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# LASERPUNCTURE TECHNOLOGY AS AN EFFORT PREPARATION OF SIAM CATFISH SEED *(Pangasianodon hypophthalmus)* MASS

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***Abstrak***

*Pemijahan ikan patin siam yang sangat bergantung dengan musim dan keadaan lingkungan menyebabkan tidak tersedianya induk matang gonad pada musim kemarau. Alternatif yang sering dilakukan adalah dengan stimulasi biologis menggunakan hormon ovaprim dan teknologi laserpunktur. Tujuan dari penelitian ini adalah untuk mengetahui pengaruh pemberian hormon ovaprim, penembakan laserpunktur dan kombinasi antara keduanya terhadap kecepatan waktu pemijahan dan nilai fekunditas ikan patin siam. Penelitian ini menggunakan metode eksperimental dengan rancangan acak lengkap 3 perlakuan dan 6 ulangan. Hasil dari penelitian ini yaitu diperoleh hasil rata-rata lama waktu pemijahan induk yang diberi perlakuan laserpunktur dan hormon ovaprim dan nilai fekunditas berbeda nyata dengan perlakuan lainnya (p<0,05). Berdasarkan hasil penelitian dapat disimpulkan kombinasi pemberian hormon ovaprim 0,5 ml/kg dan laserpunktur dosis 0,5 J/cm$^2$ selama 32,66 detik dapat mempercepat waktu pemijahan dan fekunditas ikan patin siam.*

***Kata Kunci: Laserpunktur, Hormon Ovaprim, Ikan patin siam***

**Abstract**

The spawning of striped catfish which is very dependent on the season and environmental conditions results in the unavailability of mature gonad broodstock during the dry season. The alternative that is often done is by biological stimulation using the ovaprim hormone and laserpuncture technology. The purpose of this study was to determine the effect of giving the ovaprim hormone, laserpuncture induction and a combination of the two on the speed of spawning time and the fecundity value of Striped catfish. This study used an experimental method with a completely randomized design with 3 treatments and 6 replications. The results of this study were the average spawning time for broodstock treated with laserpuncture and ovaprim hormone and the fecundity value was significantly different from the other treatments (p<0.05). Based on the results of the study, it can be concluded that the combination of 0.5 ml/kg of ovaprim hormone and laser puncture at a dose of 0.5 J/cm$^2$ for 32.66 seconds can speed up the spawning time and fecundity of striped catfish.

**Keywords: Laserpuncture, Ovaprim hormone, Striped catfish**

## 1. INTRODUCTION

The sustainability of striped catfish farming is greatly influenced by the availability of high-quality and plentiful seeds at all times, which is a crucial element in defining the industry's success. As a result, mature


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gonads must always be available on the market in adequate amounts. However, it is impossible to meet the demand for gonad mature striped catfish broodstock at any given moment in adequate numbers. As a result, it's imperative to advance the aquaculture industry by boosting gonad maturation so that reproduction occurs on par with or even ahead of production cycles. (JMittelmark dan Kapuscinski, 2008; Kiran dkk.2013).

Internal enhancements, such as locating high-quality broodstock in large amounts and adjusting gonadotropin hormone levels, as well as exterior improvements, such as enhancing the protein content of broodstock diet, can all improve broodstock handling. These advancements make it possible to raise healthy broodstock in huge quantities that is constantly in great condition. (Izquierdo dkk., 2001; ElSayed dkk., 2003; Cek & Yilmaz, 2007; Sotolu, 2010).

The highest estrogen levels are observed in the late vitelogenic stage and continue to drop when oocytes develop and ovulation, according to numerous studies, and an increase in gonadotropin levels leads estrogen levels to likewise increase in order to raise levels of vitellogenin in the blood (Kagawa et al., 1983; Sinjal, 2007). Striped bass, *Morone sexatilis,*and *Clarias macrocepalus* have both been studied (Sullivan et al., 1991). Reproductive cycle manipulation can be expedited by changing the environment, such as by elevating the pituitary gland or injecting fish with the hormone ovaprim (Yaron, 1995; Azuadi et al., 2011; Gadissa and Devi, 2013; Kiran et al., 2013). In addition to ovaprim, laser puncture induction could be provided at the point of reproduction, specifically on the ventral 2/3 of the broodstock (Kusuma, 2013; Hariani et al., 2014)

A short-wave light called a laser (Light Amplification by Stimulated Emission of Radiation) has the ability to both inhibit and stimulate certain organs and tissues (Karu, 2000). Increased synthesis of enzymes and hormones, as well as activity of membrane receptor reactions, are examples of induction biostimuli in the brain-pituitary-gonad-hepatic axis connected to biological activities; they do not induce inflammatory response. Gonadal maturation and catfish spawning can both be enhanced by laserpuncture He-Ne induction. (Karu, 2000; Koutna dkk.,2003; Kusuma dkk., 2012).

