EFFECT OF PROTEIN SOURCE SUBSTITUTION ON FEED FORMULA WITH LAMTORO LEAF FLOUR ON GROWTH AND SUSTAINABILITY LIVE CARP (*Cyprinus carpio L.*)

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ABSTRACT

Lamtoro leaf flour is a valuable nutritional biological resource for raw materials for carp feed. However, in its utilization the lamtoro leaf flour is constrained by the presence of anti-nutrient substances such as tannins and mimosin. Fermented to reduce anti-nutrients and crude fiber content. Fermentation using *Trichoderma* sp. This study aims to examine the effect of lamtoro leaf flour fermented and added to the feed on the growth of carp (*Cyprinus carpio* L.). The research method used was an experimental method with a completely randomized design (CRD) consisting of 3 treatments and 5 replications. Fish were given artificial feed with lamtoro leaf flour content that had been fermented by 0%, 10%, and 20%. The observed variables included Specific Growth Rate (SGR), Absolute Growth, Feed Conversion Ratio (FCR), Survival Rate (SR), Fish response to feed given and Water Quality. Based on the results of the study it can be concluded that the substitution of fish meal protein and corn flour with lamtoro leaf flour which has been fermented with *Trichoderma* sp. into the feed did not have a significantly different effect ($p > 0.05$) on the growth and survival of carp (*Cyprinus carpio* L.).

Keywords: Growth, tilapia, feed, lamtoro leaf, fermentation.

Introduction

Goldfish is one of the freshwater fish commodities that is much favored by the community so that many entrepreneurs cultivate it. Goldfish including freshwater fish species that like a place to live in swift waters. Goldfish can live well in an area of 150-600 meters above sea level (asl) and at a temperature of 25-300C. Although classified as freshwater fish, goldfish are sometimes found in brackish waters or estuaries with salinity (salinity) of 25-30 ‰. Goldfish are classified as an omnivore, which is fish that can prey on various types of food, both those from plants or small animals such as insects. In aquaculture activities have an important role. The feed given must be nutritional value, one of which is its protein content. The protein content in fish feed is the main source of nutrition which still relies on the use of fish meal. To overcome this, it is necessary to substitute alternative sources of animal protein using vegetable protein sources that

are relatively cheaper, available at all times, and of good quality. One of them is by using green plants namely lamtoro leaves which are wild plants. According to Restiningtyas *et al*., (2015) lamtoro leaf is a potential biological resource to be used as feed with sufficient nutritional value forage produced by the chemical composition of lamtoro leaf, 25.9% crude protein, 40% carbohydrate, 11% total ash , N total 4.2%, calcium 2.36%, phosphorus 0.23%, tannins 10.15% and mimosin 7.19%. Lamtoro Leaf Flour is a potential local biological resource for use as a source of fish food. In this research, lamtoro leaves are made as feed for goldfish cultivation because goldfish are omnivore fish, which are fish that can prey on various types of food, are expected to easily adapt to the types of feed mixed with vegetable sources such as lamtoro leaf flour. However, the use of lamtoro leaf flour as feed raw material is limited by its high content of anti-nutrient substances such as tannins and mimosin. Not only anti-nutritive substances, lamtoro leaves contain high crude fiber which fish cannot digest properly.

Therefore, researchers conducted fermentation processes to reduce crude fiber on lamtoro leaves. The working principle in the fermentation process is to break down undigested materials such as cellulose, hemicellulose into simple sugars that are easily digested with the help of microorganisms (Parakkasi, 1995). This is because the digestibility of fish is high because the crude fiber on lamtoro leaves decreases due to the fermentation process using probiotics that contain cellulolytic molds with high cellulase activity such as *Tricoderma* sp. *Tricoderma* sp. is a cellulotic mold that can produce cellulose enzymes that can degrade cellulose.

The purpose of this study was to determine and analyze the effect of protein source substitution on feed formulations with lamtoro leaf flour on the growth and survival of carp (*Cyprinus carpio* L.). This research is expected to provide information on the effect and the right dose for protein substitution in the formulation of feed with lamtoro leaf flour on the growth and survival of carp (*Cyprinus carpio* L.). This research was carried out on 19 April 2019 to 19 May 2019 in the Laboratory of Aquaculture Study Program, University of Mataram and proximate testing was conducted at the Ruminant Laboratory of the Faculty of Animal Husbandry.

