

Case study

Artificial Spawning Techniques for Catfish (*Clarias gariepinus*) at the Cultivated Fisheries Production Business Service Center, Karawang

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ABSTRACT

One of the primary challenges in the catfish cultivation industry is the uncertainty, limited availability, and often poor quality of catfish fries. Fry quality is crucial in aquaculture, yet its reliability is hampered by the natural spawning process, which is influenced by seasonal variations. To address this issue, artificial spawning techniques offer a viable solution. This activity aims to comprehend and effectively execute the precise methods of artificial spawning for catfish, ultimately striving for the production of high-quality fingerlings. Data collection involved the acquisition of both primary and secondary data. The artificial spawning process encompasses container and medium preparation, meticulous broodstock selection and breeding, the artificial spawning procedure, larval rearing, and nursery management. Results from the artificial spawning efforts revealed a fecundity of 75,000 individuals, a remarkable 90% fertilization rate, an 89% hatching success, and an impressive 90% survival rate. Notably, catfish fingerlings were found to harbor parasites such as *Gyrodactylus* sp., *Trichodinasp.*, and *Dactylogyru*ssp. This occurrence can be attributed to the high population density of fish, which contributes to a decline in water quality, emphasizing the importance of cautious management practices in aquaculture.

Keywords: artificial spawning, catfish, freshwater fish, hatchery, spawning

1. INTRODUCTION

Catfish (*Clarias gariepinus*) is a freshwater species that enjoys widespread cultivation across Indonesia's various regions. Catfish farming is gaining momentum as a popular venture. This fish species is gaining popularity due to its rapid growth, straightforward cultivation techniques, accessible marketing channels, modest capital requirements, and adaptability to narrow spaces with high stocking densities. Catfish farming holds significant promise as it features relatively short business cycles, with just one and a half months required for hatching and three months for rearing. Consequently, this accelerated business turnover leads to quicker cash flow [1].

According to [2], Sangkuriang catfish shares striking similarities with African catfish, making them challenging to distinguish. Typically, Sangkuriang catfish are commonly referred to as catfish. These catfish exhibit a sleek, scaleless body with a relatively broad mouth, approximately 25% of their total body length. A defining characteristic of Sangkuriang catfish is the presence of four pairs of antennae strategically situated around their mouth. These four pairs of barbels encompass two pairs of upper jaw barbels and two pairs of lower jaw barbels. The lower barbels serve a dual purpose, functioning both as tactile instruments during swimming and as sensors while searching for food. Sangkuriang catfish boast five distinct fins: the pectoral fin, pelvic fin, anal fin, tail fin, and dorsal fin.[3] notes that catfish predominantly inhabit freshwater environments. Ideally, they thrive in sources like river

water, irrigation canals, spring-fed groundwater, and well water. However, catfish exhibit relative resilience to less favorable water conditions, often deemed suboptimal by standard fish habitat criteria. Notably, Sangkuriang catfish can adapt to high stocking densities and even ponds with diminished oxygen levels, thanks to additional respiratory organs enabling them to extract oxygen directly from the air for respiration.

Hatching is a critical phase in the catfish cultivation process, as it addresses the need for superior fingerlings. Nevertheless, one significant hurdle in the catfish farming sector is the unpredictable, limited, and often subpar supply of fingerlings. [4] reported that the target demand for catfish fingerlings in 2015 reached 6,023,382 fish. This demand continued to surge, reaching 17,428,382 in 2017, while production struggled to keep pace with market needs [5]. To ensure a consistent and sufficient supply of high-quality fingerlings, the industry must overcome the challenge of relying on naturally spawned fingerlings, which are subject to seasonal fluctuations. Artificial spawning emerges as a solution to this dilemma.

Artificial spawning involves the stimulation of female broodstock through hormone injections like *ovaprim*, expediting gonad maturation and enabling controlled artificial spawning. This technique guarantees a timely and reliable fry supply. Additionally, artificial spawning opens the door to genetic advancements in fish breeding, including accelerated growth and disease resistance in tested strains.

At the Karawang Aquaculture Production Business Service Center (BLUPPB), catfish is one of the featured commodities. The center employs artificial spawning techniques, where hormone-stimulated broodstock fish spawn rapidly, yielding high-quality fingerlings that exhibit enhanced disease resistance and reduced fingerlings mortality. Field work activities offer a possibility for acquiring knowledge, understanding, and proficiency in artificial catfish spawning techniques. According to [6], field activities represent a training model aimed at imparting the specific skills demanded by particular occupations. Therefore, the decision to conduct this field activity at BLUPPB Karawang was driven by the desire to broaden scientific insights into artificial catfish spawning techniques. The primary objective of this field activity is to acquire the knowledge and skills necessary to execute effective and precise artificial catfish spawning techniques.

2. MATERIALS AND METHODS

This field activities are conducted at the Aquaculture Production Business Service Center (BLUPPB) in Karawang, North Pusakajaya, West Java. The equipments employed in the process of artificial catfish spawning encompass a syringe with a 0.318 mm needle diameter, having a 3 ml volume, for precisely injecting the *ovaprim* hormone into the fish. Additional equipment includes containers for storing fish sperm and eggs, scales for accurately weighing the fish, cloths for securely handling the fish, surgical scissors for delicately dissecting the fish, chicken feathers for gently stirring the NaCl solution and fish sperm, an aerator to boost oxygen levels in the water, *kakaban* serving as a substrate for egg deposition, and lift nets for capturing fish within the pond. Conversely, the materials utilized in the artificial spawning process for catfish comprise the catfish themselves as the subjects of observation, *ovaprim* hormone as the stimulating hormone for females, a 0.9% NaCl solution to dissolve the sperm, silk worms as larval feed, and PF 300 feed for nourishing the fry.

