. The egg diameter was measured using a micrometer. The fecundity, SGI and HSI were calculated using the following formulas:

\[ F = n \times G / g \]  \hspace{1cm} (1)

F is fecundity; n is the average number of eggs in sub-sample; G is gonad weight (g); and g is the sub-sample weight (g).

\[ GSI = Gw / Bw \times 100\% \]  \hspace{1cm} (2)

GSI is Gonad Somatic Index; Gw is Gonad weight (g); and Bw is Body weight of fish (g).

\[ HIS = Lw / Bw \times 100\% \]  \hspace{1cm} (3)

HSI is Hepato Somatic Index; Lw is Liver weight (g); and Bw is Body weight of fish (g).

Statistical Analysis

At the beginning, the normality and homogeneity of experimental data obtained were analysed using Lilliefors test and Bartlett test, respectively. Data transformation should be first done if data were found not normal or not homogeneous. If the assumption was fulfilled, then apply for the Analysis of Variants (ANOVA). The Mean differences test was used if there were significantly
differences among the treatments. All tests were analysed at the 0.05 level of significance using SPSS-16 software.

III. RESULTS AND DISCUSSION

The mean ± standard deviation of fecundity, egg diameter, somatic gonad index (SGI) and hepatosomatic index (HSI) obtained from experiments was presented in Table 3. It is clearly shown that among different injection frequency, the treatment-A was determined as the best performance in term of generating fecundity, egg diameter and SGI rate i.e. 19600 ± 450.00 granules/individuals, 1.10 ± 0.15 mm and 3.41 ± 0.90 % respectively. It was followed by treatment-B with the respective values of 7300 ± 556.77 granules/individuals, 0.90 ± 0.11 mm and 1.39 ± 0.14 %. Meanwhile, the treatment-C showed better performance as compared to treatment-D (the control) in term of fecundity that is 1367 ± 2367.13 granules/individuals. With regard to the egg production, the mean fecundity obtained ranging from 1033 ± 1789.79 to 19600 ± 450.00 granules/individuals. There were significantly differences in the mean fecundity among the treatments (p<0.05). The mean fecundity of treatment-A was considerably higher than other treatments (p<0.01). The treatment-B was significantly higher than treatment-C (p<0.05); whereas treatment-C and D was not significant different (p>0.05). There were significantly differences in the mean egg diameter among the treatments (p<0.05). The mean egg diameter was ranged from 0.20 ± 0.40 mm to 1.10 ± 0.15 mm. The maximum size of egg diameter for injected fish in the treatment-A or treatment-B was found larger than treatment-D without injection in the control group (p<0.05), indicating that Oodev had positive effect on the egg maturation of snakehead broodstock. No significant difference was observed in the egg diameter between treatment-C and D (p>0.05).

Overall, the result also clearly demonstrated that the treatment-A yielded the highest GSI rate among other treatments (p<0.01). The treatment-B was considerably higher than treatment-D (p<0.05), but not significant different from treatment C (p>0.05). There was no significantly difference in the SGI rate between treatment-C and D (p>0.05). The mean SGI rate was ranged from 0.40 ± 0.62 % to 3.41 ± 0.90 mm. The GSI rate gradually increases with increased frequency and time given. The other way the HSI rate decreases with increased frequency and time. The treatment-C showed higher HSI rate as compared to treatment-A and D (p<0.05), but not differ from treatment-B (p>0.05) was observed. No statistical difference in the mean HSI rate was found between treatment-A, B and D (p>0.05). The mean HSI rate was varied from 0.89 ± 0.03 % to 1.53 ± 0.23 %.

Table 4 shows the mean ± standard deviation of the three estimated weights i.e. body weight, liver weight and gonad weight, as well as the ratios of these parameters for each treatment. The body weight and the liver weight were ranged from 253.67 ± 21.33 to 266.80 ± 727.78 g and from 2.43 ± 0.61 to 3.68 ± 0.45 g respectively. The ratio of liver weight to body weight obtained from the treatment-C was considerably higher than that of treatment-A and D (p<0.05), but not differ from treatment-B (p>0.05) was detected. There were no statistically significant difference in those ratios between treatment-A, B and D (p>0.05). The ratio of liver weight to body weight was varied from 0.0991 to 0.0143. The results also clearly revealed that the treatment-A yielded the highest gonad weight (9.20 ± 2.50 g) among other treatments (p<0.05). The ratio of gonad weight to body weight was ranged from 0.0050 to 0.0345. Furthermore, the highest ratio of fecundity to body weight was 73.44 (treatment-A), while the lowest ratio was 3.88 (treatment-D). It was similarly demonstrated, the highest ratio of fecundity to body length was 62.88 (treatment-A), while the lowest ratio was 3.32 (treatment-D). Such relationships between fecundity and body weight as well as between fecundity and body length can be seen in Figure 1.

