The Effect of Different Stocking Density of Eggs on The Production of Sangkuriang Catfish Seeds (*Clarias gariepinus* Burchell 1822) by Using Filtration System

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**Abstract.** This study aims to determine the production capacity of sangkuriang catfish (*Clarias gariepinus*) with different high egg density in ponds using filtration system. The research is held in Hatchery Fisheries and Marine Science Faculty, Diponegoro University Semarang on December 2017 – January 2018. The test fish used was sangkuriang catfish eggs that hatch and then maintained with maintenance time for 17 days. This research used experimental method with Completely Randomized Design, consisting of 3 treatments with 3 replications i.e. P1 (egg 50 g), P2 (egg 60 g), and P3 (egg 70 g). Variables which was measured include fertilization rate (FR), hatching rate (HR), absolute length growth (L), absolute weight growth (WG), specific growth rate (SGR), production (ind) and water quality. Data were analyzed by multiform analysis, then Duncan double area test to know the difference between treatment to determine the best treatment. The results showed that the different density of the stocked eggs had significant effect (*P*<0.05) on the HR, L and WG, SGR, SR and production, but no significant effect (*P*>0.05) on FR. Treatment P1 showed the best result with value of FR was 92.6 ± 2.10%, HR was 91.33 ± 0.42%, L was 1.50 ± 0.10 cm, WG was 0.13 ± 0.01 gr, SGR was 22.9 ± 0.55%/day, and seed production was 25,323 ± 345.01 ind.

1. Introduction

Sangkuriang catfish is one of the superior varieties of freshwater fish that has grown in the society. This is because it can be cultivated in limited areas with high density and rapid growth and resistance to disease [1]. Catfish produce eggs 50,000-100,000/m² in one spawning [2]. Catfish production in 2012 amounting to 62,686 tons increased in 2013 by 75,236 tons or an increase of 20.02% within a year [3]. The production of catfish *Clarias gariepinus* (Burchell 1822) in Central Java at 2,939,201,940 fish within a year [4]. Increased production in the future requires the support of adequate seed supply. However, until now the demand for catfish seeds has not been fulfilled properly.

The stocking density of catfish larvae in BLUPPB Karawang 10,000 fish tarpaulin ponds with 30 cm water height and obtained seed production of 7,970 fish/m² [5]. However, there are still some obstacles encountered in hatchery activities, one of these constraints is the low value of fertilization and egg hatching because the nature of catfish eggs has adhesiveness (adhesion) so that the eggs clump in one area and low larval growth with high stocking.
density. Therefore, it is necessary to determine the optimal capacity of catfish seed production in aquaculture pond. In addition, the cultivation of sangkuriang catfish seeds produces solid waste and liquid waste from feces and food waste. It also needs to be aware that if the density of the cultivated fish is getting higher, then the ammonia loading into the cultivation system will be also higher [6]. These wastes can potentially become toxic and reduce the production capacity per m$^3$ of water.

One of the efforts that can be done is filtration system technology, which is a water flow system that flows continuously in a maintenance container for sangkuriang catfish seeds. Therefore, preliminary tests were carried out with 50 g egg stocking density for 17 days. There is a filter consisting of dacron and bioball as a filter for dirt in the ponds and using the pump as driving energy. Biofilters, often referred as biological filter or nitrification filter are commonly used in recirculating aquaculture system to remove ammonia and convert it to nitrite, and then to nitrate. Many researches have proved that biofilter system in closed recirculation water system could control water quality within tolerant limit and reduces water requirement [7]. Filters in the filtration system will filter and remove accumulated waste. The filtration systems can be used to improve water quality in aquaculture so that they are suitable for cultivation [8, 9].

2. Research Methods

The test fish used in this study were catfish eggs. The number of eggs used in each treatment is 50 g, 60 g and 70 g. The eggs are hatched and maintained in a maintenance pond for 17 days until the seed size is 1-2 cm. Furthermore, weighing the weight and length of the fish with a sampling of 10 fish every 5 days and a change of water by 50%. The containers used during this maintenance are tarpaulin ponds with a volume of 1,000 l of water.

The test feed used for catfish during the study was natural food, namely silk worms. Silk worms have a high content of protein (57%), fat (13.3%), crude fiber (2.04%), and ash content (3.6%) [10]. Feeding is carried out ad libitum, regarding the feeding of silk worms given to catfish in ad libitum, with the number being increased according to the age of the fish and given at 08.00 WIB and 15.00 WIB [11].

