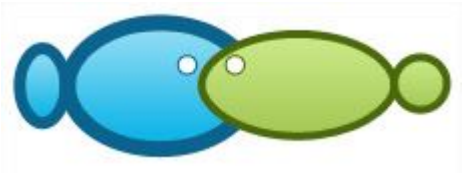

Hormonal induction for maturation, ovulation and successful level of spawning on striped snakehead (*Channa striata*)

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Abstract. Striped snakehead (*Channa striata*) is an essential commodity with potential important economic value as a consumable fish and a source of albumin-producing products. This study aimed to determine the optimal dose of hormones to be used in maturation, ovulation, and the effects of the hormones on the reproductive performance of striped snakehead in improving productivity. This study on the maturation and ovulation of striped snakehead was conducted experimentally using a completely randomized design (CRD) with 4 treatments and 3 replications. The maturation activity used 12 prospective broodstock measuring 23.75±0.80 cm in standard length and 238.08±8.66 g in weight. Striped snakeheads were marked and maintained communally in ponds measuring 3 m x 5 m with a water level of about 1 m. Maturation induction used a combination of pregnant mare serum gonadotropin (PMSG) and anti-dopamine hormones with doses as follows: (A) 2.0 mL kg^{-1}, (B) 2.25 mL kg^{-1}, (C) 2.5 mL kg^{-1} and (D) controls (without hormone induction). Hormone induction was performed once every 10 days for the 30 days of rearing. Ovulation activity, using striped snakehead female broodstock at a gonad maturation stage of GMS IV, was carried out by measuring the length (27.72±2.41 cm) and weight (336.67±39.02 g) of the fish. Ovulation induction used different doses of hormones, i.e. (A) LHRHa 0.6 mL kg^{-1}, (B) HCG 500 IU + LHRHa 0.6 mL kg^{-1}, (C) oxytocin 1 IU + LHRHa 0.6 mL kg^{-1}, and (D) controls (without hormone induction). The results of the maturation study showed that striped snakehead broodstock induced with a combination hormone PMSG + AD (2.5 mL kg^{-1}) had the highest gonadosomatic index (GSI, 3.21±0.26%) and fecundity (5.268±410 egg per fish) at the GMS IV. On the other hand, the ovulation study also showed that striped snakehead broodstock induced using a combination hormone HCG 500 IU + LHRHa 0.6 mL kg^{-1} had the fastest ovulation latency period with high ovulation rate, 21 hours 8 minutes and 100%, respectively, when compared with other treatments. The application of breeding technology on striped snakehead using hormones can accelerate the process of gonad maturation with a higher success level of spawning.

Key Words: striped snakehead, maturation, ovulation, breeding, hormone.

Introduction. Striped snakehead (*Channa striata*) is a local commodity that is spread evenly throughout Indonesia, and is found in several other countries in the Southeast Asian region (War & Altaff 2011). It has the potential to be developed as cultured fish due to both its economic value and prospective market (Gustiano et al 2015). At the national market level, the price of striped snakehead is quite promising, ranging from IDR. 60,000 to 70,000/kg in the rainy season and IDR. 40,000 to 50,000/kg in the dry season. Also, the striped snakehead is known as a high source of albumin. Albumin is a protein soluble in water which is present in blood serum. It plays a variety of roles, especially in wound healing and cancer drugs (Yuniarti et al 2013).

Striped snakehead consumption needs to increase annually. Meeting the consumption needs still relies on natural catchment. As an illustration, Data and Information Center, Ministry of Marine Affairs and Fisheries Republic of Indonesia (2013) reported that the volume of striped snakehead caught on Indonesian inland water in 2011 was 36,837 tons. However, overfishing in nature will result in a decrease in the population of striped snakehead. Nasution (2012) reported in several exploration

activities that striped snakehead was smaller in size and fewer in number during the catch.

So far, several culture activities of striped snakehead have been carried out to supress the level of capture in nature, including natural and semi-natural breeding (Hossain et al 2008; Paray et al 2013; Kusmini et al 2016; Muslim 2017; Ath-Thar et al 2017). However, the seed production result was not optimal. Increased productivity is inseparable from the reproductive performance. The development of technology to improve the efficiency of reproduction has been done through the addition of hormones to reach oocyte maturation in vivo and efficient reproduction periods. Some examples of products that can be used in maturation activities are hormone containing pregnant mare serum gonadotropin (PMSG) and anti-dopamine. These hormones contain follicle stimulating hormone (FSH) which plays a role in the early maturation of gonads or vitellogenesis. Ovulation activities were induced using a combination of hormones, luteinizing hormone-releasing hormone (LHRH) analogue and human chorionic gonadotropin (HCG), to accelerate the ovulation in striped snakehead.