The appropriate hormone preparation should be selected on the basis of the species to be spawned and the availability of the hormones. Many factors which have impact on ability of induced spawning, include: 1) condition of the fish, 2) stage of sexual maturity, 3) size of the fish, 4) previous spawning history, 5) water temperature, 6) season of the year and 7) dosage of hormone use (Rottmann et al., 1991). Hormones for induced spawning have been widely used in air-breathing fishes such as human chorionic gonadotropin (HCG) (Mollah and Tan, 1983; Inyang and Hettiarachchi, 1994), luteinizing hormone releasing hormone analogue (LHRHa) (Fermin, 1992), and ovaprim (Alok et al., 1993; Haniffa et al., 1996). Oodev is one solution when fish can not perform vitellogenesis and spermatogenesis.

The oodev hormonal treatments have been successfully used for advancing the process of gonad rematuration period in A. testudineus for 12-24 days (Sari, 2015). Furthermore, Agustinus (2016) reported that P. hypophthalmus being injected with oodev 0.7 ml kg⁻¹ fish weight every 7 days for 4 weeks showed the best performance in term of growth rate and egg diameter and also can accelerate gonad rematuration of them for 28 days after spawning. Firiati et al. (2018) affirmed that oodev hormone dose of 1 ml kg⁻¹ fish weight was effective for gonad rematuration of O. hasselti broodstock within 17 days with the GSI rate of 14.48 %, average egg diameter of 1.035 mm and body weight of 31.25 mm. For the time being, the effectiveness of oodev works in snakehead is still questionable due to lack of available evidence. Dealing with the presence of oodev itself, it is commonly
known that oodev hormone is a combination of Pregnant Mare's Serum Gonadotropin (PMSG) hormone and Antidopamine (AD) compounds. PMSG hormone contains follicle stimulating hormone (FSH) and luteinizing hormone (LH) (Moore and Ward, 1980). The role of FSH increases the chances of egg cell maturation, while the role of LH stimulates ovulation. PMSG chemical structure is similar to the FSH and LH, whereas FSH structure is greater than LH (Reeves, 1987). The PMSG hormone in oodev is thought to have triggered the gonad maturation of brood fish, where vitelogenesis only takes place when a hormone is present associated with this process. The gonadotropin hormone of PMSG injected from the outside will work on the egg by maturing the existing follicle by stimulating the growth of interstitial cells and the formation of luteal cells. The process of gonadal rematuration starts from the synthesis of vitelogenin which is the precursor of egg yolks (Wiegand, 1982). One of the conditions governing vitelogenesis is the availability of hormones associated with vitelogenesis in the body. It is known that there is a positive correlation between gonadal steroid hormone and vitelogenesis (Mackenzie et al. 1998)

It was clear from our findings that snakehead successively induced with 0.5 ml kg⁻¹ fish weight for three-frequency and time periods in the treatment-A produced more eggs about 3 times higher than twice, or about 14 times higher than once or about 19 times higher than with no injection. It was significantly different from other treatments (p<0.05). The fecundity of fish varied according to the size (length-weight), age, species, food availability and season. Larger fish tends to have greater fecundity than small fish. For a given size, females in better condition exhibit higher fecundity (Kjesbu et al., 1991). The relationship between the age of brood female and the size of the egg is the square. The young mother spawning for the first time produces small eggs (about 0.8 mm diameter); the productive females will produce large eggs (about 1 mm) and old females again producing small eggs (about 0.8 mm). A relationship between length, weight, and fecundity for the family Channidae was well-documented in C. gachua (Gaikwad et al., 2009; Widodo et al., 2013), C. striatus (Islam et al., 2013; Sakhare, 2015), and C. Limbata (Khomsab and Wannasri, 2017). Figure 1 clearly shows that the ratio of fecundity to body weight as well as the ratio of fecundity to body length increases proportionally to the time periods of oodev injections given. The longer time period of oodev injections, the higher the ratio of fecundity to body sizes. It also implies that the larger fish the higher ratio of fecundity to body sizes. Such relationships were expressed in the following equations i.e. $y = 3.578 x^{2.0343}$ and $y = 3.064 x^{2.0343}$ with the coefficient of determination ($R^2$) was 0.9748, suggesting that approximately 97% of the variation in the fecundity data could be explained by the body weight of the fish. In practical, when researchers endeavor to evaluate the commercial potential of fish stocks, the fecundity estimation must consider a variety of attributes, including size at first sexual maturity, duration of spawning season, daily spawning behavior and spawning fraction (El-Drawany, 2013).

