



THE EFFECT OF ADDITIONAL PAPAIN CRUDE ENZYME IN FED ON THE GROWTH RATE OF BARB JUVENILE (*OSTEOCHILUS HASSELTII*) IN FLOATING NET CAGE AQUACULTURE CIRATA RESERVOIR

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KeyWords

Cirata reservoir, Daily Growth Rate, Feed Efficiency, *Osteochilus hasselti*, Papain Crude Enzyme

ABSTRACT

The aim of this research was to determine the effect of addition of papain crude enzyme with different doses in feed and optimal doses that can produce the highest barb juvenile growth rate in floating fish cage aquaculture Cirata Reservoir. The research was conducted from March until May 2016 in laboratory of research and service of chemical at FMIPA UNPAD and floating net cage aquaculture of BPPUIH Cirata Reservoir. The method implemented was experimental method using Group Randomized Design (GRD) with five treatments and three replications. The treatments were five level of the amount of papain crude enzyme on feed (0%, 2.25%, 3.00%, 3.75%, and 4.50%). The results showed that the papain crude enzyme with protease activity 12.63 Unit/mg protein was able to increase the value of daily growth rate and feed efficiency of barb juvenile. The amount of 3.00% papain crude enzyme on feed gave the highest results upon daily growth rate (1.76%/ day) and, feed efficiency (37.70%) with survival rate (98.00%).

Introduction

Silver barb fish (*Osteochillus hasselti*) is a native freshwater fish of Priangan area (West Java) that has the potential for further development in the production. The fish is high in protein, and its eggs are known for the superior taste that may be compared with caviar. The potentials and excellences as protein source as well as commodity must contend with the main issue of the fish having slow growth rate.

In addition, the fish is unable to digest the protein in the feed optimally for the growth as it is not directly absorbed in the digestive system, but has to degraded first by protease to become amino acid (Fujaya, 2008), that most of the protein in the feed is not efficiently used. One of the solutions for the feeding inefficiency is by giving exogenous enzymes in artificial feeding. Exogenous enzymes, one of which, can be found in papayas called papain enzyme as proteolytic enzyme.

Pure papain enzyme may be altered with papain crude enzyme extract to mark down the cost of feeding production. This type of feed would be sufficient for fish farmers in order to enhance the growth of silver barbs.

Method

The main materials for this research include silver barbs as test fish, test feed under the brand ARTHA, and unripe papayas as the main source of papain enzyme. The method implemented in this research is experimental with Randomized Block Design (RBD) that includes five treatments and three repetitions. The five treatments consist of the percentages of papain crude enzyme extract: 0%, 2.25%, 3.00%, 3.75%, and 4.50%. The procedure of this research includes:

Preparation

- Making Papain Crude Enzyme Extract (PCEE). The making of PCEE starts with the process of blending the papayas in a blender, drying the puree, extracting the PCEE into powder using coffee grinder. The final step of this process is testing the activity.
- Identifying Enzyme Activity. The PCEE is dissolved with distilled water (H₂O) of 0.1g/mL. The sample liquid (in 0.1 mL) is poured into a reaction tube with casein (10 mg/mL in buffer fosfat of 0.1 M pH8) for 1 mL. Then, 0.9 mL of buffer fosfat is added into the tube and it is shaken until it becomes homogenous. Next, the sample is incubated for 30 minutes at the temperature of 37°C. After the incubation step, into the tube is poured TCA 8% for 3 mL, and shaken until it is homogenous. The sample is centrifuged at the 10,000 rpm speed for 5-10 minutes. The process would result in supernatant liquid that is measured using spectrophotometer at the wavelength of 280 nm.
- Preparing Farming Media. The farming media used in this experiment is 1m x 1m x 1m net placed in the main net with dimension of 6m x 6m x 3m. The net is set and arranged by tying it to the bamboo frame. Each net is set with ballasts to maintain the square shape of the net.
- Preparing Test Feed. PCEE is applied using spraying method. The process of making test feed starts with scaling the extract in appropriate measure. The enzyme is dissolved with water and sprayed to the feed. The sprayed feed is dried by aerating it, not by the direct sunlight exposure to avoid evaporation or damage of the enzyme. The process of giving the enzyme to the feed is conducted daily during the observation.
- Preparing Test Fish. Silver barb seeds used for the research weigh approximately 9 to 10 grams. The seeds are acclimated for one day before receiving treatment by starving the fish prior to the observation in order to remove feeding residu in the body. Fish are spread into the observation nets for 50 fish per net.

Experiment

The feeding frequency involves three repetitions of spreading with feeding level of 5% of the fish' biomass at 08.00, 12.00, and 16.00 hours (Watanabe, 1998). The research activity ranges for 60 days.

