

**KOMBINASI INJEKSI sGnRH-a + ad , PADA KETINGGIAN AIR YANG BERBEDA UNTUK PEMIJAHAN SNAKEHEAD , *Channa striata* DI WADAH BUDIDAYA**

**COMBINATION OF INJECTION sGnRH-a + ad, and WATER LEVEL FOR SPAWNING SNAKEHEAD, *Channa striata* IN CONCREAT TANK**

<sup>1)</sup>Untung Bijaksana, <sup>1)</sup>Septiodrian R. Balantek

<sup>1)</sup>Aquaculture Departement Faculty of Fisheries University of Lambung Mangkurat

**ABSTRAK**

Penelitian ini bertujuan untuk melihat efektivitas sGnRH-a+ad dan ketinggian air yang berbeda pada pemijahan ikan snakehead, *Channa striata* dalam tangki concreat. Penelitian ini dilakukan di Laboratorium Basah Program Studi Budidaya Perairan Fakultas Perikanan Universitas Lambung Mangkurat . Ada dua faktor utama sebagai perlakuan yaitu: Faktor A = dosis sGnRH -a + ad dengan tiga tingkat ( A1 = 0,2 ml / kg , A2 = 0,4 ml / kg , A3 = 0,6 ml / kg ) dan faktor B = tinggi air dengan tiga tingkat ( B1 = 15 cm; B2 = 20 cm; B3 = 25 cm ) diulang 4 kali untuk mendapatkan 36 unit percobaan. Pengamatan dilakukan pada parameter : berat badan, berat gonad, berat hati , estradiol - 17 $\beta$  , diameter telur, IGS, IHS, fekunditas, pemupukan dan derajat penetasan. Terdapat Hubungan yang sangat erat antara parameter pengamatan IGS dan IHS. Perlakuan yang diberikan waktu laten diperoleh kisaran antara 12 jam sampai 25 jam, fekunditas antara 5047-5072 butir, derajat fertilisasi antara 60-60,3 persen dan 76,6 derajat fertilisasi antara 83,9 persen. Perkembangan setelah menetas mencapai panjang tubuh 3,5 mm dan 45,5 mm pada hari 20 .

Kata kunci : sGnRH –a+ad , tingkat air , snakehead

**ABSTRACT**

This study aims to look at the effectiveness of injection of sGnRH-a + ad and height of water on fish spawning snakehead, *Channa striata* in the concreat tank. The research was conducted at the Wet Laboratory Aquaculture Departement Faculty of Fisheries University of Lambung Mangkurat. There are two main factors as treatment namely: Factor A = dose of sGnRH-a + ad with three levels (A1 = 0.2 ml / kg; A2 = 0.4 ml / kg; A3 = 0.6 ml / kg) and factor B = height of water with three levels (B1 = 15 cm; B2 = 20 cm; B3 = 25 cm) made repeated 4 times to obtain 36 units of the experiment. Observations made on the parameters: body weight, gonad weight, liver weight, estradiol-17 $\beta$ , egg diameter, IGS, IHS, fecundity, fertilization and hatching degrees. The strongest relationship obtained from the treatment of

observational parameter is IGS and IHS. Of treatment given the latent time of the obtained range between 12 hours to 25 hours, fecundity between 5047 to 5072 grains, the degree of fertilization between 60 to 60.3 percent and 76.6 degrees fertilization between 83.9 per cent up. Developments after hatching body length of 3.5 mm and 45.5 mm on day 20.

Keywords: sGnRH-a + ad, the water levels, snakehead

## INTRODUCTION

Snakehead, *Channa striata* (Bloch, 1793) is one of the commodities that live in fresh water that has high economic value and is also required as a treatment. While it is known that the spawning fish are very much depending on the season and needs just rely on fishing effort in the wild. Gonad maturation efforts in the cultivation container with hormonal stimulus ever attempted in the catfish, Belida, balashark, jambal, *Claris batrachus* and baung (Zairin et al. 1992; 1996; 1997; 2001; Zairin 2002; Sularto 2002 ; Isriansyah 2005; Syarifuddin 2005), conjoined jambal, *Pangasius hypophthalmus* (Ernawati 1999), catfish, snakehead, *Channa striata* (Fitriliyani 2005), baung, *Hemibagrus nemurus* (Supriyadi 2005) and *Channa striatus* (Bloch 1793) (Hossain et al., 2008).