Materials and Methods

The fish used in this study were carp (*Cyprinus carpio* L.) seeds measuring ± 10 g/head. Test fish originated from independent aquaculture in the lingsar area. The feed used in this study is artificial pellet feed. The feed was substituted for lamtoro leaf flour which had been fermented with molds of *Tricoderma* sp. in artificial feed with different doses for each treatment. Feeding to carp is done by etatization with the frequency of feeding as much as 3 times, namely in the morning, afternoon and evening. This study used an

experimental method that was carried out with a Completely Randomized Design (CRD) with 3 treatments and 5 replications. The order of treatment in this study are:

P0 treatment : Substitution of fish and corn meal protein with lamtoro fermented leaf meal by 0% of total weight of feed.

P1 treatment : Substitution of fish meal protein and corn flour with fermented lamtoro leaf flour by 10% of the total weight of feed.

P2 treatment : Substitution of fish meal protein and corn with fermented lamtoro leaf flour by 20% of the total weight of the feed.

Before making feeds, a proximate test is first performed on each ingredient and a final test is made on artificial feed.

Calculation of the specific growth rate (SGR) used the formula proposed by Hariati (1989), as follows

$$SGR = \frac{\ln Wt - \ln Wo}{t} \times 100\%$$

Information:

SGR = Specific growth rate (% / day)

Wt = Average weight of the individual at the end of the study (grams)

Wo = individual average weight at the beginning of the study (grams)

t = Duration of Research (days)

The absolute growth of carp measured was weight, using the formula (Afrianto *et al.*, 2005):

W = Wt-Wo

Information :

W = Absolute growth (grams)

Wt = Final weight (grams)

Wo = Initial weight (grams)

According to Tacon (1987), the Feed Conversion Rate is calculated based on the following formula:

$$FCR = \frac{F}{(Wt - D) - Wo}$$

Information :

F = Amount of feed given during maintenance

Wo = Initial average fish weight

Wt = Average fish final weight

D = Number of dead fish weight during maintenance

The survival rate of carp is calculated using the formula Wirabakti (2006):

$$SR = \frac{Nt}{No} \times 100\%$$

Information:

SR = Survival (%)

Nt = Number of fish at the end of the study

No = Number of fish at the beginning of the study

Data Analysis

Data obtained from the results of the study are specific growth rate values (SGR), absolute growth, calculating feed conversion ratio (FCR), survival (SR) and degree of feed preferences. Analysis using variance (ANOVA) to determine the effect of treatment. Before analyzing the variance, the normality and homogeneity tests were first performed. Normality and homogeneity tests are carried out to ensure data is spread normally, homogeneous, and additive in nature. If the analysis of variance obtained significantly different ($P<0.05$), then the Duncan's multiple region test was performed to determine the effect between treatments. Water quality data including dissolved oxygen (DO), acidity (pH), salinity and temperature levels were analyzed descriptively to support growth.

The observations during the study of the specific growth rate, absolute growth, feed conversion ratio, and survival rate were tested for normality, homogeneity, ANOVA presented in Table 1.

Table 1. Specific Growth Rate, Absolute Growth, Feed Conversion Ratio and Survival During the Study.

Observation Data	Treatment		
	P0 (Control)	P1 (10%)	P2 (20%)
SGR (%/day)	-0,00117	-0,00157	-0,00217
Absolute Growth (grams)	-0,274	-0,459	-0,472
FCR (grams)	-3,7766	-7,3022	-6,3668
SR %	92	98	94

Specific Growth Rate

Based on the results of the study the specific growth rate values showed that the treatment of the use of lamtoro leaf flour fermented in artificial feed with respective doses (0%, 10% and 20%) showed a variety of homogeneous and normal spread data and the results of the analysis of variance using a diversity analysis of one direction (one way anova)

found that the value of specific growth rate was not significant ($p \geq 0.05$) so that it could not be further tested by Duncan's double region to determine the effect between treatments. Based on the results of calculations using Excel, the value of the specific growth rate is minus, because the final biomass of fish is lower than the initial biomass, resulting in a minus value. However, if viewed from one way diversity analysis (one way anova) the specific growth rate of the treatments P0, P1 and P2 are not significantly different ($p \geq 0.05$), which means that feeding with fermented lamtoro leaf substitution of 10% and 20% gives the same growth as feeding without the addition of fermented lamtoro leaf flour (control). So that the optimal specific growth rate is P2 treatment with lamtoro leaf flour substitution of 20%, and it is expected that the fermentation of lamtoro leaf flour substitution is 20% as a substitute for protein sources in feed to support the growth of carp. It is suspected that the fermented lamtoro leaf flour in P1 and P2 treatments can increase the protein and carbohydrate content so that it can be utilized optimally as an energy source. Where the carbohydrate content in the feed of each treatment is P0 (60.77%), P1 (47.36%), and P2 (51.47%). According to Rosmawati (2005), that sufficient carbohydrates will prevent the use of protein for energy, so that the available protein can be more utilized for growth.