Observation

The observation for fish growth (measurement of the seeds' weight) is conducted for every 10 days, while the water quality (temperature, dissolved oxygen, and pH) is measure at the beginning, middle, and end of the research. After the observation has completed, the data are analysed using several formulations.

Daily Growth Rate (DGR)

Daily Growth Rate is calculated based on of Steffens' formula (1989):

$$DGR = \frac{(\ln W_t - \ln W_0)}{t} \times 100\%$$

Description:

DGR = Daily Growth Rate

lnW₀ = fish biomass day-0 (g)

lnW_t = fish biomass day-t (g)

t = Length of Cultivation (day)

Feed Efficiency Utilization (FEU)

Feed Efficiency Utilization (FEU) is calculated using Tacon's formula (1987):

$$FEU = \frac{(Wt - W0)}{F} \times 100\%$$

Description:

FEU = Feed Efficiency utilization (%)

W0 = fish biomass day-0 (g)

Wt = fish biomass day-t (g)

F = fish feed amount consumed during the study (g)

Survival Rate (SR)

Survival Rate is calculated using the following formula (Effendie, 2002):

$$SR = \frac{Nt}{N0} \times 100\%$$

Description:

SR = Survival Rate (%)

Nt = Fish Number day-t (fish)

N0 = Fish Number day-0 (fish)

Data Analysis

Data from the observation are analyzed with descriptive approach through observation review with secondary data and related literatures on the object. To identify the effects of each treatment, statistical analysis is resorted using analysis of variance (ANOVA) in F-test with significance level of 95%. If the treatment has significant effect, it is proceeded to Duncan's multiple range test and regression (Gasperz, 1994).

Result and Analysis

Daily Growth Rate

Growth means changes in length, weight or volume in a certain period of time. This growth is physically expressed through changes in number or size of the constituent cells of body tissues in a certain period of time. In energetic term, the signs of growth are inevitable in changes in total energy of the body in a certain period of time. Growth occurs when the free energy excesses after the energy available in the feed is used for standard metabolism, digestive process and supporting activities (Gusrina, 2008).

Table 1. Result Analysis of Variance Daily Growth Rate of sample

No.	Treatment	Daily Growth Rate %
1	A (EECP 0 %)	1,42 ± 0,30a
2	B (EECP 2,25 %)	1,73 ± 0,31a
3	C (EECP 3,00 %)	1,76 ± 0,36a
4	D (EECP 3,75 %)	1,60 ± 0,12a
5	E (EECP 4,50 %)	1,61 ± 0,17a

The results from analysis of variance show that the daily growth rate of the silver barb seeds fed with papain crude enzyme extract to 4.50% is insignificantly different (table above). The overall daily growth rates of the fish in this research result in inconsiderable numbers, ranging from 1.42 to 1.73%. This condition, according to Retnosari (2007) is in good category that the acceptable daily growth rate is minimum 1%.

The use of papain crude enzyme extract for 3,00% at treatment C results in the highest number of daily growth rate. This confirms the similar results from research by Pinandoyo, et. al. (2015) that show that the use of papain enzyme for 2.25% in artificial feed for black nile tilapia enhances the highest daily growth rate for 1.83%. Furthermore, it also bears the similar results from research by Amalia, et. al. (2003) that experiments with the use papain enzyme in the artificial feed for dumbo catfish in 2.25% concentrate enhances the highest daily growth rate for 1.97%.

The feed containing papain crude enzyme extract contributes in higher number of daily growth rate compared to that of without the extract. It confirms Sari, et. al. (2013) that fish need enzymes for growth. The enzymes may be endogenous or exogenous, for instance, papain, that help to accelerate the digestive process and protein hydrolysis.

The insignificant differences in each treatment show that feeding with papain crude enzyme extract is sufficient to increase the digestive system of the seeds, but they also need enzymes with higher protease activity in order to achieve maximum daily growth

rate. Reed (1975) states that enzyme concentration is one of the factors that influences the proses of protein catabolism. It indicates that feeding the fish with papain crude enzyme extract enhances the daily growth rate of silver barb seeds.

Feeding Efficiency

Feeding efficiency specifies the effect of feeding to the fish and how the feed is consumed and used that would determine the growth (Gusrina, 2008). In this research, the difference among the values of feeding efficiency in each treatment is statistically tested with analysis of variance. The results from the analysis show that feeding with papain crude enzyme extract until 4.50% to the silver barb seeds does not have significant effect to the feeding efficiency.

This result may challenge that of Amalia, et. al. (2013) that adding papain crude enzyme extract to feed dumbbo catfish with the concentrate of 2.25% bears the highest values of growth rate and efficiency for 1.97% and 62.83% respectively. In addition, Ananda (2015) confirms that giving papain enzyme as supplement in artificial feed in 2.25% engenders the highest percentage of efficiency to 52.39%.