Fish spawning with snakehead to make a nest in the water around the plant or on the outskirts of the fast-

flowing waters of a shallow weak, can spawn with the parent about 9 months of age at a size of about 21 cm (Haniffa et al., 1996). In Sri Lanka snakehead spawn several times a year, while in the Philippines snakehead can spawn every month. Snakehead spawning season in Thailand between May and October, with a peak in July through September (Wee 1982). In the Mekong River Delta, a female fish first mature snakehead (Long et al., 2002). In Central Java Kedongombo Reservoir, the fish began to mature female sex snakehead on the total length of 18.5 cm (Kartamihardja 1994). In Bangladesh, the fish can spawn in the snakehead with the provision of container cultivation hypophysis gland (Hossain et al., 2008). Snakehead in one spawning season to spawning 2-3 times (Bijaksana, 2006).

Somatic index Gonado snakehead in a container of aquaculture is the highest in April  $3:59 \pm 0:09$ ,  $3:33 \pm 0:07$  in May and June,

3:48 ± 0:10 and July are the lowest is 2.75 ± 0.08 (Bijaksana, 2008). Naturally spawning time has passed but the snakehead with a range of circumstances Gonado somatic indices as above should be spawning can take place. Loss of "signal" as the trigger ovulation injection treatment given sGnRH-a + ad with multiple doses and multiple levels of the water level so that it can be proved that the snakehead spawning can take place in a container cultivation.

This study aims to see the effect of giving sGnRH-a + ad, combined with the water levels of spawning fish in concreat tank.

## MATERIALS AND METHODS

### *Time and Place*

The experiment was conducted at the Wet Laboratory Aquaculture Department of Fisheries Faculty of the University of Lambung Mangkurat Banjarbaru began in April 2009 to June 2009.

Treatment is the injection given sGnRH-a + ad (dose) and water level (level), the observations were made on body weight, gonad weight, liver weight, estradiol-17β, egg diameter, IGS, IHS, fecundity, fertilization and

the hatching degrees spawning fish in concreat tank.



Figure 1. Snakehead used in the study

### *The design of experiments*

To determine the effect of injection of sGnRH-a + ad and height of the water against the spawning fish in concreat tank with cultivation used a factorial completely randomized design with combination treatment (Table 1), as follows:

Table 1. Combination treatment in the study

Water Level (B)	Injection sGnRH-a+ad (A)		
	0,2 ml / kg (A1)	0,4 ml / kg (A2)	0,6 ml / kg (A3)
15 cm (B1)	A1B1	A2B1	A3B1
20 cm (B2)	A1B2	A2B2	A3B2
25 cm (B3)	A1B3	A2B3	A3B3

Each treatment was repeated four times to obtain 36 units of the experiment. Each treatment combination represents an experimental unit, ie one pair of parent snakehead but only female fish are given an injection treatment of sGnRH-a + ad.

### ***sGnRH-a+ad***

sGnRH-a + ad is to stimulate spawning fish products produced by Syndel Laboratory, Vancouver, Canada. Ovaprim containing 20 g D-Arg6, Pro9-Net sGnRH and 10 mg domperidone per ml of propylene glycol (King & Young, 2001). In addition to anti-dopamine, ova-RH is a peptide contained in ovaprim has been shown to be effective from the development of peptide analogues (Anonymous 2004).

### ***Implementation Procedures***

Fish catches in natural snakehead adapted and maintained in cement tanks with a given feed and water quality are conditional. The feed is given as much as 4% of body weight / day with a frequency of 4 times / day.

Injections made after weighing the prospective parent locations to determine the dose used. Mains to be injected at the head covered with a

damp cloth. Injection done twice intramuscularly behind the dorsal fin to the left and right. The first injection of as much as 1/3 of the total dose administered and the interval of 10 hours is given a second injection of as much as 2/3 which is the remainder of the first injection dose.

Blood sampling performed at the base of the tail by using 2 ml syringes that had been given heparin. The content of estradiol-17 $\beta$  was measured with a kit COAT-estradiol-17 $\beta$ -made account Diagnostic Product Corporation Los Angeles, USA. And quantitative measurements performed using 125I radioactive substances. (Nur, et al., 1992, Bintang, 2006). Disentrifius blood samples at 5000 rpm for 5-10 minutes. Plasma collected and stored at -20 ° C, while waiting for the measurement of Radio Immuno Assay (Rouger & Liley, 1990; Zanuy et al. 1999).