Snakehead fish brood injected with oodev 0.5 ml kg⁻¹ fish weight in the treatment-A showing the biggest egg diameter and was significantly different from other treatments (p<0.05). However, the average egg diameter of C. striata (1.53 mm) in Malaysia and India (Ghaedi et al., 2013; Sakhare, 2015.) or Ophiocephalus striatus (1.60 mm) in Mekong River, Vietnam (Long et al., 2002). The vitelogenesis process can be seen from the value of the enlarged egg diameter. It is the process of vitelogenin synthesis in the liver which is the main precursor of yolk, resulted in the egg diameter increases. The diameter of the eggs increases as the weight of the gonads increases. The sexual maturity in fish is characterized by the development of egg diameter and through the distribution of its egg size (Kuo et al., 1974). The egg size can be expressed in many ways. Single diameter is commonly used, but the longest diameter (i.e. egg length and egg width) is also occasionally used. Other egg sizes include the volume of eggs, wet weight and dry weight. From an energetic standpoint, the best term for egg size is the equality of egg calorie (energy content per egg or joule per egg), because it shows the energy available to the developing embryo (Ginzburg, 1972). The large egg size is a guarantee of higher survival (Kamlar, 1992), this because the egg contains food reserve to be used by fish larvae for survives. Larvae derived from the large egg will have more egg yolk reserves as energy source before getting food from outside. The size of the egg diameter can determine the quality of the yolk content. The egg diameter is the accumulation of the vitelogenesis process that is the absorption of vitelogenin which is going to prospective yolk. Increasing the yolk granules in the number and size resulted in the oocyte volume will be greater until the maximum size then the egg is in “dormant” phase. Egg quality is affected by both internal factors (e.g. brood age, brood size and genetic) and external factors (e.g. feed, temperature, light, density, and population). The first spawning female fish produce small diameter eggs. The egg diameter increases clearly when the second spawning and the rate of increase are slower in subsequent spawning.

The results also confirmed that snakehead consecutively injected with oodev 0.5 ml kg⁻¹ in the treatment-A increasing the GSI rate about 2.5 times higher than treatment-B, or about 8.5 times higher than treatment-C or
about 7 times higher than with no injection in treatment-D. It is clearly revealed that the GSI increases proportionally towards the time periods of oodev interventions. Also, an increase of GSI value is in conjunction with the increased doses of Oodev hormone given (Fitriatin et al., 2018). It is alleged that FSH and LH activity in oodev hormone influences the gonad development as a whole (Moore and Ward, 1980). The GSI of family Chamidae varies according the type of species (Gaikwad et al., 2009; Kapil et al., 2011; Widodo et al., 2013; Tiwari et al., 2014). For example, the GSI of C. striata (3.41 %) in the present study was lower than GSI of C. gachua (6.00 %) in Godavari River, India (Gaikwad et al., 2009) or C. marulius (47.56 %) in Son River Shahdol, India (Tiwari et al., 2014), or C. striatus (8.00 %) in Badin Sindh District, Pakistan (Narejo et al., 2015). The IGS value is used to predict when the fish will be ready for spawning. Increased IGS values can be triggered by oocytes, while vitelogenin is main precursor of yolk which is the main component of the growing oocyte. When the process of vitelogenesis is continuing, the yolk granules increase in number and size, so that the volume of the oocyte enlarges and eventually will lead to increased value of GSI. The process of vitelogenin formation starts from the presence of environmental factor cues such as fotoperiode, temperature, eating activity, and other factors that will all stimulate the hypothalamus to secrete gonadotropin releasing hormone (GnRH). GnRH will be secreted into the blood stream will stimulate hypophysis to secrete gonadotrop in hormones (Darwisito et al., 2006). In the reproductive cycle, GSI increases with the maturation process, whereas the HSI in contradicting to GSI (Lodeiros et al., 2001). It was clear from our findings that the treatment-A produced the highest GSI among other treatments (3.41 %), contrariwise it provided the lowest HSI among other treatments (0.89 %) in the same amount of oodev content. In other words, the longer time period of oodev injection given, the lower the ratio of liver weight to body weight gained. This is attributable to the use of energy reserves derived from the liver instead of energy sourced from the body. The HIS of C. striata (1.53 %) in the present study was lower than HSI of C. punctatus (1.64 %) in Syilhet, Bangladesh (Hossain 2013), but slightly higher than C. striatus (1.40 %) in Penang, Malaysia (Ghaedi et al., 2013). In addition, Bijaksana (2006) argued that most of the snakehead caught during fishing season is allocating their somatic growth for reproductive growth. In line with this; temperature, pH and DO during sampling period are considered to be comfortable for snakehead fish broods.