Absolute Growth

Based on the results of the study the absolute growth rate values indicate that the treatment of the use of lamtoro leaf flour fermented in artificial feed with their respective doses (0%, 10% and 20%) shows a variety of homogeneous and normal spread data and the results of analysis of variance using one diversity analysis direction (one way anova) obtained the value of absolute growth rate is not significant ($p \geq 0.05$) so it can not be further tested Duncan double region to determine the effect between treatments. Based on the results of calculations using Excel the absolute growth rate value is minus, which is because the final biomass of fish is lower than the initial biomass so that it produces a minus value. However, if viewed from a one-way diversity analysis (one way anova) the absolute growth rate of the treatments P0, P1 and P2 are not significantly different ($p \geq 0.05$), which means that feeding with fermented lamtoro leaf substitution of 10% and 20% provide the same growth as feeding without the addition of fermented lamtoro leaf flour (control). So that the absolute optimal growth value is in the treatment of P2 with lamtoro leaf flour substitution of 20%, and it is expected that the fermentation of lamtoro leaf flour substitution is 20% as a substitute for protein sources in feed to support the growth of carp.

Although there was no increase in carp weight during the study, the average absolute growth value in P1 and P2 was

higher than in P0 (control), this is presumably due to different types of feed, where the feed given previously in the goldfish aquaculture ponds derived from the type of floating feed and the feed given in this study is the type of sinking feed.

Feed Conversion Rate

In this study it is necessary to analyze the Feed Conversion Ratio to determine the efficiency of artificial feed substitutes for fish and corn protein with fermented lamtoro leaf flour to the ability of fish to utilize feed optimally for growth. Based on the results of research the absolute growth rate values indicate that the treatment the use of lamtoro fermented leaf flour in artificial feed with the respective doses (0%, 10% and 20%) showed a variety of homogeneous data and normal spread and the results of analysis of variance using one-way diversity analysis (one wayanova) obtained value of feed conversion ratio significant ($p \geq 0.05$) so that it cannot be tested further by Duncan's multiple region to determine the effect between treatments.

Data value of FCR (Feed Conversion Ratio) is obtained by comparing the amount of feed given during maintenance with the weight of fish in each treatment. The data generated by the Feed Conversion Ratio during the study can be said to be not good due to the decrease in fish weight when compared between initial weight and final weight and low feed consumption resulting in a minus value. However, if viewed from a one-way diversity analysis (one way anova) the feed conversion ratio of P0, P1 and P2 is not significantly different ($p \geq 0.05$) which means that feeding with 10% and 20% fermentation of lamtoro leaf substitution gives growth which is the same as feeding without additional lamtoro fermented leaf meal (control), although the average results in Table 1 show the optimal feed conversion ratio at 10% but the ANOVA results show all treatments are not significantly different, so that the feeding of lamtoro leaf flour substitution fermentation of 20% or more can still be given to carp. The decrease in the amount of fish feed consumption is due to the goldfish being in the adaptation phase in artificial feed which is given mainly in the protein-substituted feed of fish meal and corn flour with lamtoro leaf flour (P1 and P2). Decreased amount of feed is thought to be a lack of response of fish to the feed given, where the feed given during the study is different from the feed given previously in the aquaculture pond where the carp was taken. Feed given previously in the aquaculture pond is a type of floating feed while in this study using sinking type of feed. Floating food has the advantage of making the FCR value better because the feed is cooked perfectly so that it optimizes the digestibility of fish while the sinking food is easily destroyed when submerged in water so that the efficiency of feeding is reduced (Harianto *et al.,* 2016).

According to Widyanti (2009), the smaller the value of feed efficiency, the fish are not efficient in utilizing feed or it can be said to be wasteful in utilizing the feed. Fish are unable to utilize feed optimally even though feed digestibility is very high.

Survival Rate

One of the successes of a cultivation activity can be seen from the survival parameters. Based on the results of research the value of survival shows that the treatment of the use of fermented lamtoro leaf flour in artificial feed with their respective doses (0%, 10% and 20%), the test results showed a variety of homogeneous data, spread abnormally. The results of analysis of variance using one-way diversity analysis (one way anova), obtained a value of life insignificance ($p \geq 0.05$) so that Duncan's multiple region test cannot be carried out to determine the effect between treatments. However, if seen from one way diversity analysis, the survival rate of treatment P0, P1 and P2 is not significantly different ($p \geq 0.05$), which means that feeding with 10% and 20% fermentation of lamtoro leaf substitution gives a survival rate of 10%. the same as feeding without additional lamtoro fermented leaf meal (control), although the average yield in Table 1 shows an optimal survival rate of 10%, but the ANOVA results showed all treatments were not significantly different, so the feeding of lamtoro fermented leaf substitution was 20 % or more can still be given to carp.