Data collection for this activity was conducted through two distinct approaches: primary data collection and secondary data acquisition. Primary data was sourced through observations, interviews, active participation, and documentation. Conversely, secondary data represents refined information derived from primary data sources, often retrieved from libraries or reports by previous researchers. Primary data denotes information obtained directly from the source and recorded for the first time, remaining unprocessed. Various methods were employed for primary data collection, including observation, interviews, active participation, documentation, and the use of specific measurement instruments tailored to the research objectives.

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Observation, a deliberate and systematic study, was employed to monitor spontaneous activities related to artificial catfish spawning. Observations involved directly scrutinizing the subject under study and systematically documenting the outcomes. In this endeavor, observations entailed recording activities and documenting aspects linked to artificial catfish spawning. Interviews served as a valuable data collection technique to unearth research-worthy issues and gain deeper insights from respondents. During interviews, questions were posed and answers sought from the responsible personnel involved in the spawning activities at the center. Active participation entailed direct and engaged involvement in field activities related to artificial catfish spawning. This immersive participation facilitated the acquisition of firsthand data and information pertinent to the process. Documentation was executed by actively engaging in fieldwork, capturing essential data and images, and comprehensively recording all field activities.

Secondary data, on the other hand, was garnered from various sources including libraries and previous research reports. As per [7], secondary data constitutes processed forms of primary data, often presented through graphs, tables, diagrams, and images to enhance information richness and usability. In this initiative, secondary data was derived from a thorough analysis of primary data through literature studies, which encompassed journals, books, and previous research findings linked to artificial catfish spawning.

2.1 Preparing Containers and Media

The initial step in artificial spawning involves preparing a brood pond for the breeding process and a spawning pond for egg incubation. The pool must be meticulously cleaned with a brush until it's pristine, then filled with running water and equipped with an aerator to enhance oxygen levels. In the spawning pool, five *kakaban* structures are placed to serve as egg-laying substrates.

2.2 Broodstock Selection

Broodstock selection is a crucial step, as not all catfish in the broodstock pond are prepared for spawning (mature gonads). Selecting suitable broodstocks is pivotal in determining both the quality and quantity of larvae produced during the spawning process. After the selection process, the breeding continues in the prepared pond. *Berok* process is carried out and it is the local term for fasting process which applied to broodstock fish for 1-2 days, aimed at purging waste and reducing the fat content in the gonads.

2.3 Artificial Spawning

Artificial spawning is a method that involves hormonal stimulation using *ovaprim*, which is injected into the female broodstock. Ovulation is then triggered with human assistance, either by stripping or sorting the broodstock's abdomen [8]. The process encompasses injecting the female broodstock with *ovaprim* hormone, dissecting the male broodstock to extract sperm, stripping/sorting the eggs from the female broodstock, mixing the eggs and sperm, and dispersing the eggs into the spawning pool.

2.4 Larval Rearing

Larval rearing occurs after the eggs hatch and continues until the larvae progress to nursery stage 1. This phase involves meticulous monitoring of water quality, efficient food management, ensuring adequate oxygen levels, and maintaining clean, flowing water. Larval care encompasses feeding, periodic water changes, aeration provision, and siphoning to remove leftover food, feces, and deceased catfish.

2.5 Nursery

Nursery activities focus on nurturing the frylings resulting from the initial frying to a size suitable for cultivation in the growth medium [9]. This artificial spawning endeavor is monitored until the catfish reach nursery stage 1. Nursery 1 commences post larval

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harvesting and extends until the fish attain a size of 1-2 cm. Critical considerations include providing ample feed quantities and grouping fish of similar sizes together to ensure uniform growth and minimize predatory behavior among them.

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3. RESULTS AND DISCUSSIONS

3.1 Preparation of Containers and Media

3.1.1 Fish Broodstock Pond

The artificial spawning process for catfish at BLUPPB Karawang involves the use of concrete pools measuring 3 x 5 x 1 meters, totalling three ponds. Concrete ponds are chosen for their durability and ability to prevent water seepage through the bottom and walls. These ponds are equipped with an inlet channel made of *Parallon* pipe that surrounds the pond's sides, and an outlet channel in the form of a PVC pipe at the pond's bottom, leading to a gutter. To prevent larval wastage, a filter is incorporated into the outlet channel.

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Fig. 1. Fish Broodstock Pond

3.1.2 Spawning Pond

In the artificial spawning of catfish, a concrete pond measuring 3 x 5 x 1 meter is employed (Figure 2). The pool is initially cleaned with a brush to remove any adhering dirt. Subsequently, it's filled with water to a 30 cm depth, and an aerator is installed to enhance oxygen levels during spawning. Five *kakabans*, weighted for submersion, are placed in the pond to serve as substrates for catfish egg laying. Before introducing the *kakabans*, they are cleaned to remove any residual eggs and then sun-dried to prevent microorganism growth. These *kakabans*, used at BLUPPB Karawang, are constructed with palm fiber sandwiched between bamboo.

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Fig. 2. Spawning Pool

The preparation of the spawning pond aligns with research by [10], where *kakabans*, serving as egg-laying substrates, are placed at the pond's bottom. These *kakabans*, measuring 140 cm by 40 cm, consist of palm fiber sandwiched between wood or bamboo

and affixed with nails. Before usage, *kakabans* are meticulously cleaned to remove unhatched eggs from previous spawnings and are sun-dried for one day to eliminate microorganisms that could hinder catfish egg hatching. *Kakabans* play a crucial role as substrates for female catfish to deposit their eggs.