To stimulate the surface active cells in the reproductive site zone of catfish to carry a sequence of energies, the dermis layer is exposed to light at the reproductive point (governoer vessel), which is located 2/3 of the distance from the anus to the pectoral fins. (Kusuma & Hariani, 2017) claim that the act of induction lasers at the site of reproduction can stimulate the creation of energy associated with particular proteins in cells and has an impact on increasing metabolism, with the outcomes mostly affecting gonad growth. According to a different investigation on the impact of biostimulation on tissue, the employment of diode lasers can contribute to promoting cell growth (Dyson, 2002). Male striped catfish can reach optimal gonadal maturity by receiving laser acupuncture with a red light output power of 20 mW and a dose of 0.5 J/cm2 (Mukti, 2020). According to (Yolanda et al., 2016), a red laser with a wavelength of 630–650 nm and a power of 1–5 mW that is exposed for 20 seconds can raise the TKG IV of manvis fish *(Pterophyllum scallare).* The TKG IV of male catfish broodstock can be raised by the 632.8 nm red He-Ne laser, according to Hariani et al. (2020). According to Kusuma and Hariani (2017), male catfish broodstock can be elevated to TKG IV by 632.8 nm red He-Ne laser induction at a dosage of 0.4 J/cm2 and 30% feed protein. Therefore it is necessary to do this research to determine the spawning speed of catfish and how it affects fecundity.

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## 2. RESEARCH METHOD

### 2.1 Time and Place of Research

This research was conducted in March-June 2021. Fish rearing, laserpuncture shooting and calculation of spawning time and fecundity were carried out at the Cultivation Installation.

### 2.2 Equipment and Materials

The equipment used in this study was a 20mW red diode type laser puncture, female Striped catfish broodstock weighing 1000-1500 grams aged 1-1.5 years in TKG I and TKG IV conditions and not yet spawning, 4x2m net size pond, stopwatch, duster, scales and camera.

### 2.3 Research Design

This study used a completely randomized design (CRD) with a factorial pattern consisting of 3 treatments and 6 replications. The first treatment is the administration of the hormone ovaprim 0.6 ml Intramuscular injection technique in fish that have reached TKG IV (Nasrudin, 2015) The second treatment is laserpuncture shooting with a dose of 0.5 J/cm2 for 32.66 seconds once a week for 4 weeks until the fish reach TKG IV (Mukti, 2020), and the third treatment is a combination of laserpuncture until it reaches TKG IV and the hormone ovaprim. Striped catfish are reared in 3 separate concrete ponds and fed factory feed with 30% protein content.

### 2.4 Spawning Time Measurement

Striped catfish that have reached TKG IV and are ready to spawn are weighed and carried out for each treatment at 17.00. Then the spawning time of each group was observed by counting the initial Striped catfish returned to the spawning pond until the fish spawned, after the parents spawned the eggs were taken to count the number.

### 2.5 Fecundity

To measure the fecundity of female fish eggs are weighed to find out the total weight, after which surgery is performed using a sectio set. The taken gonads were weighed to determine the total weight of the gonads. To measure the fecundity of Striped catfish, 0.1 gram of fish eggs were taken and counted. Fecundity can be measured using the gravimetric formula (Effendie, 2002).

$$F = \frac{G}{Q} x N \quad \ldots \quad (1)$$

Description:
F = fecundity (items)
G = gonadal weight (g)
Q = sub-gonad weight (g)
N = number of eggs in the sub-gonads (items)

### 2.4 Data Analysis

Data analysis in this study used IBM SPSS (Statistical Package for the Social Sciences) Statistics Version 24. The statistical test used was Analyze of Variance (ANOVA) completely randomized design (CRD) factorial pattern to determine the effect of the treatment given. If there is a significant difference, it will be continued with Duncan's Multiple Range Test to find out the best treatment.

## 3. RESULTS AND DISCUSSION

### 3.1 Spawning Time Measurement

The following data about spawning time showed at table 1.

**Table 1.** shows the amount of time between ovaprim injection treatment, laserpuncture


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induction, and a combination of both that Striped catfish need to spawn.

| Treatment | N | Spawning time (hours) Mean ± SD |
|---|---|---|
| Ovaprim | 6 | 15.40 ± 2.39b |
| Laserpuncture | 6 | 13.10 ± 3.32b |
| Laserpuncture and Ovaprim | 6 | 10.15 ± 1.8a |

Superscript letters (a, b, c, d) that differ in the same column shows that there is a significant difference ($p<0.05$)

Table 1 show that Ovaprim treatment need 15.40 hours, Laserpuncture need 13.10 and the combination of both need 10.15 hour to perfectly spawned. Striped catfish treated with either ovaprim injection and laserpuncture induction experienced significantly the fastest spawning periods from those treated with the other treatment and the lowest spawing period were in ovaprim injection. At the time of the first check, the catfish had not ovulated yet, so it was necessary to wait until the catfish ovulated perfectly. As stated by Nur et al. (2017), the latent time in fish that has been induced by hormones can last a long time, so routine checks are necessary. This occurs presumably because the eggs have not developed optimally so that the egg follicles have not ruptured, the spawning speed depends on the condition of the mature gonads. Mature gonads are influenced by the amount of gonadotropin hormone (GtH), especially GtH-II released by the pituitary in the bloodstream

The combination between laserpuncture induction and oavprim injection cause to the huge increase in the hormones estrogen and testosterone—which are important for ovulation and spawning process. By creating synaptic connections between GABAergic nerve terminals and GnRH neurons in the hypothalamus, Kusuma et al (2020) claims that the induction effect of laserpuncture can cause GABAergic neurons to generate y-aminobutyric acid (GABA). High quantities of GtH-I and GtH-II hormones are produced in the blood as a result of GnRH regulation of pituitary GtH release. The hormone estrogen synthesis is stimulated by GtH-I. It has been demonstrated that Striped catfish with laserpuncture induction at the reproductive sites can stimulate granulosa cells to make more estrogen than the control group.