IV. CONCLUSION

Oodev hormone had a significant effect in speeding up the process of gonadal rematuration in the snakehead injected. Oodev dose of 0.5 ml kg$^{-1}$ those given in 3 times/9 days (treatment-A) is the most effective treatment to rematurate the gonad of snakehead female broodstock within 30 days, producing fecundity of 19,600 granules/individuals, egg diameter of 1.10 mm, and GSI rate of 3.41 %. The results may applicable when outdoor environmental quality is improved. Despite these findings, the outdoor environmental quality should be considered into account. Some of the information presented here may be applicable in other geographical areas.

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Table 1: Tools, instruments and materials used in snakehead gonad rematuration experiment

| No. | Tools and Materials                  | Size/Merk          | Utility                                      |
|-----|-------------------------------------|--------------------|----------------------------------------------|
| 1.  | Concrete pond                       | 4x7x1.2 m          | Placement of hapa                            |
| 2.  | Small hapa 12 units                 | 0.4x0.3x1m         | Placement of fish brood tested               |
| 3.  | Large hapa 2 units                  | 1.5x1.5 m          | For brood fish reserves                      |
| 4.  | Waterchecker U10                    | Horiba             | Measurement of water quality                 |
| 5.  | Scoop net                           | 30 cm              | For collecting the fish samples              |
| 6.  | Name label                          | 15x10 cm           | Mark off hapa                                |
| 7.  | Digital balance                     | ACS                | Weighing of fish samples                     |
| 8.  | Ruler                               | 30 cm              | Measurement of body length                   |
| 9.  | Washbasin                           | 50 cm              | Container for brood fish                     |
| 10. | Knife                               | 10 cm              | Dissection of the selected brood fish        |
| 11. | Rope                                | 10 m               | Hapa installation                            |
| 12. | Injection                           | 1 ml               | Oodev injection                              |
| 13. | Snakehead brood                     | 48 fish            | Experimental fish sample                     |
| 14. | Frogs                               | 4% of body weight/day | Feed/bait served for snakehead              |
| 15. | Oodev hormone                       | 0.5 ml kg⁻¹ fish weight | Dose of oodev to be injected to snakehead fish brood samples. |

Table 2: The experimental treatments for investigating the snakehead gonad rematuration

| Treatment | Oodev dose/fish weight | Frequency | Time period          |
|-----------|------------------------|-----------|----------------------|
| A         | 0.5 ml kg⁻¹            | 3 times   | Day-3, day-6, day-9  |
| B         | 0.5 ml kg⁻¹            | 2 times   | Day-3, day-6         |
| C         | 0.5 ml kg⁻¹            | 1 time    | Day-3                |
| D         | No injection           | 0 time    | Control              |
Table 3: The mean ± standard deviation of the parameters observed for each treatment.

| Treatment | Fecundity (granule/individual) | Egg diameter (mm) | GSI (%) | HSI (%) |
|-----------|-------------------------------|-------------------|---------|---------|
| A         | 19600 ± 450.00 **             | 1.10 ± 0.15       | 3.41 ± 0.90 ** | 0.89 ± 0.03 |
| B         | 7300 ± 556.77                | 0.90 ± 0.11       | 1.39 ± 0.14 *  | 1.11 ± 0.03 |
| C         | 1367 ± 2367.13               | 0.20 ± 0.40       | 0.40 ± 0.62    | 1.53 ± 0.23 * |
| D         | 1033 ± 1789.79               | 0.20 ± 0.40       | 0.50 ± 0.54    | 0.93 ± 0.09  |