Vitellogenin absorption would lead to oocyte reaching a given size to be ready for ovulation. The addition of exogenous hormones in the final stage of gonad development and spawning of striped snakehead has been carried out previously. Ath-Thar et al (2017) stated that hormones containing a combination of PMSG, anti-dopamine with a dose of 1.5 mL kg^{-1} of fish could stimulate gonadal maturity until the vitellogenesis phase with an increase in egg diameter of 0.38±0.015 mm. Those results indicated that the reproduction can still be optimized by increasing the dose. The present study aimed to determine the optimal doses of hormones to be used in maturation, ovulation, and their effects on the reproductive performance of striped snakehead in improving productivity.

Material and Method. The study was conducted from March to November 2018 at two locations in Indonesia: 1) Germplasm Research Station which is an installation unit of the Institute for Freshwater Aquaculture Research and Fisheries Extension (BRPBATPP), 2) UPPI farm of striped snakehead breeding unit in Keyudan Village, Minggir District, Sleman Regency of Yogyakarta.

Fish sample. The fish used in maturation activities were 12 prospective broodstock of striped snakehead measuring 23.75±0.80 cm in standard length and 238.08±8.66 g in weight. In ovulation activities, striped snakehead broodstock was used at a ratio of 12:12 (females and males). The used striped snakehead female broodstock was in GMS IV gonad maturation stage and measured 27.72±2.41 cm in length and 336.67±39.02 g in weight, while the male broodstock measured 20.15±10.07 cm in length and 289.21±2.12 g in weight. The selection of broodstock was carried out based on the completeness of morphology and the level of gonadal maturity.

Maturation of striped snakehead broodstock. This study on the maturation of striped snakehead was conducted experimentally using a completely randomized design (CRD) with 4 treatments and 3 replications. Striped snakehead was reared communally in a pond measuring 3 m x 5 m with a water level of 1 m. The fish were first marked using a microchip to facilitate observation. During the observation, broodstock was fed using commercial feed (pellet, 30% protein). The feed was provided at a ratio of 3% of the total fish biomass. Feeding was done twice daily, at 07.00 am and 5.00 pm. Maturation was conducted through induction using a combination of PMSG and anti-dopamine hormones with doses as follow: (A) 2.0 mL kg^{-1}, (B) 2.25 mL kg^{-1}, (C) 2.5 mL kg^{-1} and (D) controls (without hormone induction). Hormone induction was performed once every 10 days for 30 days. The parameters measured were gonadal maturity stage (GMS), egg diameter (mm), gonadosomatic index (GSI; %), and fecundity (egg per individual). The measurement of GMS and egg diameter was carried out by observing egg size, while GSI was determined based on total gonad weight. The fecundity was calculated based on the gravimetric method (Effendie 2002).

Ovulation of striped snakehead broodstock. This study on the ovulation of striped snakehead was conducted experimentally using a completely randomized design (CRD) with 4 treatments and 3 replications. The ovulation treatment was performed on striped snakehead female broodstock at the gonad maturation stage of GMS IV. Ovulation was induced using hormones, namely (A) LHRHa 0.6 mL kg^{-1}, (B) HCG 500 IU + LHRHa 0.6 mL kg^{-1}, (C) oxytocin 1 IU + LHRHa 0.6 mL kg^{-1}, and control (without hormone treatment). LHRHa induction was conducted twice, the first was 40% of the total dose and the second (after 6 hours) was 60% of the total dose. The induction of the HCG hormone was carried out 24 hours before the first LHRHa hormone induction, whereas oxytocin was carried out at the same time as the first LHRHa induction. Induction was done intramuscularly on broodstock with a slope of 45º. Spawning was carried out based on fullsib at a ratio of 1:1 (1 male to fertilize one female) in a plastic box measuring 100 cm x 80 cm x 60 cm. The parameters measured were the latency of ovulation (hours) and ovulation rate (%). The observation of the latent time and ovulation rate was conducted by observing and calculating based on study Radona et al (2018) in Asian redtail catfish *Hemibagrus nemurus*.

Reproduction performance of striped snakehead. The reproduction performance of striped snakehead broodstock was observed after the ovulation. The parameters measured were fertilization rate (FR; %), hatching rate (HR; %) and survival rate (SR; %). A total of 150 eggs were fertilized and stocked into a basket of 12 cm x 10 cm x 5 cm with a water level of about 3 cm. A total of 9 baskets, representing 3 treatment and 3 replications was used. The value of the fertilization rate was observed 2 hours after fertilization, while the hatching rate was observed after 30 hours of fertilization. The survival rate was observed 2 days after hatching. The reproductive biology parameters observed were measured using Effendie (2002) formula.