Egg diameter was measured by taking samples of the egg by way of surgery every 30 days. Eggs were taken, fixed with buffered formalin solution. Subsequently made an egg diameter frequency distribution (Tamaru, et al., 1991).

Fecundity was calculated based on the weight of individual eggs per gonad (g) multiplied by the number of

egg samples (grains) divided by the weight of the egg sample (g) further in the individual body weight (g). To find out the end of the IGS and IHS research performed surgery on some of the test fish. IGS and IHS was determined using the formula proposed by Crim & Glebe (1990)

### **Data Analysis**

Data from the combined treatment is displayed in graphical form so that the picture looks the best results in the observation, namely: body weight, gonad weight, liver weight, estradiol-17 $\beta$ , egg diameter, IGS, IHS, fecundity, fertilization and the degree of hatching degrees.

To determine the effect of induction of sGnRH-a + ad and height of water on the observation parameters, was tested using variance (ANOVA). To find the best treatment DUNCAN Further follow-up test conducted to determine the response patterns generated test followed by orthogonal polynomial contrasts (Gomez & Gomez 1995). Data processing for statistical testing used SPSS 15.0 program.

## **RESULTS**

Injection of a combination of observations sGnRH-a + ad and the water levels are presented in Table 2. Indicators of reproductive aspect is body weight, gonad weight, liver weight, IGS, HIS, estradiol-17 $\beta$ , egg diameter, fecundity, fertilization and the hatching degrees.

Results of analysis of the weight range of injectable treatments gonat of sGnRH-a + ad and height of water level indicates that the treatment A3B1, A2B2, A2B3, A3B3 was not significantly different but significantly different to the treatment A1B3, A1B1, A3B2, A2B1 and A1B2. Treatment A2B3, A3B3, A1B3, A1B1, A3B2, A2B1 and A1B2 are not significantly different. ( $P < 0.05$ ), with a value of  $r^2 = 0.500$  (Figure 2).

Table 2. Average value of the experimental observations of reproductive status

Treat	Body Weight /ek/gr	Gonad Weight /ek/gr		Hevato Weight/ek/gr		IGS (%)		HIS (%)		E2 (pg/ml)	
A1B1	250.00	4.63	± 0.05	2.10	± 0.08	1.84	± 0.03	0.84	± 0.03	195.55	± 3.74
A2B1	248.75	4.63	± 0.00	2.10	± 0.10	1.86	± 0.00	0.84	± 0.04	197.83	± 9.54
A3B1	247.50	4.63	± 0.00	2.13	± 0.06	1.87	± 0.00	0.86	± 0.02	195.33	± 4.10
A1B2	246.25	4.63	± 0.00	2.15	± 0.13	1.87	± 0.00	0.87	± 0.05	191.48	± 4.86
A2B2	245.00	4.60	± 0.00	2.18	± 0.05	1.87	± 0.00	0.88	± 0.02	187.55	± 4.52
A3B2	242.50	4.63	± 0.08	2.20	± 0.10	1.90	± 0.03	0.90	± 0.04	187.55	± 4.57
A1B3	240.00	4.65	± 0.05	2.23	± 0.05	1.94	± 0.02	0.92	± 0.02	187.54	± 3.06
A2B3	237.50	4.68	± 0.06	2.20	± 0.05	1.97	± 0.02	0.92	± 0.02	191.39	± 0.17
A3B3	235.00	4.70	± 0.06	2.15	± 0.13	2.00	± 0.03	0.91	± 0.05	195.31	± 2.46

Treat	Egg Diameter (mm)		Fecundity (butir)		Fertilization		Hatching	
A1B1	1.48	± 0.05	5047.50	± 6.4 5	3043.75	± 12.50	2552.50	± 6.45
A2B1	1.43	± 0.06	5053.75	± 6.4 5	3041.25	± 6.29	2548.75	± 11.90
A3B1	1.38	± 0.05	5060.00	± 4.7 9	3042.50	± 12.91	2548.75	± 2.89
A1B2	1.30	± 0.05	5067.50	± 6.4 5	3046.25	± 2.89	2550.00	± 4.08
A2B2	1.25	± 0.05	5072.50	± 4.7 9	3048.75	± 6.45	2547.50	± 6.45
A3B2	1.30	± 0.00	5070.00	± 10. 31	3043.75	± 6.45	2495.00	± 9.13
A1B3	1.33	± 0.05	5066.25	± 9.5 7	3041.25	± 7.50	2438.75	± 10.80
A2B3	1.38	± 0.05	5063.75	± 4.0 8	3042.50	± 4.79	2383.75	± 4.08
A3B3	1.43	± 0.00	5061.25	± 5.0 0	3045.00	± 2.50	2332.50	± 4.79