The occurrence of goldfish deaths in the P0 (Control) treatment was suspected to be stressed due to lack of oxygen in the maintenance media so as to make one by one death in fish while the death of the fish in the P1 and P2 treatments was suspected to be stressed due to lack of oxygen in the maintenance media and in the fermented lamtoro leaf flour still contained substances anti-nurses are like mimosin and tannin. Kurniasih, (2013) states that the decline in growth rate in tilapia is caused by the presence of several antinutrients in the flour of lamtoro leaf namely mimosin and tannins. Mimosin toxicity to fish and shrimp is reported in the form of decreased growth and feed efficiency and increased mortality. Tannin toxicity is reported, among others, binding food nutrients (protein and minerals) so that it cannot be utilized by fish, disrupts the work of digestive enzymes and reduces the absorption of vitamin B12. The presence of anti-nutrient content in lamtoro leaf flour is due to the lack of precise methods to reduce the content of the anti-nutrient substances. Biotic and abiotic factors such as water quality can affect survival, where biotic factors consist of age and fish's ability to meet food needs and abiotic factors including food availability and quality of life media (Handayani *et al*., 2017).

Fish Response To Feed Given

The limited availability of high quality feed raw materials such as fish meal, corn and trash fish stocks in the sea as raw materials for the production of fishmeal is increasingly thinning making the price increase, so that alternative sources of raw materials that have high nutritional value, especially nutrients in the protein, are sought. In this study substitute fish meal protein and corn flour with vegetable raw material sources, namely lamtoro leaves. Lamtoro leaf is a feed for ruminants such as goats and cattle, but lamtoro leaf can also be given to fish because it contains high protein of 29.82% - 32.12% (Hindrawati *et al.*, 2011).

In this research, it is necessary to observe the response of fish to the feed given to determine the response of fish to artificial feed added with green plants such as lamtoro leaves. From the results of a 30-day research experiment, the response of fish to feed containing lamtoro leaf (P1 10% and P2 20%) was given on the first day, some fish tasted first and some fish directly ate the feed and the following days goldfish do not respond too much to the feed. This is presumably because the feed mixed with lamtoro leaf flour which is fermented makes the distinctive odor of fish in the feed decreases and feels a bit bitter so that the goldfish do not respond too much to the feed. P0 (control) treatment on the first day when given fish feed immediately eat it but the following days the response to feed is reduced. This is presumably because the pellets given during the study were different from the feed given previously in the aquaculture pond where the carp were taken. The response of fish is also reduced due to different types of feed given. The feed given earlier in aquaculture is a type of floating feed while the feed in this study is a type of sinking feed. Before the research began carp had been adapted to sinking food for 2 weeks, but during the research carp still needed the process of adaptation with sinking feed.

From the observations of the response of fish to the feed given, both feeds substituted with lamtoro leaf flour or without substitute for lamtoro leaf flour affect the decline in carp growth and feed conversion ratio values, but the Anova test results between treatments were not significantly different ($p \geq 0$, 05).

Water Quality Parameters

Water quality is one of the parameters that must be a concern in fish farming activities. Freshwater quality measurement parameters consist of temperature, DO and pH. Observation of water quality is done 2 at the beginning and end of the study. The results of water quality measurements are presented in Table 2.

Table 2. Water Quality Parameter Data on Carp (*Cyprinus carpio* L.) Seeds During the Study.

Water Quality Parameters	The range	Appropriateness
Temperature ^{0}C	28	28-32^{0}C
DO	3,1-4,1 mg/L	≥ 5 mg/L
pH	7,1-7,2	6,8 – 8,5

Literature Tatangindatu *et al.*, (2013).

Measurement of water quality parameters above shows reasonable results according to the literature to be used as a medium for carp (*Cyprinus carpio* L.) cultivation.

CONCLUSIONS

Conclusion:

1. Based on the results of the study it can be concluded that the protein substitution of fish meal and corn flour with lamtoro leaf flour which has been fermented with *Trichoderma* sp. into the feed gave no significant effect ($p \geq 0.05$) between the treatment of growth and the survival of carp (*Cyprinus caprio* L.).
2. P2 treatment of 20% fermented lamtoro leaf flour substitution gives optimal value to the growth rate of carp (*Cyprinus caprio* L.).
3. During the maintenance of 30 days the goldfish biomass has decreased because it is in the process of adaptation to the fermented lamtoro leaf meal substitution feed.

Suggestion

1. It is recommended to conduct research with a longer period of observation, because the experimental data shows that the indicator is still in the adaptation stage.

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