3.2 Broodstock Selection

Broodstock selection is a critical step because not all catfish in the broodstock pond are suitable for spawning. As outlined by [11], specific conditions must be met for catfish to be considered eligible for breeding. The selection criteria for good catfish broodstock, as per [12], include the following:

- 1) Non-Sibling: Male and female broodstocks should not share the same genetic lineage, as this can result in poor egg quality.
- 2) Age and Size: Catfish should be at least one year old. Female catfish should weigh between 0.7-1 kg and measure 25-30 cm, while males should weigh between 0.5-0.75 kg and measure 30-35 cm.
- 3) Pond Environment: Broodstock should originate from ponds with stable water conditions, including temperature and pH, to acclimatize them to the pond environment.
- 4) Maturity: Broodstock should be more than one year old for females and over eight months for males.
- 5) Physical Condition: They should have symmetrical bodies, devoid of defects, injuries, and exhibit agility.
- 6) Mature Gonads: The gonads of both male and female catfish should be matured.

Male catfish can be identified by reddish genital coloration, tapered urogenital shape, relatively flat head bone structure, darkening of body color before gonadal maturation, and milky white discharge upon gentle massaging near the urogenital area (Figure 3.). Female catfish can be recognized by their reddish genital coloration, rounded urogenital shape, slightly concave head bone structure, brighter body coloration, enlarged abdomens, and clear greenish egg release upon gentle massaging (Figure 4.)



Fig. 3. Male Catfish Broodstock



Fig. 4. Female Catfish Broodstock

At BLUPPB Karawang's catfish facility, two broodstock meeting these criteria were selected, each weighing 750 grams. Choosing suitable broodstocks is vital for determining both the quality and quantity of larvae produced during spawning. Artificial spawning at BLUPPB Karawang employs a ratio of 1:2, with one male and two females in one spawning pond, in line with [13]. This ratio aims to maximize external fertilization with human assistance, increasing egg fertilization rates and, consequently, the number of larvae produced compared to natural spawning.



Fig. 5. Catfish Broodstock Selection

3.3 Fish Fasting

Fish Fasting Fish fasting is the process of withholding food from the broodstock fish for 1-2 days with the aim of removing waste and reducing the fat content in the gonads. During their stay in the breeding pond, the fish are not fed until they have completed the fasting period. Fertilization is carried out 1 day after the broodstock selection process. Throughout the fish fasting process, water must continuously flow in the pond using a cover to prevent the broodstock catfish from jumping during this period. Fertilization is conducted using a small stream of water while the broodstock fish are not given any food.

The purpose of the fish fasting process is to ensure that the fish adapt well and are not stressed during spawning. This practice aligns with research conducted by [14], where fasting is performed to ensure that the broodstock fish are truly ready to spawn and are not ill, as the readiness of the broodstock fish significantly impacts the quantity and quality of eggs produced. Fertilization is carried out to confirm that the contents of the fish's stomach are eggs rather than food. [15] noted that if the female fish's stomach becomes deflated after fasting, it indicates that the stomach was distended not due to eggs but because of food.

3.4 Artificial Spawning

3.4.1 Preparation of Equipment and Materials

After selecting the broodstock and assessing their readiness, the next step involves weighing the female catfish and proceeding with the injection of the *ovaprim* hormone. Before initiating the hormone injection, it's essential to prepare the necessary tools and materials, which include the following: catfish broodstock, *ovaprim* hormone, physiological solution (0.9% NaCl solution), surgical scissors, syringe, plastic container, towel/rag, tissue, scales, and chicken feathers.

3.4.2 *Ovaprim* Hormone Injection

The purpose of injecting the ovaprim hormone is to stimulate gonad maturation and increase the number of eggs released. Before administering the injection, prepare ovaprim at a dosage of 0.3 ml/kg of the broodstock's weight, then mix it with 0.9% NaCl solution at a 1:1 ratio (*ovaprim*:NaCl). Injections should be performed at night around 22:00 WIB to minimize stress on the catfish. Only one injection is carried out, using a syringe, on each female's back.



Fig. 6. Ovaprim Hormone Injection in Female Catfish

The injection site is on the back, specifically in the muscle near the dorsal fin, at a depth of 2 cm. After the injection, return the female catfish to the pond. The *ovaprim* hormone stimulates gonadal development, resulting in the production of mature egg and sperm cells ready for spawning. *Ovaprim* functions by stimulating gonadotropin hormones in the fish's body, accelerating ovulation, and enhancing egg quality in a relatively short period while reducing mortality rates [16].

Without *ovaprim*, the spawning process in broodstock fish may be delayed due to insufficient gonadotropin content, and there won't be external hormonal stimulation to increase gonadotropin levels in the fish's body [17]. Research by [18] suggests that injecting African catfish with 0.3 ml of *ovaprim* per kilogram of body weight can significantly increase gonadotropin hormone levels in the blood, stimulating egg development and expediting the spawning process with a latency time of 552 minutes.

3.4.3 Sperm Stripping and Surgery

Approximately 8 hours after injecting the female catfish, typically at 07:00 WIB in the morning, the stripping process can commence to release the eggs. This procedure involves wrapping the female fish in a cloth, leaving the abdomen and genital opening exposed. Carefully and firmly, the eggs are extracted from the female broodstock by massaging her abdomen from the pectoral fins toward the anus (Figure 7.). The eggs are then collected in a clean, dry container. Following the stripping, sperm fluid is provided. This is achieved by surgically extracting the sperm sac from the male catfish. The male broodstock is dissected with surgical scissors starting from the genital area toward the head, and then the sperm sac is removed and cleaned using tissue (Figure 8.).



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Fig. 7. Stripping Eggs from Female Catfish



Fig. 8. Dissection of a Male Catfish



Fig. 9. Sperm Extraction

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Male catfish have two sperm sacs, and sperm is extracted by cutting the sperm sacs on the side, squeezing them, and diluting the sperm with 0.9% NaCl solution. The recommended ratio is 250 ml of 0.9% NaCl solution per male broodstock's sperm. It's crucial to perform the fertilization process promptly (mixing sperm and eggs) as sperm fluid remains viable for only about 2 minutes. This aligns with the findings of [19], who suggested that crushed sperm has a viability of approximately 1 minute.