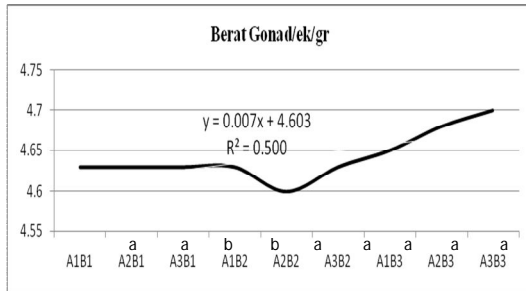


Figure 2. Injection of sGnRH-a relationship + ad and height of the water to the weight of gonads

IGS analysis results of the injection of a dose range of sGnRH-a + ad showed that treatment of 0.6 ml / kg was significantly different to the treatment of 0.4 ml / kg and 0.2 ml / kg. Treatment of 0.4 ml / kg was significantly different to the treatment of 0.2 ml / kg. Furthermore the height of 20 cm water were significantly different to the treatment 25 cm and 15 cm height 25 cm, but the treatment and 15 cm are not significantly different. ( $P < 0.05$ ). A2B1, A1B3 and A1B1 are not significantly different with  $r^2 = 0.889$ . (Figure 3).

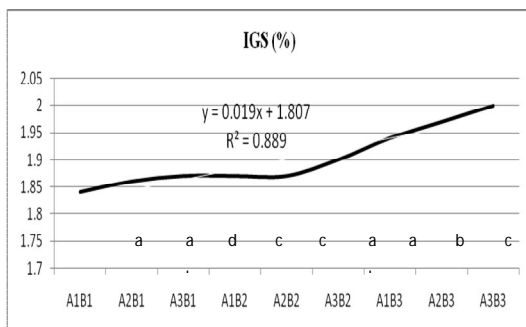


Figure 3. Relationship sGnRH-a + injection ad and the water levels of IGS

Results of analysis of IHS variety of injection dose combination treatment sGnRH-a + ad and height of the water showed that the treatment A3B1, A3B3, A1B2, A2B3, A2B1, A1B3, A3B2 and a2b2 are not significantly different but significantly different to the treatment A1B1. Treatment A2B3, A2B1, A1B3, A3B2, A1B1 a2b2 and not significantly different with  $r^2 = 0.917$ . (Figure 4).

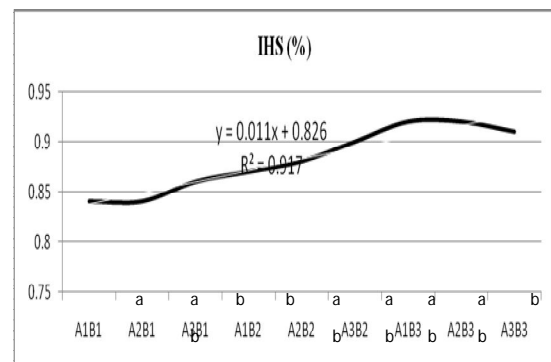


Figure 4. Relationship sGnRH-a + injection ad and the water levels of IGS

Results of analysis of various treatment combinations of E2 on sGnRH-a-dose injection + ad and height of the water showed that the treatment of A1B1 and A3B1 are not significantly different but significantly different to the treatment A2B1, A2B2, A1B2, A3B2, A1B3, A2B3 and A3B3. Treatment A2B1 A2B2 significantly different to the treatment, A1B2, A3B2, A1B3, A2B3 and A3B3. A2B2 and A1B2 treatment was not significantly

different but significantly different to the treatment A3B2, A1B3, A2B3 and A3B3. A3B2 and A1B3 treatment was not significantly different but significantly different to the treatment of A2B3 and A3B3. A2B3 and A3B3 treatment was not significantly different with  $r^2 = 0.205$ . (Figure 5).

