

Replacement of fish meal with fermented Moringa leaves meal and its effect on the growth performance of red tilapia (*Oreochromis* sp.)

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Abstract. The purpose of the present research was to evaluate the nutritional content of fermented *Moringa oleifera* leaves meal and to find out the effect of fish meal replacement with fermented Moringa leaves meal (FMLM) in diets on the growth performance of red tilapia (*Oreochromis* sp.). The treatments include replacement of the fish meal with FMLM at 0, 10, 20, and 30%. Red tilapia with total length of 9-10 cm was fed twice daily for two months at a feeding rate of 3% of total biomass. The results revealed that fermentation increased the moisture, protein, ash, total lipid, calcium, and phosphorous content in Moringa leaves meal (MLM). The fermentation process actually reduced the anti-nutrient substances (phenol, tannin, phytic acid, and hydrogen cyanide (HCN)) in MLM. Fish meal replacement with FMLM in diets significantly affected feed digestibility. FMLM can be used as a replacement for fish meal in diet at 30% with only limited effect on the growth performance of red tilapia.

Key Words: mixture of bacteria, Moringa leaves meal, growth, feed digestibility, red tilapia.

Introduction. In 2014, the production of global fish cultivation contributed to 44.14% of the total production of caught and aquaculture fisheries activities (FAO 2016). The aquaculture production is greatly dependent on the quantity and quality of feeds. Feed comprises the largest cost portion at 50 to 70% of the total aquaculture production costs (Tacon & Metian 2008). Fish meal is the most crucial animal protein source for fish feed (Miles & Chapman 2006). Lunger et al (2007) stated that the low level of supply and the high level of demand of fish meal cause an increase in feed prices. The production of fish meal has decreased an average of 1.7% per year from 2008 to 2010 (Tacon et al 2011). This condition challenged the researchers to find sustainable feed material sources to replace fish meal. Kokou & Fountoulaki (2018) revealed that using an alternative replacement for fish meal in fish feed is increasingly needed because of the reduction of fisheries stock. Therefore, the researchers have focused on using the plant-based protein sources which were cheap and locally available (Hlophe & Moyo 2014). Omnes et al (2017) stated that plant-based protein can be used as a replacement for fish meal. Kokou & Fountoulaki (2018) proved that plant-based materials have already been successfully used as sustainable alternative feed materials to replace fish meal for several aquaculture species.

Moringa (*Moringa oleifera*) has frequently been studied as an alternative and promising protein source in fish feed. Moringa is a source of plant-based protein which can be used for fish feed formula, because it contains protein up to 30.3% and has 19 amino acids like methionine, cysteine, and tryptophan (Moyo et al 2011), as well as vitamins B, C, K, and beta carotene (Ganzon-Naret 2014). Moringa leaves contain high energy, minerals and phenolics, particularly flavonoid and phenolic acids as natural antioxidant source (Valdez-Solana et al 2015). In general, feed is formulated from various kinds of feed materials to fulfill fish nutrient needs, so that their physiological functions can work normally, including protecting their natural defense systems, growth,

and reproduction (Alemayehu et al 2018). Richter et al (2003) reported that Moringa leaves meal (MLM) is able to replace fish meal and wheat flour in tilapia feed by 10% and produce good growth performance. Ayotunde et al (2016) indicated that MLM is potentially to replace fish meal in *Clarias gariepinus* feed until 10% without reducing the growth. MLM can be used in *Pangasius bocourti* feed up to 100 g kg⁻¹ without negative effects on the growth, feed utilisation, digestibility, and serum biochemistry (Puycha et al 2017). MLM up to 10% in feed produces the highest growth performance in *Sparus aurata* (Mansour et al 2018). Karina et al (2015) stated that as much as 16 to 32% of MLM can be formulated into tilapia (*Oreochromis niloticus*) feed components.

Plant-based protein source feed materials often have lower dried material digestibility than fish meal. As a consequence, it will cause an increase in waste production, because there are nutrients that are not digested (Glencross et al 2012). The low level of digestibility is due to anti-nutrient substances (anti-nutritional factors), including crude fiber and polysaccharides which cannot be digested (Krogdahl et al 2010). Moringa leaves contain anti-nutrient, such as saponin, tannin, phytic acid and hydrogen cyanide (HCN) (Francis et al 2001). Fermentation is a technique which is used to improve the nutrient quality of an agricultural by-product, so that it can be used in a diet formulation as a source of protein (Bertsch & Coello 2005). Fermentation reduces the anti-nutrient content, improves the crude protein content, and reduces the crude fiber content from the plant material (El-Batal & Abdel Kareem 2001). Ijarotimi et al (2013) reported that fermentation improves the essential amino acid content, fatty acid content, and phytochemical content in Moringa seed meal. *Bacillus pumilus* CICC 10440 and solid-state fermentation are successfully used to increase the release of nutrients in MLM (Zhang et al 2017). This research strives to evaluate the nutrient content of fermented Moringa leaves meal (FMLM) and discover the effect of fish meal replacement with FMLM in feed towards the growth performance of red tilapia (*Oreochromis* sp.).