Analysis of data. Data were tabulated and analyzed based on the analysis of variance (ANOVA) with a 95% confidence interval. Differences among treatments were analyzed further with Duncan test using SPSS 18.

Results and Discussion

Maturation of striped snakehead broodstock. The GSI and fecundity values of striped snakehead maturation study are presented in Table 1, while the egg diameter and GMS data are presented in Table 2.

Table 1

Characteristics of striped snakehead broodstock induced with PMSG + AD hormone

Hormone treatment	*Repli-cations*	*Length (cm)*	*Weight (g)*	*Gonad weight (g)*	*GSI (%)*	*Average of GSI (%)*	*Fecundity (egg/indv)*	*Average of fecundity (egg/indv)*
PMSG + AD (2.0 mL kg^{-1})	1	23.5	230	5.8	2.52	2.74 ± 0.22^{ab}	3,654	$4,154\pm618^{ab}$
	2	24	246	7.5	3.05		5,025	
	3	23	230	6.1	2.65		3,782	
PMSG + AD (2.25 mL kg^{-1})	1	25.5	250	7.9	3.16	2.98 ± 0.27^{b}	5,056	$4,604\pm541^{ab}$
	2	23.8	245	7.8	3.18		4,914	
	3	23.1	235	6.1	2.60		3,843	
PMSG + AD (2.5 mL kg^{-1})	1	23	238	6.8	2.86	3.21 ± 0.26^{b}	4,692	$5,268\pm410^{b}$
	2	25	247	8.6	3.48		5,504	
	3	24	240	7.9	3.29		5,609	
Controls (without hormone induction)	1	24.1	245	6.2	2.53	2.32 ± 0.15^{a}	4,216	$3,575\pm502^{a}$
	2	23	231	5.1	2.21		3,519	
	3	23	220	4.9	2.23		2,989	

Remarks: numbers followed by different superscript letters in the same line indicate significant differences ($p < 0.05$).

Table 2
Egg diameter and final GMSD of striped snakehead induced with PMSG + AD hormone for 30 days

Hormone treatment	*The initial average of egg diameter (mm)*	*Initial GMS*	*Final average of egg diameter (mm)*	*Final GMS*
PMSG + AD (2.0 mL kg^{-1})	0.4 ± 0.10	GMS I	1.0 ± 0.10	GMS III
PMSG + AD (2.25 mL kg^{-1})	0.4 ± 0.10	GMS I	1.1 ± 0.06	GMS III
PMSG + AD (2.5 mL kg^{-1})	0.4 ± 0.10	GMS I	1.4 ± 0.02	GMS IV
Controls (without hormone induction)	0.4 ± 0.10	GMS I	0.9 ± 0.03	GMS II

Based on the results of the observation, striped snakehead broodstock induced with a combination of PMSG + AD (2.5 mL kg^{-1}) had the highest GSI and fecundity with values of 3.21±0.26% and 5,268±410 egg, respectively. Statistically, the GSI and fecundity were significantly different from the control treatment. Moreover, striped snakehead broodstock induced with PMSG + AD using a dose of 2.5 mL kg^{-1} reached the GMS level IV with bigger egg diameters compared to other treatment (Table 1). Both external and internal factors influence gonad maturity in fish. External factor influencing gonad maturity is environment such as temperature, currents, and the presence of the opposite sex, while the influence of internal factor is in the forms of differences in the species, age, and physiological conditions (Setijaningsih & Asih 2011; Bijaksana 2012).

Induction, using a combination of PMSG + AD (2.5 mL kg^{-1}) hormones, was able to accelerate the gonad maturity stages of striped snakehead compared to other treatments, i.e., PMSG + AD (2.0 mL kg^{-1}), PMSG + AD (2.25 mL kg^{-1}) and controls. This result suggests that the artificial hormonal stimulation with an optimal dose can manipulate the body's hormonal system of fish, as an environmental signal as a natural stimulus is not required (Muslim 2017).

Ovulation of striped snakehead broodstock. The result of striped snakehead ovulation study using a combination of hormones (latent time of ovulation and ovulation rate) are presented in Table 3. Meanwhile, the reproductive performance such as FR, HR and SR are presented in Table 4.