The treatment ovaprim, which combines an analog of salmon gonadotropin releasing hormone (sGnRH-a) with antidopamine, can speed up the ovulation and spawning processes in fish. The pituitary is stimulated to release gonadotropins by the GnRH in ovaprim (Lam 1995). The findings demonstrated that the combination of sGnRHa with the dopamine antagonist domperidone proved successful in increasing ovulation, similar to other groups of cyprinids (Szabo, 2003). Injection ovaprim, a synthetic gonadotropin-releasing hormone with a dopamine antagonist, has been shown to be helpful for catfish as well, with a dose of 0.6 mL/kg being the most efficacious for gonadal maturation and spawning stimulation in striped catfish broodstock.

In nature, external environmental factors such as rain, temperature, chemistry, physics, water, time (night), and others have an impact on spawning. Environmental factors will have an impact on the endocrine system's ability to create hormones that aid in gonad development and spawning processes. Fish vitellogenesis, ovulation, and spawning are processes that are influenced by hormones and the environment. The pituitary gland and estrogen regulate the reproductive stages, which can be hastened by the addition of reproductive hormones (Lam 1995, Fujaya 2004). Hormonal engineering typically affects the vitellogenesis process to hasten fish that are challenging to spawn outside of

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their habitat's development and spawning (Yaron 1995).

### 3.2 Fecundity

The following data about spawning time showed at table 1.

**Table 2.** shows the amount of fecundity between ovaprim injection treatment, laserpuncture induction, and a combination of both that Striped catfish need to spawn.

| Treatment | N | Fecundity Mean ± SD |
|---|---|---|
| Ovaprim | 6 | 15.40 ± 2.39b |
| Laserpuncture | 6 | 13.10 ± 3.32b |
| Laserpuncture and Ovaprim | 6 | 19.15 ± 1.8a |

From table 2 it can be seen that the fecundity value in the ovaprim treatment is 55875 and in the laserpuncture treatment is 67132 and in the combination laserpuncture and ovaprim treatment is 79642. The highest fecundity value was obtained in the combination treatment and was significantly different ($p<0.05$) from the fecundity value in the ovaprim treatment.

In sows, a laser puncture at the site of conception has been shown to more swiftly stimulate the hypothalamus-pituitary-gonadal axis, hence promoting the release of gonadotropins. According to (Kert and Rose, 1989), lasers are a short-wave light that can stimulate biological tissues. For example, they can boost the capacity of nerve regeneration, both central and peripheral, as well as cellular activity and the production of hormones and enzymes. Additionally, lasers can alter cell membrane polarization and transmembrane ion transport.

The circulation subsequently transports GtH-I and GtH-II to the gonads, where there is a rise in these hormones compared to before laser puncture induction. Following that, the rise in GtH-I levels will promote the synthesis of steroid hormones like estrogen. elevated estrogen levels as a result of liver laser puncture that induced vitellogenin production. The vitelogenin receptor, acting through vitelogenin endocytosis, transports vitelogenin from the circulation to the oocyte for absorption. The cathepsin enzyme transforms vitelogenin into vitelin, also known as yolk protein, which is then deposited in the growing oocyte (Hariani, 2015). The Ovaprim hormone comprises GnRH, which stimulates the pituitary to create FSH and LH. FSH is responsible for promoting egg maturation, and LH is responsible for promoting ovulation. Together, these two hormones have the ability to hasten egg maturation and release. Compared to ovaprim injection and feeding alone, laserpuncture induction increases metabolic activity to create GtH-II more quickly and abundantly. Because catfish are stimulated not only through the nerves but also through the blood stream, a combination of the two is the best option. Stimulation through the bloodstream will have a slower effect on the hypothalamus-pituitary-gonadal axis, which will affect the speed of spawning catfish followed by ovulation (Zhuo et al., 2011).

## 4. CONCLUSIONS AND SUGGESTIONS

Based on the results of the research that has been done, it can be concluded several things as follows: The combination of laserpuncture induction once a week for four weeks and injection of the hormone ovaprim when fish are about to be spawned has a very significant effect on spawning time and the value of fecundity because it gets stimulation from meridian points and

blood flow so that it can increase the gonadotropin hormone to the maximum and is expected to be applied to cultivators. Based on the research that has been done, the authors suggest to treat it carefully so that fish are not easily stressed.

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