Material and Method. The research was conducted at laboratory of Aquaculture, Department of Fisheries, Faculty of Agriculture, Universitas Gadjah Mada, Indonesia from February to May 2019.

Moringa leaves meal preparation. Moringa leaves were obtained from Bantul Regency, Yogyakarta Administrative Region Province, Indonesia. The leaves were dried in an oven at a temperature of 40 to 50°C for 6 to 8 hours. The dried leaves were grinded to become flour with measurements of 1 mm, then the leaves were kept in an airtight plastic bag.

Fermentation of Moringa leaves meal. The bacteria used in this research were T2A (*Bacillus* sp.), T3P1 (*Bacillus* sp.) and JAL11 (*Lactococcus raffinolactis*), which previously were grown in a sterile Tryptic Soy Broth medium for 24 hours and added with 450 mL of sterile Phosphate Buffer Saline, homogenized, mixed evenly with 500 g of MLM that was airtight, and fermented at a temperature of 34 to 37°C for 168 hours. The sample was dried at a temperature of 50 to 60°C for 7 hours, refined, and kept in an airtight plastic bag before being used for a chemical analysis.

Analysis of the Moringa leaves meal nutrient, anti-nutrient and fish feed contents. The analyses of the MLM, FMLM, and fish feeds nutrient content including the moisture, protein, ash, total lipid, crude fiber, calcium, and phosphorous contents, were done based on AOAC (2005). An anti-nutrient substance analysis in the form of phenol, tannin, and HCN were done based on Harborne (1987), and phytic acid was done on Wheeler & Ferrel (1971). The nutrient and anti-nutrient content were shown in Table 1.

Table 1
Nutrient and anti-nutrient contents of Moringa leaves meal (MLM) and fermented Moringa leaves meal (FMLM)

	MLM	FMLM
<i>Nutrient content (%)</i>		
Moisture	9.04±0.00 ^a	13.33±1.32 ^b
Ash	9.70±0.21 ^a	14.05±0.25 ^b
Protein	25.77±0.08 ^a	34.77±0.09 ^b
Total lipid	4.80±0.52 ^a	6.25±0.13 ^b
Crude fiber	11.60±0.13 ^b	8.24±0.56 ^a
NFE	39.09±0.18 ^b	23.36±0.47 ^a
Energy (kcal DE 100g ⁻¹)	296.46±0.26 ^b	295.10±0.23 ^a
Calcium	1.65±0.00 ^a	2.52±0.03 ^b
Phosphorous	0.30±0.00 ^a	0.52±0.00 ^b
<i>Antinutrient content (%)</i>		
Phenol	1.41±0.00 ^b	1.32±0.00 ^a
Tannin	1.49±0.00 ^b	1.39±0.00 ^a
Phytic acid	0.28±0.00 ^b	0.12±0.02 ^a
HCN	0.06±0.00 ^b	0.04±0.00 ^a

The different superscripts on the same row show a significant difference ($p < 0.05$); ¹ MLM = Moringa leaves meal; ² FMLM = fermented Moringa leaves meal; ³ NFE = nitrogen-free extract; calculated = 100-(%CP+%CL+%moisture+%ash+%CF); ⁴ Energy = {(protein x 4.5 kcal g⁻¹)+(lipid x 9.1 kcal g⁻¹)+(NFE x 3.5 kcal g⁻¹)/100 (NRC 1993); ⁵ DE = digestible energy.

Fish feeds formulation. The feeds formulation was made at a protein content of 32% using the Pearson's Square Method. The treatments involved replacing fish meal with FMLM at 0, 10, 20, and 30% in red tilapia. The result of proximate analyses of fish diets were shown in Table 2.

Table 2
Ingredient and proximate composition (% on dry weight basis) of diets containing different level of fermented Moringa leaves meal (FMLM)

