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Growth Performance of Tilapia Fed with Feed Containing Moringa Leaf Meal and Shrimp Meal

Suardi Laheng[1*], Dwi utami Putri[1], Ika Wahyuni Putri[1], Darmawati[1], Farida Igirisa[1] and Pina[1]

[1]Aquaculture Study Program, Faculty of Fisheries, Universitas Madako Tolitoli, Jl. Madako 1, Kelurahan Tambun, Tolitoli 94515, Indonesia

*Correspondence :
suardiaseq@gmail.com

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Abstract

The research objective of this study was to determine the impact of feed enrichment using Moringa leaf meal and shrimp meal on the growth performance of Tilapia. The initial weight of experimental fish was 6.10 ± 0.55 g/ head. The treatments given were Treatment A (feed without Moringa leaf meal and shrimp meal), Treatment B (commercial feed + 20% shrimp flour), Treatment C (commercial feed + 10% Moringa leaf meal + 20% shrimp meal), and Treatment D (commercial feed + 20% Moringa leaf meal + 20% shrimp meal). In total, there were 4 treatments with 3 replications each. Fish rearing lasted for 40 days. The results showed that the treatments had no effect on amount of feed consumption, protein efficiency ratio, specific growth rate, feed efficiency, survival rate, coefficient of variation and hepatosomatic index ($p>0,05$), but had an effect on the amount of protein consumption and body protein ($p<0, 05$). In conclusion, the enrichment of feed containing Moringa leaf meal and shrimp meal had no effect on the growth performance of Tilapia.

INTRODUCTION

One of the sectors in food security which supports the welfare of the Indonesian people is the Marine and Fisheries sector. The edible fish commonly cultivated by the community is Tilapia. The national production of Tilapia in 2015 was recorded at 567,078 tons, and this figure continued to increase until 2017 reaching 1,265,201 tons (Hadie *et al*., 2018; KKP, 2018).

One of the supporting factors for successful fish farming is feed quality. High feed prices are usually influenced by the good quality of feed protein, which can make successful production activities. The production activities of fish farming contribute around 60% of production costs, and therefore fish feed becomes a factor in fish farming whose quality needs to be given special attention (Gabriel *et al*., 2007). Furthermore, NRC (2011) explained that nutrition from the feed is the raw material for the formation of enzymes and hormones, the repair of damaged body tissues and materials, as well as the formation of meat that supports fish growth.

Techniques in increasing the utilization and digestibility of feed are the provision of natural elements originating from vegetable and animal ingredients. Vegetable ingredients that have the potential to increase the growth and health of fish are Moringa leaf meal. Moringa leaves have some beneficial functions such as increasing the specific

immune system and the growth of *Macrobrachium rosenbergii* (Kaleo *et al*., 2019), serving as an antioxidant in mice (Karthivashan *et al*., 2015a), preventing oxidative stress in Tilapia (Ibrahim *et al*., 2019), and improving the efficiency of feed utilization in Tilapia (*O. niloticus*) (Maslang *et al*., 2018).

Another potential of Moringa leaves is that they contain vitamins and minerals. According to Yaméogo *et al*. (2011), Moringa leaves contain 27.2% protein, 3512 mg/100 g minerals Ca, 717 mg/100 g Mg, 1296 mg/100 g K, 78 mg/100 g Na, 52.1 mg/100 g Fe, and 252 mg/100 g P. However, despite having rich nutrients, Moringa leaves also contain antinutrient compounds such as polyphenols of 43.02 g/kg, saponins of 79.32 g/kg, flavonoids, and alkaloids (Hlophe and Moyo, 2014; Karthivashan *et al*., 2015b).

The information obtained shows that Moringa leaves have the macronutrient and micronutrient potential to increase fish growth, but Moringa leaves also contain antinutrients that can inhibit fish growth. To be used as fish feed, further handling of Moringa leaves is needed such as through a combination of Moringa leaves with nutrient-rich raw materials, namely shrimp meal. Shrimp meal is known to contain high protein of 44.14-64.60% (dry weight) (Zainuddin *et al*., 2019). High protein in shrimp meal is believed to be able to reduce the negative impacts of antinutrients contained in Moringa leaf meal, so it could be effective in increasing the growth of Tilapia. The research by Oku *et al*. (2018) showed that catfish could grow optimally by the provision of feed enriched with 10% Moringa leaf meal and 20% shrimp meal. On this basis, this present study was conducted on the growth of Tilapia (*O. niloticus*), fed with an enriched diet of Moringa leaf meal and shrimp meal.