3.4.4 Fertilization

Fertilization is the process by which sperm cells fertilize egg cells, resulting in the formation of a zygote. In the artificial spawning process, fertilization is achieved by gently mixing the available sperm with the eggs, using a chicken feather for careful stirring until an even distribution of sperm is achieved (Figure 10.). After thorough mixing, add sufficient water and gently agitate once more. Initially, both egg and sperm cells are in an inactive state while within the bodies of the female and male broodstocks. When released into water, these cells become active, with the previously inert spermatozoa utilizing their whip-like tails for mobility [20]. Fertilization happens rapidly because sperm remain active for only about one minute when exposed to water. Subsequently, the fertilized eggs are evenly distributed (Figure 11.) in special spawning ponds until the hatching process commences [21]. Fish fertilization is monospermic, meaning only one sperm cell will penetrate the micropyle and fertilize the egg.



Fig. 10. Mixing Eggs and Sperm



Fig. 11. Egg Distribution

[22] noted that when eggs are in water, it leads to an expansion of the perivitelline space and the closing of micropyles within one minute, preventing further sperm entry and reducing fertilization potential. Therefore, when spreading the eggs, it must be done evenly to ensure successful fertilization. Even distribution is essential to counteract the adhesive nature of the eggs, which can lead to eggs sticking together and potentially obstructing oxygen exchange for embryo development, ultimately resulting in the death of the eggs [23].

Fertilized eggs exhibit noticeable development, appearing larger and adopting a dark green coloration, whereas unfertilized eggs remain white like milk. Based on the artificial spawning process conducted, it can be affirmed that the fertilization of catfish eggs was successful, as evident from the clear and greenish appearance of the fertilized catfish eggs compared to unfertilized ones. This success can be attributed to the prior injection of *ovaprim* hormone, which not only expedites the spawning process but also enhances the quality of the eggs. As supported by research from [24], the success of fertilization is significantly influenced by the condition and quality of both the eggs and sperm, which, in turn, rely on the quality of the catfish broodstock.

3.5 Egg Hatching

Fertilized spawning eggs undergo development, eventually hatching into larvae, while unfertilized eggs perish and decompose. Parameters observed during the egg hatching process encompass fecundity, the degree of egg fertilization (fertilization rate), and egg hatchability (hatching rate).

3.5.1 Fecundity

Fecundity, or fertilization success, denotes a broodstock's ability to produce eggs based on the body weight of the fish or the number of eggs contained in the ovaries of the female fish, with mature gonads ready for release during spawning. Fecundity of catfish is determined by weighing the broodstocks before and after spawning, and then calculating the

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difference. In this artificial catfish spawning, an average egg weight per fish was found to be 0.002 grams. Thus, the weight of the gonads in one broodstock is 150 grams. Fish fecundity is determined using the gravimetric method with the formula [6]:

$$F = B_g / B_s$$
$$F = 150 / 0.002$$
$$F = 75,000 \text{ (eggs)}$$

Information:

F = Total number of eggs (eggs)
B_g = Weight of entire gonad (g)
B_s = Weight of a small portion of the gonad (g)

Based on the fecundity calculations, the fecundity of catfish is determined to be 75,000 eggs. This value falls within the range of good fecundity values, as per the [25], which stipulates a good fecundity range for catfish broodstock between 50,000 and 100,000 eggs per kilogram of fish body weight. The fecundity value achieved in this artificial spawning process even exceeds the results of natural catfish spawning research conducted by [26], which yielded a fecundity result of 56,599 eggs. This discrepancy is due to the injection of the *ovaprim* hormone in the artificial spawning process, which stimulates gonad maturation, resulting in more optimal egg production.

[6] asserts that individual female fecundity varies based on factors such as age, size, species, environmental conditions, and food availability. [27] adds that Sangkuriang catfish holds several advantages over other catfish species, including 33.33% higher fecundity than African catfish and a faster age at first gonad maturity. This accelerated gonad maturation is attributed to the stimulation of the *ovaprim* hormone, leading to increased fecundity and higher-quality eggs compared to catfish not exposed to *ovaprim* hormone stimulation.

3.5.2 Fertilization Rate

The Fertilization Rate (FR) represents the degree of egg fertilization, specifically the percentage of eggs fertilized among the total number of eggs released, expressed as a percentage (%). Observations of egg fertilization occur one hour after the egg and sperm mixing process. Following the fecundity calculations, the next step is to calculate FR. From the artificial spawning conducted, a fecundity of 75,000 eggs was determined, with 67,500 eggs successfully fertilized. FR can be calculated using the formula from [20]:

$$FR = P_o / P \times 100 \%$$
$$FR = 67,500 / 75,000 \times 100 \%$$
$$FR = 90 \%$$

Information:

FR = Degree of egg fertilization (%)
P = Total number of eggs
P_o = Number of fertilized eggs

Based on the calculations conducted, the Fertilization Rate (FR) value is determined to be 90%. This value is notably high and aligns with the standards outlined in BSN (2014) for catfish fry production. Specifically, a good fertilization rate for catfish eggs is expected to be at least 60%. The FR value achieved through artificial spawning surpasses that of natural spawning, consistent with research conducted by Iswanto et al. (2016), which recorded a fertilization degree ranging from 76.53% to 99.22% (average 91.48 ± 5.38%) for artificial spawning. Therefore, the FR value attained in this artificial spawning process is impressively high, even exceeding the simultaneous natural spawning conducted at BLUPPB Karawang

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with an FR value of 80%.