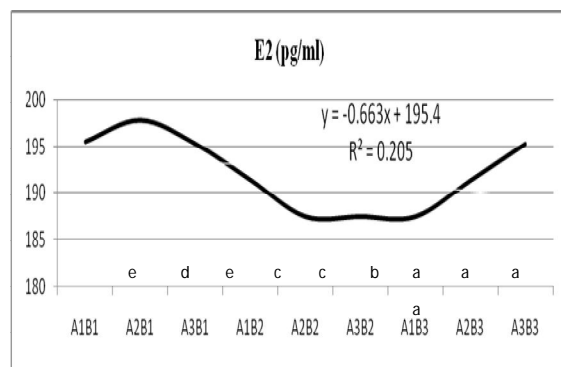


Figure 5. Relationship sGnRH-a + ad injection and the water levels of estradiol-17 $\beta$  concentrations

Results of analysis of egg diameter range of doses of injectable treatments sGnRH-a + ad showed that treatment of 0.2 ml / kg and 0.6 ml / kg was not significantly different but significantly different to 0.4 ml / kg. Further treatment of the water level indicates that the treatment was significantly different height of 15 cm to 25 cm height treatment and 20 cm. Treatment of 25 cm and 20 cm are not significantly different.

Results of analysis of various treatment combinations of egg

diameter and height of the water ovaprim showed that treatment of A1B1 and A3B1 are not significantly different but significantly different to the treatment A2B1, A2B3, A2B2, A1B3, A1B2, A3B3 and A3B2. Treatment was significantly different to the treatment A2B1 A2B3, A2B2, A1B3, A1B2, A3B3 and A3B2. Treatment A2B3, A2B2, A1B3, A1B2, A3B3 and A3B2 are not significantly different with  $r^2 = 0.075$ . (Figure 6).

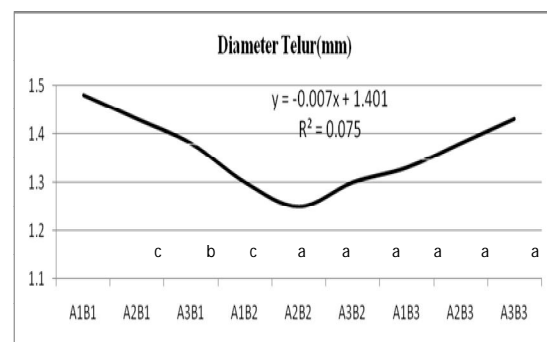


Figure 6. Injection of sGnRH-a relationship + ad and height of water on egg diameter

Results of analysis of various treatment combinations of fecundity of the water levels ovaprim and A2B1 showed that the treatment was significantly different to the other treatments. Treatment A1B2, A3B1, a2b2, A3B2, A1B3 and A3B3 are not significantly different but significantly different to the treatment of A2B3 and A1B1. Treatment A3B2, A1B3, A3B3 and A2B3 not significantly different but



significantly different to the treatment A1B1. Treatment A1B3, A3B3, A2B3 and A1B1 are not significantly different with  $r^2 = 0.329$ . (Figure 7).

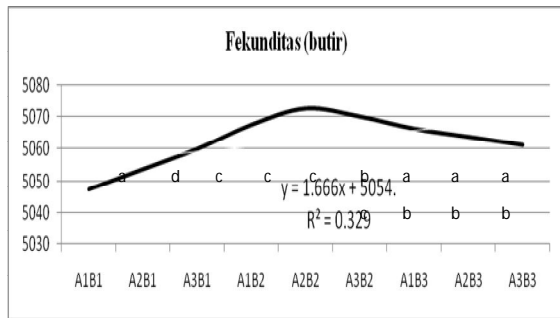


Figure 7. Relationship sGnRH-a + injection ad and the water levels of fecundity

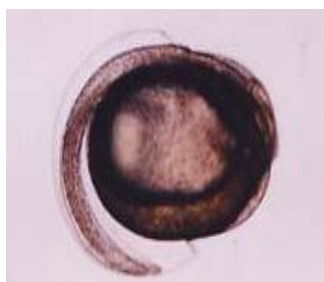
Observations of larval development after Hatching (0 hours, 4 hours, 8 hours, 14 hours, 36 hours, 48 hours, 3 days until the day of the 20) carried out by measuring the total body length as shown in Table 3.

Table 3. *Channa striata* Blkr larval development

After hatching	0	4	8	14	36	48	3 hr	6 hr	9 hr	15 hr	20 hr
Body size (mm)	3.5	3.6	3.9	4.2	5.1	5.5	5.9	7.9	12.5	17.3	45.5

Spawning the larvae obtained immediately separated from its parent and maintained in an aquarium with the water level 5 cm.

Hatching observation of current developments daily up to age 20 are presented in Figure 8.