METHODOLOGY

Place and Time

The research activity was conducted from January to February 2020 for 40 days of cultivation in UPT Local Fish Breeding Center (BBI) Tatanga, Palu. The measurements of feed protein and body protein of fish were carried out in the Laboratory of Fish Feed, Tadulako University.

Research Materials

The tools used were fishnets, scoops, basins, thermometers, digital scales, rulers, pH meters, and DO meters. The ingredients used were Moringa leaf meal, shrimp meal, tapioca starch, vitamin and mineral mixes, Tilapia fish seeds (weight 6.10±0.55 g/head and length 6.61±0.48 cm/head), and commercial feed (30-33%).

Research Design

This research was an experimental research. Artificial feeds were ground into a fine powder and reprinted after given treatments. Next, the feed was added with vitamin and mineral mixes as a substitute for vitamins and minerals of commercial feeds lost during the repelling process, and added with tapioca starch functioning as an adhesive. Furthermore, the feed was dried in the sun. The dried feed later underwent a proximate analysis to determine its protein content (Table 1) (Oku *et al*., 2018).

This study used the following treatments; (A) Feed without the addition of Moringa leaf meal and shrimp meal, (B) The addition of 0% Moringa leaf meal and 20% shrimp meal in feed, (C) The addition of 10% Moringa leaf meal and 20% shrimp meal in feed, and (D) The addition of 20% Moringa leaf meal and 20% shrimp meal in feed.

Table 1. Composition of feed ingredients.

Ingredients in the Feed Manufacturing	Amount of Ingredient			
	A	B	C	D
Commercial Feed (%)	94	74	64	54
Moringa leaf meal (%)	0	0	10	20
Shrimp meal (%)	0	20	20	20
Vitamins and Minerals (%)	3	3	3	3
Tapioca starch (%)	3	3	3	3
Amount (%)	100	100	100	100
Feed Protein (%) *	26.40	30.11	31.87	33.48

Where: * Feed protein analysis was carried out in the Laboratory of Fish Feed, Faculty of Animal Husbandry and Fisheries, Tadulako University, Palu City.

Work Procedure

The Manufacturing of Moringa Leaf Meal and Shrimp Meal

Firstly, the Moringa leaves were separated from the stalks, and then dried in the sun. Then, the dried Moringa leaves were mashed using a blender, and the resulting powder was sieved until Moringa leaf meal was obtained. Furthermore, the manufacturing of shrimp meal was done by steaming the shrimp first and drying them in the sun. Furthermore, the dried shrimp were ground or mashed in a blender, and later sieved until smooth to produce shrimp meal (Oku *et al.*, 2018).

Fish Rearing

Fish were kept in a concrete pond that was insulated using a 50 cm x 50 cm x 100 cm (W x W x H) net. The rearing pond was equipped with an inlet and outlet with a water depth of 73 cm. The net was filled with fish as much as 10 fish / container, and feeding was given as much as 5%. The experimental fish were reared for 40 days and were fed 3 times a day at 08.00, 12.00, and 16.00 WITA.

Acclimatization of the fish was done for 1 week by feeding fish with commercial feed (without Moringa leaf meal and shrimp meal) with a feeding rate of 5% and the frequency of feeding 2 times a day. Furthermore, the fish were starved for 1 day to reduce the impact of the previously given feed. Following that, 120 fish were each weighed for all treatments. Weighing was done in a dry weight system. The weighing result was the initial weight of the fish which was used as a reference for feeding.

The measured water quality included temperature, pH and oxygen. The weight and survival rate of the fish were measured every 10 days, specifically on the 10^{th}, 20^{th}, 30^{th} and 40^{th} days. On the 40^{th} day, the fish were starved for 1 day, and then the fish were weighed and their length was measured. Three fish from each treatment were chopped finely at the proximate to determine the protein content in their body (Oku *et al.*, 2018).

Parameters

Parameters observed in this research were the amount of feed consumption, amount of protein consumption, specific growth rate, feed efficiency, coefficient of variation of fish weight, protein efficiency ratio, survival rate, hepatosomatic index, and water quality.