Significance level: ** (p<0.01), and * (p<0.05)

Table 4: The mean ± standard deviation of the estimated weights and their ratios for each treatment. A = three time injections, B = two time injections, C = once injection, and D = no injection (control).

| Treatment | A | B | C | D |
|-----------|---|---|---|---|
| Body weight (g) | 266.8 ± 727.78 | 253.67 ± 21.33 | 258.03 ± 10.13 | 266.57 ± 11.60 |
| Liver weight (g) | 2.43 ± 0.61 | 2.84 ± 0.38 | 3.68 ± 0.45 | 2.48 ± 0.16 |
| Gonad weight (g) | 9.20 ± 2.50 | 3.83 ± 0.31 | 2.10 ± 1.49 | 1.33 ± 1.53 |
| Ratio of liver weight to body weight | 0.0091 | 0.0112 | 0.0143 * | 0.0093 |
| Ratio of gonad weight to body weight | 0.0345 * | 0.0151 | 0.0081 | 0.0050 |

Significance level: ** (p<0.01), and * (p<0.05)

Table 5: Comparative fecundity, egg diameter, GSI and HSI of family Channidae from different geographical areas

| Species      | Fecundity (granule/individual) | Egg diameter (mm) | GSI % | HSI % | Locations       | Country | References           |
|--------------|--------------------------------|-------------------|-------|-------|-----------------|---------|----------------------|
| Channa striata | 1,033 - 19,600 | 0.20 - 1.10 | 0.40 - 3.41 | 0.89 - 1.53 | Sungai Batang | Indonesia | Present study        |
| C. striata   | - | 1.00 - 1.60 | - | - | Danau Panggang | Indonesia | Fitriliyani, 2005   |
| C. striatus  | 28,332 - 41,068 | 1.21 - 1.33 | 10.4 - 11.5 | 0.70 - 1.40 | Penang | Malaysia | Ghaedi et al., 2013 |
| C. striatus  | 3,000 - 12,000 | 0.70 - 1.30 | 2.10 - 8.00 | - | Badin Sindh | Pakistan | Narejo et al., 2015 |
| C. striatus  | 4,900 - 14,028 | 1.53 | - | - | Beed District | India | Sakhare, 2015 |
| C. punctatus | 2,538 - 32,987 | - | 0.53 - 5.34 | - | Syilhet ditches | Bangladesh | Mian et al., 2017 |
| C. punctatus | 2,116 - 11,332 | - | - | - | Pond | India | Marimuthu and Haniffa, 2006 |
| C. punctatus | 3,678 - 27,853 | - | - | - | Varuna River | India | Lalita et al., 2011 |
| C. punctatus | 2,654 - 26,294 | - | 0.19 - 5.64 | 0.84 - 1.64 | Syilhet | Bangladesh | Hossain, 2013 |
| C. gachua    | - | - | 5.57 - 6.00 | - | Godavari River | India | Gaikwad et al., 2009 |
| C. gachua    | 2,539 - 7,194 | - | - | - | Bhubaneswar | India | Mishra, 1991 |
| C. gachua    | - | - | 5.31 - 5.63 | - | Dinoyo District | Indonesia | Widodo et al., 2013 |
| C. limbata   | 956 - 4,652 | 1.96 - 3.74 | - | - | Ta Bo - HuaiYai | Thailand | Khomsab and Wannasri, 2017 |
| C. marulius  | - | - | 8.21 - 47.56 | - | Son River | India | Tiwari et al., 2014 |
| Ophiocephalus striatus | - | 0.20 - 1.60 | - | - | Mekong River | Vietnam | Long et al., 2002 |
Fig. 1: The ratio of fecundity to body weight (top) and the ratio of fecundity to body length (bottom) increase proportionally to the time periods of oodev intervention at the same amount of 0.5 ml kg⁻¹ fish weight.

The treatment-A showed the highest ratios among other treatments (p<0.05).

A = three time injections, B = two time injections, C = once injection, and D = no injection (control).