Preliminary tests were carried out first on the stocking density of 50 g eggs with a maintenance period of 17 days and counted for 1 g of eggs amounting to 562 eggs so that it was obtained 50 g of eggs amounting to 28,100 eggs, 60 g of 33,720 eggs and 70 g of 39,340 eggs. Furthermore, higher density research was conducted to determine the optimal density in the 3 m diameter aquaculture pond. The experimental design used in this study was a Completely Randomized Design (CRD) with 3 treatments and 3 repetitions. The treatments of this research were:

- Treatment of P1: egg stocking density 50 g.
- Treatment of P2: egg stocking density 60 g.
- Treatment of P3: egg stocking density 70 g.

2.1. Calculation

Data collection conducted in this study is FR, HR, Pm, H, SGR, SR, production and water quality.

a. Fertilization rate (FR)

The FR calculation formula as follows [12]
FR (%) = \frac{\text{Number of fertilized eggs}}{\text{Total number of eggs counted}} \times 100\% \quad (1)

b. Hatching rate (HR)
The HR calculation formula as follows [12]
\[
\text{HR} \% = \frac{a}{a+b+c} \times 100\% \quad (2)
\]
Description:
\begin{align*}
a &= \text{Normal number of hatched egg} \\
b &= \text{Abnormal number of hatched egg} \\
c &= \text{Non-hatched egg.}
\end{align*}

c. Absolute length growth (L)
Absolute length growth is calculated using the formula as follows [13]
\[
L = L_t - L_0 \quad (3)
\]
Description:
\begin{align*}
L &= \text{Absolute length increase (cm)} \\
L_t &= \text{Fish body length at the end of the study (cm)} \\
L_0 &= \text{Fish body length at the beginning of the study (cm)}
\end{align*}

d. Absolute weight growth (WG)
Absolute weight growth is calculated using the formula used as follows [14]
\[
\text{WG} = W_t - W_0 \quad (4)
\]
Description:
\begin{align*}
\text{WG} &= \text{Absolute weight growth (g)} \\
W_t &= \text{End of maintenance fish weight (g)} \\
W_0 &= \text{Initial fish weight maintenance}
\end{align*}

e. Specific growth rate (SGR)
SGR is calculated using the formula as follows [15]
\[
\text{SGR} = \left( \frac{\ln L_t - \ln L_i}{T} \right) \times 100\% \quad (5)
\]
Description:
\begin{align*}
\text{SGR} &= \text{The daily rate of growth of fish (\%/day)} \\
L_t &= \text{The average length of the test biota at the end of the study (cm/g)} \\
L_i &= \text{The average length of the test biota at the beginning of the study (cm/g)} \\
T &= \text{Maintenance time (day)}
\end{align*}

f. Survival rate (SR)
The SR calculation formula as follows [12]
\[
\text{SR} = \frac{N_t}{N_0} \times 100\% \quad (6)
\]
Description:
SR = survival rate (%)
Nt = Total egg in the final of preservation
No = Total egg in the first spawning.

g. Production
Production of catfish culture obtained from the number of eggs stocked resulted in the number of seeds in maintenance for 17 days in a 3m³ pond with a water height of 20 cm.

h. Water quality
Water quality checks on the research media included several parameters including temperature, dissolved oxygen (DO), acidity (pH), and ammonia. Observation of water quality consisting of ammonia was carried out every 5 days, while pH, DO and temperature measurements were carried out twice a day at 08.00 WIB and 15.00 WIB.

2.2. Statistical Analysis
The data obtained include FR, L, WG, SGR, SR and production. Then done normality testing, testing homogeneity, and additivity test to ensure the data is spread normally, homogeneously, and additively. Furthermore, further testing is carried out namely variance analysis (ANOVA). Data analysis was performed using Ms Excel 2007 and SPSS version 20.0. Data were analyzed for variance (F test) at 95% confidence level. If the analysis of variance was found to be significantly different ($P<0.05$), Duncan’s multiple region test was conducted to determine differences between treatments. Water quality data were analyzed descriptively and compared with the value of water quality feasibility in fish farming to support the survival and growth of fish.

3. Result and Discussion
3.1. Result
The results of the calculation of Fertilization rate (FR) is presented in Figure 1 and Hatching rate (HR) is in Figure 2. The figures showed that treatment P1 had the highest number of FR and HR, while P3 showed the lowest.