Hatching time



four hours age



eight hour age



eleven hours age



twenty-five hours age



twenty days age

Figure 8. *Channa striata* Blkr larval development

Data latency time. time of ovulation, fecundity, fertilization and the degree of hatching after injection of sGnRH degrees are presented in Table 4.

Table 4. Latent time after injection of sGnRH-a + ad with a high water

Treat	TKG	Latensi Time (jam)	Fecundity (butir)	Fertiliation (%)	Hatching (%)
A1B1	IV	25	5047	60.3	83.9
A2B1	IV	18	5053	60.2	83.8
A3B1	IV	15	5060	60.12	83.8
A1B2	IV	20	5067	60.11	83.7
A2B2	IV	17.5	5072	60.1	83.6
A3B2	IV	14	5070	60.0	81.9
A1B3	IV	23	5066	60.03	80.2
A2B3	IV	19	5063	60.08	78.3
A3B3	IV	12	5061	60.16	76.6

## DISCUSSION

Reproduction of fish is under the control of the hypothalamic-pituitary-gonadal. Some of the factors involved in the reproduction of fish, namely environmental signals, the hormonal system and reproductive organs. Hormonal control of reproductive system in fish can be divided into two main factors, namely gonad maturation, and ovulation and

spawning (Aida et al, 1991 in the Zairin 2003).

Based on observations during the study note that the mother was able to ovulate. This suggests that the hormone sGnRH-a + ad with several levels of doses can stimulate the final maturation process. Use of ovaprim lowest dose 0.4 ml / kg with a height of 15 cm of water to give results that are not significantly different from other treatments.

According to Bijaksana (2008), corm fish weighing about 250 g per fish in natural waters can produce about 3000 eggs in a container grain cultivation, but only about 2000 eggs. On treatment provision sGnRH-a + ad with a dose of 0.2, 0.4 and 0.6 ml / kg, the fertilized egg was about 60 percent and a hatch about 80 percent.

Furthermore Hossain et al. (2008), argued that the use of the pituitary at a dose of 45 mg / kg on the first injection and 80 mg / kg on the second injection fecundity yield approximately 17 273 grains with about 58 percent of fertilization and fertilization about 62 percent by weight of the parent 0.5 - 0.6 kg. Caught from the wild parent in June with a heavy 600-700 gr produce 95 to 99 percent of fertilized (Fitriliani 2005).

Final stages of egg maturation is characterized by the position of cell nuclei in the animal pole, while the eggs are still in a dormant phase, or not having the final maturity is characterized by the position of the cell nucleus was still in the middle (Nagahama 1983, Billard 1992 and Harvey & Carolsfeld. 1993 ).

The success of ovulation with some ovaprim administration by injection treatment indicated that the development of fish eggs snakehead

as a test fish, has been in a mature phase. Fish that are kept in a container snakehead aquaculture is able to adapt to the environment and the feed given. After the age of 10 days of larval fish can be reared in ponds snakehead enlargement (Marimuthu & Hanifa., 2007).

Natural process of ovulation and spawning, environmental signals received by the central nervous system and forwarded to the hypothalamus. Instead the response is the hypothalamus releases the hormone GnRH which then works on the pituitary gland. At this stage pituitary does not secrete gonadotropin-I,-II gonadotropin hormones but acting on the theca layer of the oocyte. Its effects are, theca layer will synthesize the hormone  $17\alpha$ -hydroxyprogesterone in the layer which is then converted into  $17\alpha$  granulosa,  $20\beta$ -dihydroxyprogesterone by the enzyme  $20\beta$ -hydroxy steroid dehydrogenase. Further maturation of steroid trigger will stimulate the formation of stimulus factors which led to maturation of the egg nuclei migrate toward microfil then fused. Once the core melting process, a layer of the follicle will rupture and the egg is released to the ovarian cavity in a process called ovulation (Yaron 1995).

The fulfillment of this stage of the process occurs so well that eggs can be said to have reached physiological maturity and ready to be fertilized by sperm. If the optimal maturity of the gift is an encouraging acceleration ovaprim the release of an egg. Suspected breeders ( $\pm 200$  g per fish) were used as the parent fish is cooked first test is based on maintaining it for 7 to 9 months in cages.

### **CONCLUSION**

Injection treatment of sGnRH-a + ad on all the parameters were not significantly different except at IGS parameters and egg diameter. The height of water treatment was significantly different in all parameters except the parameter gonat weight, liver weight and the IHS. Induction treatment interaction sGnRH-a + ad, The height of water in all parameters were significantly different except in liver weight parameters.

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