The feed consumed by fish during the study became a reference in measuring the amount of feed consumption. A 0.01-gram scale was used to measure the feed given every day.

The formula for the amount of protein consumption is:

$$APC = AFC \times FP$$

Where:
APC = amount of protein consumption (g)
AFC = amount of protein consumption (g)
FP = feed protein (%)

The formula for specific growth rate based on Ozovehe (2013) is:

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

Where:
SGR = specific growth rate (%)

Wt = final weight (g)
Wo = initial weight (g)
t = rearing period (days)

The formula for feed efficiency based on Watanabe (1988) is:

$$FE = \frac{(Wt + Wd) - Wo}{F} \times 100\%$$

Where:
FE = feed efficiency (%)
Wt = final weight (g)
Wd = dead fish weight (g)
Wo = initial weight (g)
F = feed consumed (g)

The variation of fish weight was expressed in the coefficient of variation by Baras *et al*. (2011):

$$CV = \frac{SD}{\bar{x}} \times 100$$

Where:
CV = coefficient of variation (%)
SD = standard deviation
$\bar{x}$ = mean of fish weight (g)

Fish at the beginning and the end of the rearing period were measured for their body protein using the Kjeldahl method (Manuel *et al*., 2019).

The formula for protein efficiency ratio based on Manuel *et al*. (2019) is:

$$PER = \frac{Wt - Wo}{Pi} \times 100$$

Where:
PER = protein efficiency ratio (%)
Wt = final weight (g)
Wo = initial weight (g)
Pi = protein intake (g)

The formula for survival rate based on Madinawati *et al*. (2011) is:

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

Where:
SR = survival rate (%)
Nt = final number of fish
No = initial number of fish

A total of three fish in each treatment were used to measure the hepatosomatic index. Measurements were made at the beginning and end of the study. The fish were weighed to determine their body weight and then dissected. Afterward, the liver was weighed to determine the liver weight of the fish. The formula for hepatosomatic index based on Effendie (2002) is:

$$HSI\ (\%) = \frac{\text{liver weight (g)}}{\text{body weight (g)}} \times 100$$

Every ten days water quality was measured in situ including dissolved oxygen, water pH and temperature. Measurements were conducted in the morning, afternoon and evening.

Data Analysis

Parameters that were tested statistically were the performance of feed consumption and protein consumption of Tilapia. Data tabulation used MS. Office 2013, while the ANOVA test used the SPSS program version 16 with a 95% confidence level. Significantly different treatments were further tested using the Honestly Significant Difference (HSD) test.

RESULTS AND DISCUSSION

Based on the observations, it was shown that the treatments did not affect growth performance (initial weight, final weight, total feed consumption, specific growth rate, feed efficiency, survival rate), protein efficiency ratio, and hepatosomatic index ($p>0.05$). The p* results of this study are in line with the results of research by Manuel *et al*. (2019) showing that Tilapia feed added with 10-30% Moringa leaf meal did not affect final weight, total feed consumption, specific growth rate, and feed conversion ratio. Another study conducted by Dogmeza *et al*. (2006) showed that Moringa in the form of extracts did not affect the growth of Tilapia. Research by Richter *et al*. (2003), however, showed different results in which the addition of 12% of Moringa leaf meal was able to improve fish growth performance.

Table 2. Growth performance of tilapia given feed enriched with Moringa leaf meal and shrimp meal.

Test Parameter	Treatment of Experimental Feed			
	Control	MLM 0% + SM 20%	MLM 10% + SM 20%	MLM20% + SM 20%
B0 (g)	6.15±0.22 [a]	6.15±0.34 [a]	6.05±0.18 [a]	6.08±0.45 [a]
Bt (g)	32.63±4.11 [a]	31.68±1.96 [a]	31.15±0.97 [a]	31.67±2.01 [a]
AFC (g)	34.40±1.91 [a]	33.41±2.15 [a]	32.36±0.32 [a]	33.38±1.16 [a]
SGR (%)	4.16±0.26 [a]	4.10±0.21 [a]	4.10±0.01 [a]	4.13±0.34 [a]
FE (%)	76.80±8.55 [a]	76.51±5.46 [a]	77.54±2.27 [a]	76.64±6.05 [a]
SR (%)	100±0.00 [a]	100±0.00 [a]	100±0.00 [a]	100±0.00 [a]
CV (%)	21.75±1.64 [a]	22.19±1.64 [a]	24.12±1.64 [a]	21.25±1.64 [a]

Where: Different letters in the same line show significantly different treatment effects ($p<0.05$). MLM= Moringa Leaf Meal, SM= Shrimp Meal, B0= Initial weight, Bt= Final weight AFC= Amount of feed consumption, SGR= specific growth rate, FE= Feed Efficiency, SR= *survival rate*, CV = Coefficient of Variation.