The primary factor influencing the degree of egg fertilization in spawning results is closely linked to egg quality. High-quality eggs from mature female broodstocks significantly contribute to successful egg fertilization. In line with [28], fertilized fish eggs above 50% are considered high, those between 30% and 50% are medium, and those below 30% are deemed low. The elevated FR is attributed to the administration of hormones HCG and *ovaprim* to the female broodstock, inducing optimal egg maturation. [29] explain that several factors influence the degree of egg fertilization, including egg quality, fish sperm quality, and water quality, especially temperature and turbidity. During the hatching process, fertilized and unfertilized eggs can be differentiated visually, with fertilized eggs being clear and unfertilized ones appearing milky white.



Fig. 12. Unfertilized Egg

3.5.3 Hatching Rate

Hatching Rate (HR) represents the hatchability of eggs, expressed as the percentage of eggs that hatch. Egg hatching can be triggered by factors such as egg movement, temperature fluctuations, light intensity, and dissolved oxygen levels. Various factors related to water and egg quality play roles in mitigating egg mortality. Calculating the HR involves initially sampling the larvae to determine the total number of successfully hatched larvae. In the spawning process conducted here, it was determined that 60,000 eggs out of 67,500 fertilized eggs hatched. The HR value can be calculated using the formula per [20]:

$$\begin{aligned} \text{HR} &= P_t / P_o \times 100\% \\ \text{HR} &= 60,000 / 67,500 \times 100\% \\ \text{HR} &= 89\% \end{aligned}$$

Information:

HR = Hatchability of eggs (%)
 P_t = Number of hatched eggs
 P_o = Number of fertilized eggs

Based on the conducted calculations, the HR value stands at 89%. This HR value achieved through artificial spawning is high and aligns with the standards set by [30] for catfish fry production, where a good hatchability value for catfish eggs is expected to be at least 60%. In various studies by previous researchers, HR values obtained through artificial spawning consistently exceeded those from natural spawning. For instance, research by [31] recorded hatching rates ranging from 64.93% to 91.96% (average $80.45 \pm 6.28\%$) for artificially spawned catfish eggs. The HR value resulting from artificial spawning also surpasses that of natural spawning carried out at BLUPPB Karawang, which recorded an HR value of 87%.

[32] suggest that the increase in hatchability of catfish eggs when exposed to *ovaprim*

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solution is due to the elevated Follicle Stimulating Hormone (FSH) content, which promotes follicle development and subsequently enhances egg hatchability. HR values are influenced by a combination of internal and external factors. Internal factors include egg quality and sperm quality, while external factors encompass dissolved oxygen (DO) levels, pH, temperature, and ammonia concentration. Successful high egg hatchability can be attributed to factors such as egg quality, water quality, and careful handling during hatching [33].

3.6 Larvae Rearing

Newly hatched catfish larvae are incredibly tiny, making them challenging to spot. During the first three days post-hatching, the larvae are sustained by their own yolk sacs and are not yet ready to feed. Consequently, there's no need to provide them with food in the hatching pond during this period. Typically, on the second day after hatching, they appear white and then gradually develop a mushroom-like appearance. Throughout their care, monitoring water quality parameters, including pH, temperature, and dissolved oxygen (DO), is crucial. This consistent vigilance helps ensure the larvae remain healthy and minimizes mortality. These measurements are performed twice daily. Once the larvae have transformed from *kakaban* to larvae, they are removed from the tank and gently rinsed to release any remaining larvae by agitating the water.

Unhatched eggs should be promptly removed from the tank and discarded. Siphoning can be performed using a small scoop or a small hose to remove both the unopened eggs and any accumulated waste water. Maintaining and servicing the aerator is equally important during the larval care phase to ensure it operates efficiently. The feeding process begins after seven days when the larvae have darkened. By this time, they are usually ready to be transferred to the first nursery pond. During the initial three days in the rearing tank, the catfish larvae don't require external feeding, as they still rely on their yolk sacs for nourishment. On the fourth day, the larvae are introduced to approximately 20 grams of silk worms. Subsequently, after ten days, the larvae are transitioned to a diet consisting of *Fengli-0*.

Feeding is a crucial aspect of ensuring optimal growth and weight gain in the larvae. The choice and form of feed depend on the age and species of the fish. Feeding is carried out by evenly dispersing the feed throughout the pond to promote uniform larval growth. Newly hatched catfish larvae possess food reserves in the form of yolk sacs, which serve as their initial food source, negating the need for external feeding. The hatching of eggs and absorption of yolk sacs occur more rapidly at higher temperatures, as noted by [10]. Typically, feeding can commence when the larvae reach 3-8 days of age or when they can swim and have turned black, as observed by [34]. The feed used in this stage comprises silk worms (*Tubifex* sp.).

3.7 Survival Rate

The Survival Rate (SR) can be determined based on data gathered during the harvest of catfish larvae. SR is calculated as the ratio of the number of larvae at the end of the rearing period to the number of larvae at the beginning, multiplied by 100%. As per [35], the formula for SR calculation is as follows:

$$SR = N_t / N_o \times 100\%$$

$$SR = 54,000 / 52,500 \times 100\%$$

$$SR = 90\%$$

Information:

SR = Survival Rate (%)

N_t = Number of individuals at time t

N_o = Number of individuals at stocking

The SR calculations reveal a remarkable value of 90%. This high SR signifies excellent

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larval survival and aligns well with the standards outlined in [30], where a minimum SR of 60% is considered good for catfish egg fertilization. The high SR indicates that larvae produced through artificial spawning exhibit superior quality compared to natural spawning practices conducted at BLUPPB Karawang, where the SR stands at 75%. This improvement can be attributed to the complete removal of sperm from male fish through surgical processes and the thorough removal of eggs from female fish via the stripping method. This results in larvae with enhanced resilience against pests and diseases.