Table 2. The values of feeding efficiency of silver barb seeds

No.	Treatment	Average of Feed Efficiency (%)
1	A (EECP 0 %)	29,07 ± 6,04a
2	B (EECP 2,25 %)	35,89 ± 6,84a
3	C (EECP 3,00 %)	37,70 ± 9,05a
4	D (EECP 3,75 %)	33,33 ± 4,67a
5	E (EECP 4,50 %)	33,74 ± 4,31a

The values of feeding efficiency of silver barb seeds in this research range from 29.07 to 37.70% still fall into low and insufficient category. Craig and Helfrich (2002) find that the value of feeding efficiency fall into sufficient category if the number is higher than 50%. The results may be caused by the slow response of the fish as based on the observation during the research it appears that silver barbs respond to the feed relatively slowly that affects the feeding consumption.

The use of papain crude enzyme extract may result in low feeding efficiency, on the other hand, it enhances the daily growth rate from 1.42% to 1.76%. It can be concluded that adding papain crude enzyme extract does not induce negative impact on the growth of the seeds and it may become one of the solutions to overcome the growth problems.

Survival Rate

The survival rate of silver barb seeds in each treatment does not show significant differences because the fish respond positively to the test feed. It indicates that adding papain crude enzyme extract in artificial feed to 4,50% does not have negative impact on the survival rate of the test fish. The average number of survival rate of silver barb seeds is higher than 97%. This result is stronger than that of research by Pinandoyo, et.al. (2015) on black nile tilapia that the highest number of survival rate of the fish fed with papain enzyme is 96.67% and the lowest is 86.67%. Amalia, et.al. (2013) find even lower numbers in their result of the research on dumbbo catfish, from 83.33% to 91.67%.

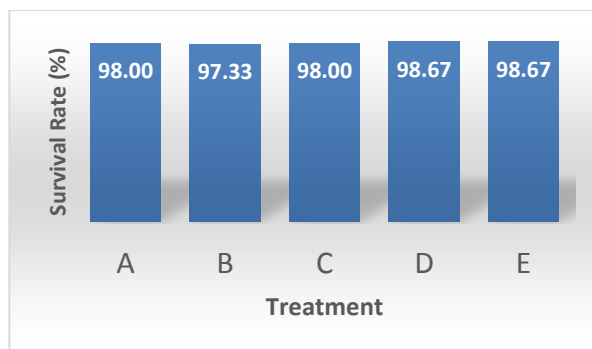


Fig 1. The survival rate of silver barb seeds in each treatment

Water Quality

During the maintenance for this research, the average temperature is between 28.6 to 30.6oC. The range is sufficient for breeding the seeds, as Asmawi (1983) says that the right temperature for silver barb seeds to grow is 18-28 oC. The highest temperature in this research exceeds the standard condition, but it is also still tolerable for the seeds as they are able to survive at the rate of 97%. Khoironi in Nurkarina (2013) confirms that the lethal limit of water temperature for farming is $\leq 11^{\circ}\text{C}$ and $\geq 42^{\circ}\text{C}$.

Table 3. Water Quality along research

Parameters	Sample	Standard	References
Themperature	28,6 - 30,6 oC	18 - 28 oC	Asmawi 1983
pH	6,3 - 7,2	6 - 9	PP RI/82/2001
DO	2,8 - 4,1 ppm	3 - 6 ppm	PP RI/82/2001

The pH level that is too low or too high may cause stress to the fish that would effect their growth. The pH range in this research is between 6.3 and 7.2; the numbers indicate acceptable condition for fish growth. Government Regulation of Republic of Indonesia No. 82/2001 (PP RI/82/2001) on Water Quality Management and Water Pollution Control recommends pH level of 6 – 9 suitable for breeding fish.

The amount of dissolved oxygen during the farming is between 2.8 and 4.1 mg/L. The range number of dissolved oxygen content falls into sufficient and tolerable category for silver barb, referring to PP RI/82/2001 that the range must be between 3-6 mg/L. Dissolved oxygen is an important environmental factor for fish growth because fish require oxygen for respiratory system and metabolism to support movement, growth, and reproduction. An optimum number of dissolved oxygen content influences appetite that promotes absorption of feed and, eventually, high growth rate (Efendi, 2004).

Conclusion

Based on the findings of this research, it may be concluded that:

1. Papain crude enzyme extract as a supplement for artificial feed for silver babr seeds until 4.50 % as the source of exogenous enzyme in the form of protease enzyme plays insignificant role in daily growth rate, feeding efficiency, and survival rate of the fish, referring to the results from analysis of variance with testing frequency of 5%.
2. The percentage of 3.00 % in adding papain crude enzyme extract for artificial feed results in the highest number of daily growth rate (1.76 %), feeding efficient (37.70%), with survival rate of 98% on silver barb seeds.

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