The results showed that the treatments had no effect on the coefficient of variation of Tilapia ($p>0.05$). The coefficient of variation for all treatments ranged from 21.25-24.12%. These results indicate that the variation in the diversity of the experimental fish was classified in the moderate category in the narrow and homogeneous diversity. This is in accordance with the statement of Tampake *et al*. (1992) stating that the coefficient of variation of 0-20% was included in the low category, 20-50% was in the moderate category, and >50% was in the high category. Fish feed is one of the causes of differences in the value of the coefficient of variation (Robisalmi and Dewi, 2014). According to Harianto *et al.* (2014), one of the factors in the occurrence of fish weight diversity is that there is competition among fish in obtaining feed. In general, small fish are less competitive in getting food than large fish.

Many studies on Moringa leaf meal have been carried out on various types of fish. For example, research of Hlophe and Moyo (2014) on Mujair fish given feed substitution of 100% Moringa leaf meal with fish meal showed a decrease in fish body weight and feed conversion ratio. The research of Ayotunde *et al*. (2016) on catfish given feed substitution of 10-30% Moringa leaf meal with fish meal showed body weight loss. On the other hand, research by Puycha *et al*. (2017) in catfish showed no effect on fish growth and feed conversion ratio with the treatment of Moringa leaf meal of 10-20%.

The decrease in body weight of Tilapia fed with Moringa leaf meal was thought to be a negative effect on the antinutrients of Moringa leaves. Hlophe and Moyo (2014) stated that Moringa leaf is one of the vegetable ingredients containing saponins of 79.32 g/kg, polyphenols of 43.02 g/kg, phytic acid of 25.41 g/kg, and tannin of 12.10 g/kg. Antinutrients contained in vegetable ingredients have a negative effect on growth, causing fish to experience a decrease in growth speed (Francis *et al*., 2001) and interference on nutrient absorption due to digestive enzymes not working normally (Muhammad *et al*., 2011; Francis *et al*., 2001). According to NRC (2011), if the absorption of nutrients is inhibited, the formation of enzymes and hormones will also be inhibited so the distribution of nutrients in the body is not optimal, thereby causing the growth speed of fish to slow down. The protease, amylase, and lipase enzymes are digestive enzymes that have an important role in the digestion of food (NRC, 2011) while growth hormone is a hormone which plays an important role in the growth of fish.

Table 3. Body protein (BP), amount of protein consumption (APC), protein efficiency ratio (PER), hepatosomatic index (HSI) on Tilapia.

Test Parameter	Treatment of Experimental Feed			
	Control	MLM 0% + SM 20%	MLM 10% + SM 20%	MLM 20% + SM 20%
APC (kg)	908.25±50.54 [a]	1005.92±64.60 [ab]	1031.31±10.19 [b]	1117.40±38.80 [b]
BP (%)	13.67±0.42 [c]	12.46±0.06 [ab]	13.26±0.05 [bc]	11.95±0.17 [a]
PER (%)	0.77±0.09 [a]	0.77±0.05 [a]	0.78±0.02 [a]	0.77±0.06 [a]
HSI (%)	1.04±0.28 [a]	0.80±0.05 [a]	0.94±0.17 [a]	0.89±0.18 [a]

Where: Different letters in the same line show significantly different treatment effects ($p<0.05$). MLM= Moringa Leaf Meal, SM= Shrimp Meal.