According to [36], assessing the effectiveness of *ovaprim* involves considering factors such as spawning success rates and the time interval between spawning and the last injection. Based on the remarkable 90% SR achieved, the artificial spawning conducted at BLUPPB Karawang can be deemed highly successful. This success is attributed to the *ovaprim* hormone's stimulation in catfish broodstocks, which promotes the production of high-quality eggs and larvae, as evidenced by the low mortality rate.

3.8 Nursery I

This artificial spawning technique is employed until the catfish reach the age suitable for Nursery 1. Nursery 1 plays a crucial role in the early development of catfish. Nursery refers to the process of nurturing juvenile fish from hatching to a size that can be safely reared in growth media [9]. Nursery 1 begins following the harvesting of larvae and continues until the fish reach a size of 1-2 cm, typically spanning a period from when the fish are 14 days old until they attain this size. Subsequently, the process progresses with two additional nurseries until the fish reach a size of 7-10 cm.



Fig. 13. Catfish nursery phase

During the nursery phase, catfish are provided with suitable food in accordance with their mouth size. With their relatively large mouths, catfish can consume organisms found at the pond bottom as well as artificial feeds. Catfish fry in the nursery phase are fed artificial feed, specifically PF 300 mixed with vitamins, and the feeding regimen consists of two meals daily – once in the morning at 08:00 WIB and again in the afternoon at 16:00 WIB. According to [37], feeding frequency may range from 2 to 3 times daily. When using fry feed, both natural and artificial, it's crucial to adhere to usage guidelines and pay attention to the feed's expiration date as indicated on the packaging label. The nutritional composition of catfish fry feed can be found in Table 1.

Table 1. Nutrient Content of Fry Feed

Nutrient	Content
Protein	42%
Fat	7%
Crude Fiber	2%
Ash Content	13%
Water Content	12%

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The nutritional content within catfish fry feed is suitable for promoting catfish growth. This is attributed to the feed's protein content of 42%, aligning with [9] recommendation that effective catfish feed should contain protein levels exceeding 25%. Additionally, the pellet characteristics adhere to fish feed standards outlined in the [38] encompassing protein (20-35%), fat (2-10%), ash content (<12%), and water content (<12%).

Following this, the grading stage is executed. Grading involves segregating catfish based on their size to achieve uniformity, reduce cannibalism, and enhance feeding efficiency. Grading serves the purpose of categorizing fish fries by size to minimize cannibalism, which frequently occurs when catfish of different sizes coexist and resources become scarce.



Fig. 14. Fry Grading Process

3.9 Pests and Diseases

During the activities, several types of parasites were identified in the catfish fries, including *Gyrodactylus* sp., *Trichodina* sp., and *Dactylogyrus* sp. These occurrences are attributed to the high population density of fish, which can lead to a decline in water quality [39]. Additionally, increased contact among fish due to overcrowding facilitates the rapid spread of parasites. [40] concurs with this, stating that higher fish density elevates the risk of direct transmission of parasites or injuries that can serve as entry points for pathogens. Furthermore, the age and size of the fish also play a role in ectoparasite attacks, with *Monogenea* more frequently targeting fish fry around 1.5-2 months of age due to their increased susceptibility compared to adult fish [41].

The intensity of parasite infestation, such as *Gyrodactylus* sp., *Dactylogyrus* sp., and *Trichodina* sp., was measured at approximately 0.73-2, indicating a relatively mild infestation [42]. The fry appear healthy with no visible signs of parasite attacks. This favorable condition is attributed to effective pond management practices, high-quality fish food, and the enhanced immunity of the fish due to *ovaprim* stimulation in the female fish, resulting in high-quality eggs. Additionally, the use of pellet feed supplemented with vitamins strengthens the fish's immune system. It's noteworthy that BLUPPB Karawang avoids using alternative feeds, ensuring the quality of the distributed fish fries.

3.9.1 Water Quality

Water quality measurements in three catfish cultivation ponds indicate that the conditions are suitable for fish cultivation, despite some fluctuations. These variations remain within the tolerance limits for Sangkuriang catfish frylings. Key water quality measurements for three BLUPPB Karawang catfish fry ponds are presented in Table 2.

Table 2. Water Quality in Three BLUPPB Karawang Catfish Fry Ponds

Parameters	Pond Quality			Standard SNI Value	Unit	Result Description
	K1	K2	K3			
Temperature	28.6	28.4	28.6	22-34	°C	Optimum

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pH	6.56	6.96	6.91	6.5-8.6		Optimum
DO	7.7	7.1	7.2	>1	mg/L	Optimum

Gyrodactylus sp. and *Trichodina* sp. tend to thrive and reproduce more actively in warmer water temperatures. Their ideal temperature range for growth typically falls between 25-30°C, with the highest population density observed at 28°C. Temperature significantly influences various chemical reactions in aquatic environments, including oxygen solubility and fish metabolism. High water temperatures can accelerate the growth of certain parasites.

However, the pH levels in the fish ponds are generally less than optimal. The measured pH tends to be too alkaline for catfish cultivation. Inadequate pH levels can disrupt fish metabolic balance and weaken their immune systems. Fish may become stressed and refuse to eat when exposed to pH levels that are too acidic or alkaline, making them more vulnerable [43].

Despite suboptimal pH levels, the dissolved oxygen content in BLUPPB Karawang's ponds falls within the normal range and complies with SNI standards. Dissolved oxygen content is a critical water quality parameter in fish farming as it directly impacts fish survival. Oxygen is essential for respiration, both for aquatic plants and fish. Maintaining adequate dissolved oxygen can be achieved by using aerators in each cultivation pond.