![Figure 1. Fertilization rate (FR) of sangkuriang catfish (%)]
Treatment P1 also had the highest value of absolute weight growth of sangkuriang catfish seeds (Figures 3). The weight growth rate of sangkuriang catfish seeds during 17 days for all treatment shown at Figures 4.
Similar with the weight growth, the growth of absolute length of P1 showed the best (Figure 5) and length growth rate (cm) of P1 also showed the best (Figures 6).

![Figure 5](image_url)

**Figure 5.** Absolute length growth of sangkuriang catfish seeds (cm)

![Figure 6](image_url)

**Figure 6.** Length growth rate (cm) of sangkuriang catfish seeds

The SGR of sangkuriang catfish seed of P1 showed the highest among the other treatment (Figure 7), whereas the P3 was the lowest.

![Figure 7](image_url)

**Figure 7.** Specific growth rate (SGR, %/day) of sangkuriang catfish seeds
Figure 7. Specific growth rate of sangkuriang catfish seeds (%/day).

The results of value SR is presented in Table 1, while the production of sangkuriang catfish seeds is in Figure 8. From those data, the P1 still observed the best among others.

Table 1. Survival Rate (SR) of sangkuriang catfish seeds (%)

| Treatment | P1     | P2    | P3    |
|-----------|--------|-------|-------|
| Survival Rate (%) | 98.7 ± 0.005^a | 84.6 ± 0.021^b | 69.06 ± 0.043^c |

Values (mean of duplicates ± SD) in same row with different letters are significant different (P < 0.05)

Figure 8. Production of sangkuriang catfish seeds (ind).

The results of measurements of water quality during the study in all treatment were still in tolerance limits. Completed data are presented in Table 2 and 3.

Table 2. Water quality data of sangkuriang catfish seeds during research

| Treatment | hour | Temperature (°C) | pH     | DO (mg/L) |
|-----------|------|------------------|--------|-----------|
| P1        | 08.00| 25.0-26.4        | 7.81-8.45 | 3.40-4.03 |
|           | 15.00| 25.2-26.7        | 7.40-8.47 | 3.25-3.97 |
| P2        | 08.00| 25.0-26.4        | 7.54-8.40 | 3.15-4.40 |
|           | 15.00| 25.1-26.9        | 7.53-8.16 | 3.13-3.89 |
| P3        | 08.00| 25.1-27.0        | 7.00-8.48 | 3.15-3.94 |
|           | 15.00| 25.1-28.0        | 7.45-8.49 | 3.16-3.89 |
| worthiness|      | *25-30°C         | *6.5-8.5   | *>3mg/L    |

*[2]
Table 3. Ammonia Data on Maintenance of sangkuriang catfish seeds during research

| Treatment | Time (day) | Ammonia (mg/L) |
|-----------|------------|----------------|
| P1        | 0          | 0.097          |
|           | 7          | 0.145          |
|           | 12         | 0.166          |
|           | 17         | 0.233          |
| P2        | 0          | 0.344          |
|           | 7          | 0.194          |
|           | 12         | 0.183          |
|           | 17         | 0.326          |
| P3        | 0          | 0.364          |
|           | 7          | 0.210          |
|           | 12         | 0.197          |
|           | 17         | 0.351          |

Tolerance Limit [16] *<1.57 mg/L

3.2. Discussion
3.2.1. Fertilization rate (FR) and Hatching rate (HR)
The results of variance analysis showed that different egg stocking densities did not have a significant effect \((P>0.05)\) on the fertilization rate in sangkuriang catfish. The average value of fertilization rate from the highest to the lowest was the treatment of egg stocking density of 50 g (P1) of 92.6 ± 2.10%, egg stocking treatment of 60 g (P2) of 88.2 ± 1.72% and egg stocking treatment of 70 g (P3) of 87.1 ± 1.15%. It is suspected that the quality and quantity of sperm from the parent sangkuriang catfish and the maturity of eggs from broodstock female catfish in each treatment are the same. The fertilization rate is largely determined by the quantity and quality of sperm which are influenced by nutrition, temperature, and frequency of broodstock male use. In addition, the level of fertilization rate is also influenced by egg maturity associated with the vitelogenesis process [17].