Based on the measurement results, there was no effect of feed treatments on growth performance, protein efficiency ratio, and hepatosomatic index ($p>0.05$). Meanwhile, the results of the measurement of body protein and the amount of protein consumption showed an effect between treatments ($p<0.05$) (Table 2). Treatments C and D had the highest protein consumption while the lowest amount of protein consumption was in treatment A (control). Feed protein in treatment C of 31.87%, in treatment D of 33.48%, and in treatment A of 26.40% caused differences in protein consumption. The high amount of protein consumption in treatments C and D was not followed by an increase in protein efficiency, which showed no effect between treatments. On the other hand, the measurement of body protein showed that Treatment A (control) obtained the best results compared to all feed treatments containing Moringa leaf meal and shrimp meal. The presence of antinutrients contained in Moringa leaves is one of the causes of the decrease in the body protein of Tilapia. The anti-nutrient content of vegetable ingredients in feed affects palatability, which is assumed to inhibit the utilization of feed protein. According to Hlope and Moyo (2014), the antinutrients of Moringa leaves can bind digestive enzymes and decrease the bioavailability of nutrients for fish.

This is in line with the research of Ayotunde *et al*. (2016) stating that catfish experienced a decrease in body protein when given feed enriched with 10-30% Moringa leaf meal. The results of protein efficiency and protein content in the body of Tilapia (*O. niloticus*) which are not in line with the results of protein consumption are thought to be due to the presence of antinutrients in the form of saponins of 79.32 g/kg and polyphenols of 43.02 g/kg of Moringa (Hlophe and Moyo, 2014). Saponins are reported to have negative effects, such as inhibiting protein digestibility, causing abnormal intestinal morphology, reducing mucosal enzymes in the intestine, and inhibiting the work of lipase enzymes in the pancreas (Francis *et al*., 2002; NRC, 2011; Tiamiyu *et al*., 2016). In addition, saponins also decrease the absorption of vitamins Fe, Mg, Ca, and Zn in the body and cause hypoglycemia through the inhibition of glucose in the intestine (Francis *et al*., 2002).

Polyphenols are classified into two main groups, which are non-flavonoids and flavonoids. Non-flavonoids are phenolic acid, stilbenes, lignans, and tannins. Meanwhile, flavonoids are flavonols, flavones, flavanols, flavanones, anthocyanidins, and isoflavones (Panche *et al*., 2016; Singh *et al*., 2017). Excessive consumption of polyphenols can inhibit the work of digestive enzymes and reduce growth (Santos-Buelga and Scalbert, 2000). The negative effects of polyphenols are that they can inhibit the catalytic process and they can inhibit the binding of food substrates in the intestine, thereby inhibiting the proteolytic processing (Velickovic and Stanic-Vucinic, 2018). As a result of these negative effects, energy from fat and protein cannot be utilized optimally (Yuangsoi and Masumoto, 2012). Efforts to reduce the negative

effects of the antinutrients of Moringa leaves can be conducted through solvent extraction and enzyme treatment on plant-based feeds, soaking, heat treatment, and drying (Afuang *et al*., 2003). In addition, fermentation techniques are also effective in reducing the negative impacts of Moringa leaves (Putra *et al*., 2018).

Table 4. The results of water quality measurements during the study.

Parameter	Treatment				Standard (KKP, 2009)
	A	B	C	D	
Temperature (°C)	27	29	27	28	23-30
pH	7.5	7.7	7.7	7.9	6.5-8.5
DO (ppm)	6.2	6.3	6.2	6.3	>5

The rearing of Tilapia for 40 days showed the optimal range of water quality for the survival rate of Tilapia (Table 4). Fish will grow optimally, and the survival rate will be high if the water quality is in accordance with the needs of the fish (Begum *et al*., 2014). Gunadi *et al.* (2015) further stated that the living environment of fish is one of the determining factors for fish growth. However, in this study, the optimal water quality could not be utilized properly by Tilapia. This was observed in the body protein of Tilapia treated with Moringa leaves which tended to decrease. This might occur as the content of Moringa leaves caused the gills to experience hyperplasia. According to Rapatsa and Moyo (2014), symptoms of hyperplasia were found in Mujair fish after being fed with Moringa leaves. Hyperplasia inhibits respiration and expiration of gasses in the fish body (Jamin and Erlangga, 2016).

CONCLUSION

Feed enrichment using Moringa leaf meal and shrimp meal had no effect on the growth performance of Tilapia. It is highly suggested that further research use Moringa leaves that have been processed by solvent extraction and enzyme treatment, soaking, heat treatment, drying, and fermentation, aiming to reduce the content of antinutrients.

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