In summary, the water quality in BLUPPB Karawang remains suitable for fish cultivation, with fluctuations that remain within tolerance limits for Sangkuriang catfish frylings. The observed parasite attacks on catfish fry are likely due to high fish density, which facilitates rapid parasite transmission.

3.9.2 Handling

Fish afflicted by *Dactylogyrus* sp. typically exhibit symptoms such as difficulty in swimming, weakness, and reduced mobility due to impaired respiration. At high infestation levels, fish may even experience gill bleeding [43]. Additionally, the fish's skin takes on a slimy and pallid appearance due to excessive mucus secretion stimulated by the infection. On the other hand, fish affected by *Gyrodactylus* sp., especially at high intensity, will display signs like pale skin, red spots on specific areas of their skin, abnormal mucus production, and skin shedding. *Trichodina* sp. usually infects the external parts of fish, including the skin, fins, and gills, but it can also infiltrate internal organs such as the urinary tract, rectum, and cloaca.

To prevent parasite infestations by *Gyrodactylus* sp., *Dactylogyrus* sp., and *Trichodina* sp., several measures can be taken. One method involves applying lime (CaO) at a rate of 25 kg/ha to ponds that have been dried for 2-3 days beforehand. Alternatively, you can employ a disinfection approach using methylene blue at a dose of 1 gram/m³[43].

If fish become infected with these parasites, treatment can be carried out by immersing the affected fish in a table salt (NaCl) solution at a concentration of 12.5-13 grams/m³ for 24-36 hours. Another effective treatment method is to immerse the fish in a 40 ppm formalin solution for 24 hours.

6. CONCLUSION

Based on this comprehensive activity, it can be deduced that artificial catfish spawning encompasses various crucial steps. These steps include the preparation of containers and media, the meticulous selection of broodstock, the delicate process of stripping, and the core of the operation, the artificial spawning process. The latter involves several stages, including the injection of the *ovaprim* hormone, the stripping of female broodstock, the careful dissection of male fish sperm, and ultimately, the pivotal moments of fertilization and the

dispersal of eggs into designated spawning ponds. In this controlled process, a specific ratio is maintained, typically involving 2 female broodstock for every 1 male broodstock, each weighing approximately 750 grams. Following the artificial spawning process, the outcome is remarkable: a fecundity of 75,000 individuals, an impressive fertilization rate of 90%, and an exceptional hatching rate of 89%. These results unequivocally demonstrate the high quality of the larvae produced through artificial spawning techniques. However, as the fry progress to nursery 1, it's worth noting that parasites, including *Gyrodactylus* sp., *Trichodina* sp., and *Dactylogyrus* sp., have been observed on the fry. This occurrence can be attributed to the elevated fish population density, which can inadvertently lead to a reduction in water quality. Nevertheless, prompt and effective prevention and remedial measures have been systematically applied to tackle these parasites.

Maintaining optimal stocking density and continuously monitoring and managing water quality parameters, such as dissolved oxygen content, water temperature, and pH, are integral aspects of catfish population density control. Additionally, preventing and controlling diseases necessitate meticulous pond maintenance, ensuring the quality of the feed, and guaranteeing the health of fish fry by protecting them from parasites and diseases. Moreover, it is imperative to emphasize the significance of utilizing high-quality, certified feed and providing essential supplements and vitamins to support the overall well-being and robust development of the catfish population. This multifaceted approach underscores the importance of comprehensive care and management throughout the catfish breeding process.

REFERENCES

1. Sunardy. Catfish (*Clariassp*) Hatchery Business Cycle. Self-Help Distribution. Jakarta.2014.
2. Djokosetiyanto D. Eating Habits of Catfish (*Clariassp*). Indonesian Aquaculture Journal, 2012; 11(2):168 - 178.
3. Sudarto. Genetic characteristics of catfish. Bogor: Freshwater Aquaculture Research Center.2004.
4. The Fisheries and Maritime Service of West Java Province. 2014. Analysis of Basic Marine and Fisheries Data 2014. West Java: Ministry of Maritime Affairs and Fisheries.
5. Murhananto. Rearing Dumbo Catfish in the Yard. Jakarta. AgromediaPustaka Publisher. 2002.
6. Omar SBA. Fisheries Biology Practical Module. Makassar: Department of Fisheries. Faculty of Marine and Fisheries Sciences. Hasanuddin University. 2005.
7. Umar, H. 2004, Research Methods for Business Theses and Theses, Jakarta, PT. King Grafindo
8. Meilala, A. Koi Fish (*Cyprinus carpio*) Hatchery Techniques at the Jepun Fish Fry Center, Tulungagung Regency, East Java. Field practice. Surabaya: Faculty of Fisheries and Maritime Affairs. Airlangga University. 2018.
9. Suyanto SR. Catfish cultivation. Jakarta: PenebarSwadaya. 2007.
10. Sunarma A. Increasing Business Productivity of Sangkuriang Catfish (*Clarias gariepinus*). Bandung: Paper presented at the Business Meeting of the Directorate General of Cultivated Fisheries, Marine Fisheries Service, Bandung, 4-7 October 2004.
11. Darseno. 2010. Catfish Cultivation and Business. Agromedia Library. Jakarta. Pages 41-43.
12. Andrianto and Indarto. Catfish Hatchery and Rearing. Jakarta: PenebarSwadaya.2005.
13. Khairuman and Amri K. Intensive Local Catfish Cultivation. Jakarta: Agro Media Library. 2002.
14. Kurnia K. Masamo Catfish (*Clariassp*) Hatchery Technique using the Clear Water System Method at the Aquaculture Production Business Service Center (BLUPPB) Karawang, West Java. Field Work Practices.2015.