Based on the observation results of 12 striped snakeheads, there were a total of 8 ovulated broodstock out of 12. Broodstock of striped snakehead was induced with the combination of hormone HCG 500 IU + LHRHa 0.6 mL kg^{-1} showed the fastest latency ovulation time with high ovulation rate compared to other treatments, 21 hours 8 minutes and 100% (Table 3). The differences in latent time and ovulation rate were thought to be due to the influence of the content and dose of the hormone induced. The use of gonadotropin hormone in an implant way can increase the maturity of the gonads outside the fish spawning season (Levavi-Sivan et al 2010; Zohar et al 2010). Selvaraj et al (2012) reported that the use of HCG in striped snakehead could improve gonad maturity outside the spawning season. According to Farastuti et al (2014), in the fish induced with HCG + LHRHa hormone, there would be higher secretion of gonadotropin in the blood to stimulate final oocyte maturity and ovulation stages further. Moreover, Rakhmawati (2015) demonstrated that the morphological characteristics of oocytes could be stimulated using a dose of HCG to increase the effectiveness of hormonal stimulation to obtain larger oocyte diameter.

The reproductive performance parameters (FR, HR and SR) revealed the opposite results, as control treatment (without hormone treatment) had the highest value about 95.33%, 85.59%, and 80.88%, respectively. The resulting value statistically showed a significant difference ($p < 0.05$) compared the hormone treatment with LHRHa 0.6 mL kg^{-1}, but without significant differences ($p > 0.05$) compared to the HCG 500 IU + LHRHa 0.6 mL kg^{-1} treatment. This is presumably because the oocytes produced from the striped snakehead control broodstock (without hormone treatment) was more mature and with natural ovulation so that the oocyte quality was maintained without any induction of stimulation. Muslim (2017) in a study also reported that the character of reproduction, especially in the FR values produced in spawning striped snakehead are naturally higher than semi-natural spawning.

Table 3

Latent time of ovulation, ovulation rate and spawning level of striped snakehead using a combination of hormones

Hormone treatment	*Replication*	*Length (cm)*	*Weight (g)*	*Ovulation 100%*	*Latent time of ovulation*	*Average time ovulation*	*Dv*	*Ovulation rate (%)*
LHRHa 0.6 mL kg^{-1}	1	25	280	No	x	29 hours 22 minutes	1 hour 7 minutes	66.6
	2	28.5	340	Yes	28 hours, 15 minutes			
	3	26.3	300	Yes	30 hours, 30 minutes			
HCG 500 IU + LHRHa 0.6 mL kg^{-1}	1	32	400	Yes	20 hours, 20 minutes	21 hours 8 minutes	1 hours 6 minutes	100
	2	27	300	Yes	24 hours			
	3	27.5	340	Yes	21 hours			
Oxytocin 1 IU + LHRHa 0.6 mL kg^{-1}	1	33	420	No	x	28 hours	0 hours	33.3
	2	25.5	340	Yes	28 hours			
	3	29	360	No	x			
Controls (without hormone induction)	1	26.5	320	Yes	96 hours	110 hours	14 hours	66.6
	2	26.3	320	Yes	124 hours			
	3	26	320	No	x			

Table 4

Reproductive performance of striped snakehead broodstock induced with a combination of hormones

Hormone treatment	*Replications*	*FR (%)*	*Average of FR (%)*	*HR (%)*	*Average of HR (%)*	*SR (%)*	*Average of SR (%)*
LHRHa 0.6 mL kg^{-1}	1	88.00	88.93 ± 1.91^{a}	74.55	70.32 ± 3.05^{a}	78.05	73.12 ± 5.16^{a}
	2	91.60		69.00		71.52	
	3	87.20		67.43		67.35	
HCG 500 IU + LHRHa 0.6 mL kg^{-1}	1	92.40	92.80 ± 1.82^{b}	78.35	78.71 ± 1.48^{b}	76.24	75.82 ± 2.50^{ab}
	2	90.80		77.09		72.57	
	3	95.20		80.67		78.65	
Controls (without hormone induction)	1	95.20	95.33 ± 0.50^{b}	81.93	85.59 ± 2.59^{c}	81.03	80.88 ± 0.15^{b}
	2	96.00		87.50		80.95	
	3	94.80		87.34		80.68	

Remarks: numbers followed by different superscript letters in the same line indicate significant differences ($p < 0.05$).

Conclusions. Combination hormone of PMSG + AD (2.5 mL kg^{-1}) was able to accelerate the process of gonad maturity in striped snakehead. Combination hormone of HCG 500 IU + LHRHa 0.6 mL kg^{-1} showed a higher level of artficial spawning in striped snakehead compared to other treatments.