The results of variance analysis showed that egg stocking density was significantly different \((P<0.05)\) on the degree of hatching of eggs in sangkuriang catfish. The average value of hatching eggs from highest to lowest is P1 treatment of 91.3 ± 0.42%, P2 treatment of 83.7 ± 2.05% and the lowest in P3 treatment of 80.8 ± 0.68%. This is presumably because of the high egg density, the space for movement of the embryo becomes narrow. The egg of density in a hatching medium affects the hatchability of eggs. High egg density has the opportunity to narrow the space of the embryo so that this condition has a bad effect on eggs that have not hatched or can cause death of newly hatched larvae [18].

Based on the research that has been done, the number of egg densities of egg stocking treatment was 50 g (P1), egg stocking density was 60 g (P2), and egg stocking density was 70 g (P3) which amounted to 28,100; 33,720 and 39,340 eggs, respectively. The results fertilization rate of egg stocking treatment were 50 g (P1), egg stocking density was 60 g (P2), and egg stocking density was 70 g (P3), respectively 26,021; 29,741 and 34,265 eggs. The results of the egg hatching rate were 50 g (P1) of egg stocking density, 60 g (P2) egg stocking density and 70 g (P3) egg stocking density respectively, 26,655; 28,224 and 31,787 eggs. Whereas the results of the production of catfish with egg-stocking treatment were 50 g (P1), egg stocking density was 60 g (P2), and egg stocking density was 70 g (P3) in a number of
25,321; 23,874 and 21,952 fingerlings. The treatment of egg stocking density of 70 g (P3) has a number of fertilized eggs, hatching eggs and little seed production. This is thought to be high density so the interaction between fish is very high, due to the narrow space. densities of 30, 40 and 50 fish / l cause the interaction between fish is very high, due to the increasingly narrow living space [19].

3.2.2. Growth rate
The results of variance analysis showed that egg stocking density was significantly different ($P<0.05$) on the growth of absolute weight and length and specific growth rate in sangkuriang catfish. The absolute growth of weights and lengths is associated with different levels of egg stocking density. It is suspected that the high stocking density will cause fish to have high competitiveness in utilizing food and space, so that it will affect growth. The observations of catfish weight growth from the first week to the fourth week using a biofloc system had a higher increase for 1,100/m$^3$ density compared to a density of 1,300/m$^3$ [19].

The results of Duncan's test analysis showed that the highest absolute weight growth value was 50 g (P1) egg stocking density, which was 0.13 ± 0.01 g while the absolute length growth value was 1.50 ± 0.10 cm. This is allegedly due to different levels of stocking density. Low fish density has a wider range of motion so that feed consumption tends to be higher which will affect appetite. The highest growth yield was 1,100/m$^3$ while the lowest was 1,300/m$^3$ with a biofloc system [19].

The lowest growth results in absolute weight and length in egg stocking treatment were 60 g (P2) and egg stocking density 70 g (P3), i.e. absolute weight values of 0.10 ± 0.01 g and 0.07 ± 0.01 g while the absolute length value is 1.20 ± 0.10 cm and 1.0 ± 0.10 cm. It is suspected that high fish density will affect the space for movement. If there is competition for space, then between individuals who are unable to compete will be affected by growth. In addition, it can reduce the appetite of fish which will indirectly slow the growth rate. The highest weight gain value was obtained in 5 stocking / m$^2$ stocking treatment with a value of 12.06 ± 1.56 g [20].

Based on the results of the analysis of variance ($P<0.05$) SGR values showed that different egg densities gave a very real influence on sangkuriang catfish. This is presumably because the increase in high stocking density will interfere with the physiological process and behavior of fish to the space of movement which in turn can reduce the health and physiological conditions of the fish so that the process causes growth to decline. Generally it can be said that the higher the stocking density applied, the lower the growth will be, because there will be good competition for space, DO and feed which affect growth [21].

Based on the Duncan test results, the highest SGR value was 50 g (P1) egg stocking density of 22.9 ± 0.55%/day, the second was 60 g (P2) egg stocking treatment of 21.2 ± 0.67%/day and the lowest value in egg stocking treatment was 70 g (P3) by 19.6 ± 0.56%/day. It is suspected that growth is related to the level of density in fish. Increased density resulted in competition to get food, so that the utilization of food became uneven between fish with one another which resulted in variations in fish size. In addition, increasing density will limit the movement space and movement activities of fish. The high density causes the narrower the space for fish which affects feed competition and physiological conditions of fish, such conditions can ultimately inhibit the growth of catfish seeds [21].
3.2.3. Survival rate (SR) and Production