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Comment [T39]: The references must be full detailed; should include the journal name, volume and issue number, and the DOI: number for the journal article

15. Mahyuddin K. Complete Guide to Catfish Agribusiness. Jakarta: Penebar Swadaya. 2008.
16. Sukendi. The Effect of a Combination of Ovaprim and Prostaglandin F_{2α} Injection on Ovulation Stimulation and Egg Quality of Dumbo Catfish (*Clarias gariepinus* Burcheel) Bogor: Bogor Agricultural Institute Postgraduate Program. 1995
17. Fujaya Y. Fish Physiology. Jakarta: Publisher PT Rineka Cipta. 2004.
18. Sinjal H. Effectiveness of Ovaprim on Spawning Time, Egg Hatchability and Larval Survival of Dumbo Catfish (*Clarias gariepinus*). Journal of Aquaculture, 2014; 2 (1): 14-21
19. Gusrina. Fish cultivation. Jakarta. Department of National Education. 2008.
20. Effendie Ml. Fisheries Biology. Yogyakarta: Nusatama Library Foundation. 2002.
21. Susanto H. Catfish Cultivation. Jakarta: Penebar Swadaya. 1995.
22. Waynarovich E and Horvath I. Artificial Propagation of Warm Water Finfishes. A Manual for Extensions. FAO. 1980.
23. Baharudin, Ahmad MB, Syakirin and Mardiana TY. The Effect of Soaking in Tea Solution on the Hatchability of Sangkuriang Catfish (*Clarias gariepinus*) Eggs. PENAAquatika 2016; 14(1): 9-17
24. Ardias N. The role of NaCl on the degree of fertilization, egg hatching and survival of koi fish (*Cyprinus carpio*) larvae. Thesis. Bogor: Faculty of Fisheries and Marine Sciences, Bogor Agricultural Institute. 2008.
25. National Standardization Agency. Fry Production of Dumbo Catfish (*Clarias gariepinus* x *C. fuscus*) Spread Fry Class. Indonesian National Standards 01-6484. 4. Jakarta: National Standardization Agency. 2000
26. Engky, Sinjal. 2019. Effectiveness of ovaprim on spawning time, egg hatchability and larval survival of African catfish, *Clarias gariepinus*. Aquaculture. Vol. 2 No. 1: 14 – 21
27. Wulandari, C. Harahap, F and Gultom, B. 2017. The Effect of Giving the Hormone "Ovaprim" with "Wovafh" on the Hatchability of Mother Sangkuriang Catfish (*Clarias Gariepinus*) Eggs at the Samosir Regency Fish Fry Center. Proceedings of the III National Seminar on Biology and its Learning, Medan State University. ISBN: 978-602-5097-61-4
28. Fani F, Inalya I, Rani Y, A'yunin Q, and Evi T. Use of Clay for Successful Spawning of Siamese Patin Fish (*Pangasianodon hypophthalmus*). Scientific Journal of Fisheries and Marine Affairs, 2018; 10 (2): 91-94.
29. Keshavanath P. Nutritional studies on mahseer, Tor khudree (Sykes). In: Coldwater Aquaculture and Fisheries (eds. Singh HR and Lakra WS), pp. 219-228. Delhi: Nar-endra Publishing House. 2000.
30. BSN. 2014. Standards for Dumbo Catfish (*Clarias* sp.). SNI 6484.3:2014.
31. Iswanto B, Suprpto R, Marnis H and Imron. Reproductive Performance of Pearl Catfish (*Clarias gariepinus*). Aquaculture Media, 2016; 11(1): 1-9
32. Manickam PJKP. Induction of Maturation and Ovulation by Pimozide LHRH Analogue Treatment and Resulting High Quality Egg Production in the Asian Catfish, *Clarias batrachus* L. Aquaculture. 1989; 83:193 – 199.
33. Sutarjo, G. A. 2015. The effect of sucrose concentration with the cryoprotectant dimethyl sulfoxide on the quality of carp (*Cyprinus carpio* Lin.) eggs in the cryopreservation process. Gamma Journal, 9(2):20-30
34. Dahlan and Handoyo B. Mass-Scale Catfish Hatchery Using Clear Water System at BLUPPB Karawang. Karawang: Aquaculture For Business and Food Security. 2016.
35. Murtidjo BA. Several Methods for Freshwater Fish Hatchery. Yogyakarta: Kanisius Publishers, 2001.
36. Peter RE, Lin HR and Craak CVD. Induced Ovulation and Spawning of Cultured Freshwater Fish in China: Advent in Application of GnRH Analogue and Dopamine Antagonist. Aquaculture 1988; 74:1 – 10.
37. Rukmana HR. Cultivation and Postharvest of Dumbo Catfish. Semarang: CV. Various Sciences of IKAPI Members. 2003.
38. Indonesian National Standards. 2006, Artificial Feed for Catfish (*Clarias* sp.) (SNI 01-4087-2006), National Standardization Agency, Jakarta.

39. Shafrudin D, Yuniarti and Setiawati M. The effect of African catfish fry density (*Clarias* sp.) on production in a cultivation system with nitrogen control through the addition of wheat flour. *J Indonesian Aquaculture*, 2006; 5 (2): 137-147.
40. Pillay TVR and Kutty MN. *Aquaculture Principles and Practices* (2nd edition). UK: Blackwell Publishing.2005.
41. Ozturk MO. An investigation on metazoan parasites of common carp (*Cyprinus carpio*) in lake Eber. *Turkiye Parazit Derg*, 2005; 29 (3): 204 - 210.
42. Fish Quarantine Center. 2015. *Guidelines for Implementing Quarantine Fish Pest and Disease Monitoring (HPIK)*. Jakarta: Fish Quarantine Center.
43. Kordi MGH. *Management of Fish Pests and Diseases*. Jakarta: Rineka Cipta and Bina Adiaksara.2004.

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