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References

Ath-Thar M. H. F., Gustiano R., Kusmini I. I., Prakoso V. A., Putri P., 2017 [Hormonal induction for gonadal maturation of the striped snakehead *Channa striata*]. Jurnal Riset Akuakultur 12(1):9-20. [in Indonesian]

Bijaksana, 2012 [Snakehead domestication *Channa striata* Blkr, swamp water optimization efforts in South Kalimantan Province]. Jurnal Lahan Suboptimal 1(1):92-101. [in Indonesian]

Center of data and information, Ministry of Marine Affairs and Fisheries Republic of Indonesia (Pusdatin KKP), 2013 [Marine and fisheries statistics 2011]. Jakarta, Ministry of Marine Affairs and Fisheries Republic of Indonesia. [in Indonesian]

Effendie M. I., 2002 Fisheries biology. Pustaka Nusatama Foundation, Yogyakarta, 163 pp. [in Indonesian]

Farastuti E. R., Sudrajat A. O., Gustiano R., 2014 [Induction of ovulation and spawning of soro fish (*Tor soro*) using a combination of hormones]. Limnotek 21(1):87-94. [in Indonesian]

Gustiano R., Kusmini I. I., Ath-thar M. F. H., 2015 [Understanding of genetic resources on Indonesian native freshwater fish species for aquaculture development]. IPB Press, Bogor, 51 pp. [in Indonesian]

Hossain M. K., Latifa G. A., Rahman M. M., 2008 Observations on induced breeding of snakehead murrel, *Channa striatus* (Bloch, 1793). International Journal of Sustainable Crop Production 3(5):65-68.

Kusmini I. I., Gustiano R., Subagja J., Ath-thar M. H. F., Prakoso A., Radona D., 2016 [Naturally and semi-artificial breeding of striped snakehead]. Rekomendasi Teknologi Kelautan dan Perikanan, Balitbang KP, pp. 323-335. [in Indonesian]

Levavi-Sivan B., Bogerd J., Mananos E. L., Gomez A., Lareyre J. J., 2010 Perspectives on fish gonadotropins and their receptors. General and Comparative Endocrinology 165(3):412-437.

Muslim, 2017 [Naturally and semi artificial spawning of striped snakehead (*Channa striata*)]. Jurnal Akuakultur Rawa Indonesia 5(1):25-32. [in Indonesian]

Nasution Z., 2012 [Institutional management of fisheries resources “lelang lebak lebung” and proverty of fishing communities (case study in Ogan Ilir-South Sumatera)]. Thesis, IPB Graduate School, 162 pp. [in Indonesian]

Paray B. A., Haniffa M. A., Manikandaraja D., Milton M. J., 2013 Breeding behavior and parental care of the induced bred striped murrel *Channa striatus* under captive conditions. Turkish Journal of Fisheries and Aquatic Science 13(4):707-711.

Radona D., Subagja J., Prakoso V. A., Kusmini I. I., Kristanto A. H., 2018 [Reproductive biology and level of spawning of Asian redtail catfish *Hemibagrus nemurus* (Valenciennes, 1840) from Cirata population incubated at different temperature settings]. Jurnal Riset Akuakultur 13(2):131-136. [in Indonesian]

Rakhmawati E., 2015 [Induction of female gonadal on striped snakehead (*Channa striata, Bloch*) with HCG hormone injections in culture system]. Undergraduate thesis, Department of Aquaculture, Faculty of Fisheries and Marine Sciences, IPB. [in Indonesian]

Selvaraj S., Francis T., Venkatasamy M., Santhoshkumar S., 2012 Effect of HCG implants on change in testosterone and estradiol level in blood serum of murrel, *Channa striatus*. Tamilnadu Journal Veterinary and Animal Sciences 8(5):290-298.
Setijaningsih L., Asih S., 2011 [Breeding success of kelabu fish (*Osteochilus melanopleura* Blkr) as a local conservation effort through environmental manipulation and hormones]. Prosiding Forum Nasional Pemacu Sumber Daya Ikan III, 18 Oktober 2011, Balai Penelitian Budidaya Air Tawar, Bogor, pp. 1-7. [in Indonesian]
War M., Altaff K., 2011 Preliminary studies on the effect of prey length on growth, survival and cannibalism of larval snakehead *Channa striatus* (Bloch, 1793). Pakistan Journal of Zoology 46(1):9-15.
Yuniarti D., Sulistiyati T., Suprayitno E., 2013 [Vaccum drying temperature influence on quality albumin powder of striped snakehead]. THPi Student Journal 1(1):1-9. [in Indonesian]
Zohar Y., Munoz-Cueto J. A., Elizur A., Kah O., 2010 Neuroendocrinology of reproduction in teleost fish. General and Comparative Endocrinology 165(3):438-455.

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