Based on the results of the analysis of variance analysis showed that egg stocking density was significantly different \((P<0.05)\) on the SR and production of sangkuriang catfish seeds. The treatment with the highest SR and production value was P1 \((98.7 \pm 0.005\% \text{ and } 25,321 \text{ ind, respectively})\), following by P2 \((84.6 \pm 0.021\% \text{ and } 23,874 \pm 870.5 \text{ ind, respectively})\) and the lowest was P3 \((69.06 \pm 0.043\%. \text{ and } 21,952 \pm 241.8 \text{ ind, respectively})\). This is presumably due to different stocking density levels between treatments so that high density will affect the space of movement in the container to be narrow and competition in feed. The high stocking density will affect competition for space and environmental conditions which will then affect growth and production \([21]\).

In addition, it can also be expected that the maintenance of sangkuriang catfish seeds with a filtration system, at 50 g egg stocking density treatment (P1) has reached the carrying capacity limit in the maintenance pond. This is marked by a significant decrease in the real effect on the production of sangkuriang catfish seeds when the density is increased. The decrease in production in cultivation can be caused by a decrease in growth or morbidity and mortality of fish cultured \([22]\).

The lowest yield of seed production in P3 suspected that high fish density will cause many stools to be released, indirectly affecting the quality of water, especially ammonia. High ammonia will lead to lower survival rates in fish due to toxicity. The dirt, urine and excess food can reduce water quality, especially increasing the ammonia content in the water. Organic substances are processed into ammonia \((\text{NH}_3)\) by decomposers. The high amount of \text{NH}_3\ in the maintenance medium can be toxic to fish \([6,23]\).

3.2.4. Water quality

The DO in spawning containers and maintenance of sangkuriang catfish larvae can be said to be quite good, ranging from 3.13 to 4.4 mg/L. This is in accordance with the opinion that DO in catfish farming ranged from 3.26 to 5.66 mg/L \([24]\). The optimum dissolved oxygen concentration for catfish growth is \(>3 \text{ m/L}\). Catfish is still able to live in oxygen concentrations \(<3 \text{ mg/L}\) because it has additional respiratory equipment to take oxygen from the air \([25]\).

Based on the results of the study, the value of the acidity level (pH) in the spawning container and the maintenance of sangkuriang catfish seeds can be said to be quite good, ranging from 7 to 7.86. Good of value pH for catfish growth is between 6.5-9.0 \([26]\). Good of value pH range for spawning, egg hatching and larvae maintenance that is 6.5 to 8.5 \([2]\). Value of pH <5 is very bad for the life of catfish, because it causes clumping of mucus in the gills and can cause mortality. While the pH above 9 can inhibit growth, because it causes less appetite for catfish \([26]\).

Based on the results of the study, the temperature value in spawning containers and maintenance of sangkuriang catfish seeds is 25-26.5\(^\circ\)C. The temperature range can be said to be optimal for the maintenance of sangkuriang catfish seeds. The good of temperature range for spawning, hatching eggs and larvae maintenance is 25-30\(^\circ\)C\([2]\). The temperature values ranging from 26.1 to 27.9\(^\circ\)C are still within the limits of feasibility and support the survival of catfish larvae \([27]\).

Based on the research conducted, the results of ammonia between treatment P1, P2 and P3 experienced differences. The results of treatment P1, ammonia values were 0.097-0.233 mg/L, P2 were 0.326-0.344mg/L, while P3 were 0.351-0.364 mg/L. Ammonia values increase
with increasing density. This is in line with previous study [6]. However, the ammonia value in each treatment was still feasible for the maintenance of sangkuriang catfish seeds. The range of ammonia values for fish maintenance is <1.57 mg/L [11].

4. Conclusions and Suggestion

4.1. Conclusion

The conclusion that can be drawn from this study is that the optimal egg stocking density in this study is at P1 with 50 g egg stocking density, which results in a FR of 92.6 ± 2.10%, the HR of 91.3 ± 0.42%, long growth and absolute weight 1.50 ± 0.10gr and 0.13 ± 0.01cm, SGR of 22.9 ± 0.55%, SR of 98.7 ± 0.005% and seed production amounting to 25,321±348.0 ind.

4.2. Suggestion

Based on study the optimal egg density for a 3 m diameter with 1 m³ water tarpaulin pool is 50 g. Further studies on filter media other than bioball in catfish seedlings to obtain growth rates. and higher life is need.

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