	FMLM level			
	0%	10%	20%	30%
<i>Ingredient (% dry matter)</i>				
Fish meal	42.00	32.00	22.00	12.00
Soybean meal	35.00	35.00	35.00	35.00
FMLM	0.00	10.00	20.00	30.00
Rice bran	10.00	10.00	10.00	10.00
Starch	8.00	8.00	8.00	8.00
Mineral mixture	2.00	2.00	2.00	2.00
Vitamin C	2.00	2.00	2.00	2.00
Fish oil	1.00	1.00	1.00	1.00
Cr ₂ O ₃	0.6	0.6	0.6	0.6
<i>Proximate analysis (%)</i>				
Moisture	15.41±3.08 ^d	14.79±3.90 ^c	14.16±0.02 ^b	13.25±0.12 ^a
Protein	31.32±1.43 ^d	30.41±0.00 ^c	27.24±1.39 ^a	29.23±0.30 ^b
Total lipid	13.74±0.07 ^a	14.55±0.42 ^c	14.01±0.13 ^b	15.43±0.12 ^d
Ash	13.16±0.03 ^d	12.42±0.39 ^c	10.65±0.25 ^a	11.19±0.14 ^b
Crude fiber	3.09±0.31 ^a	4.14±0.32 ^b	5.44±0.41 ^d	4.65±0.24 ^c
NFE	23.28±0.98 ^a	23.69±1.00 ^b	28.50±0.44 ^d	26.25±0.18 ^c
Energy (kcal DE 100g ⁻¹)	347.45±0.82 ^a	352.16±0.47 ^c	349.82±0.65 ^b	363.82±0.20 ^d

¹ FMLM = fermented Moringa leaves meal; ² NFE = nitrogen-free extract; calculated = 100-(%CP+%CL+%moisture+%ash+%CF); ³ Energy = {(protein x 4.5 kcal g⁻¹)+(lipid x 9.1 kcal g⁻¹)+(NFE x 3.5 kcal g⁻¹)/100 (NRC 1993); ⁴ DE = digestible energy.

Fish and experimental conditions. Feeding trial in red tilapia was done for 60 days in 2019. The red tilapia with total length of 9 to 10 cm were reared in a fiber tank measuring 50x50x70 cm³ with 15 units with rearing density of 15 fish/tank. The fish was fed twice daily at 8:00 AM and 2:00 PM with feeding rate of 3% of total biomass. The research was conducted at the Research Station in Department of Fisheries, Faculty of Agriculture, Universitas Gadjah Mada, Indonesia. The growth of red tilapia was checked on days 0, 15, 30, 45, and 60, by measuring the absolute weight of the fish.

Feces collection and feed digestibility measurements. The red tilapia was adapted to the treatment containers for five days. The collection of the red tilapia feces was done for 14 days treatment. The feed treatment containing Cr₂O₃ 0.6% was given to the red tilapia twice daily at 8:00 AM and 2:00 PM with a 3% of total biomass. The feed digestibility test was done through an indirect method (Takeuchi 1988). Before feeding time, the red tilapia feces were retrieved by using a siphoning hose from each container. The feces were stored in a labeled bottle in a freezer. The feces and feed were analyzed for the protein and Cr₂O₃ content. The protein content was analyzed by using a micro-Kjeldahl method, while the destruction method was used to determine the Cr₂O₃ in the feces and feed through detection by using a spectrophotometer (Spectonic Genesys 20 Visible) with a wavelength of 350 nm.

Data calculation. Final weight, specific growth rate, feed conversion ratio, feed efficiency, protein efficiency ratio, feed digestibility, and survival rate were calculated by using the following formulas:

$$\text{Final weight (g)} = W_t - W_o$$

where: W_t = fish weight at the end of the cultivation;
 W_o = fish weight at the beginning of the cultivation.

$$\text{Specific growth rate (\% day}^{-1}\text{)} = \frac{\ln W_t - \ln W_o}{t} \times 100$$

where: W_t = fish weight at the end of the cultivation;
 W_o = fish weight at the beginning of the cultivation;
 t = cultivation time (day).

$$\text{Feed conversion ratio} = \frac{F}{W_t - W_o}$$

where: F = dry weight of the feed given during the cultivation;
 W_t = fish weight at the end of the cultivation;
 W_o = fish weight at the beginning of the cultivation.

$$\text{Feed efficiency (\%)} = \frac{W_t - W_o}{F} \times 100$$

where: W_t = fish weight at the end of the cultivation;
 W_o = fish weight at the beginning of the cultivation;
 F = dry weight of the feed given during the cultivation.

$$\text{Protein efficiency ratio} = \frac{\text{Additional growth rate}}{\text{Amount of protein consumed}}$$

$$\text{Feed digestibility (\%)} = 100 - \left[100 \times \frac{a}{a'} \times \frac{b}{b'} \right]$$

where: a = Cr₂O₃ in feed (%);
 a' = Cr₂O₃ in feces (%);
 b = nutrients (protein) in feed (%);
 b' = nutrients (protein) in feces (%).

$$\text{Survival rate (\%)} = \frac{N_t}{N_o} \times 100$$

where: N_t = total fish at the end of the cultivation;
 N_o = total fish at the beginning of the cultivation.

Water quality parameters measurement. Measurement of water quality was carried out on days 0, 15, 30, 45 and 60 by measuring dissolved oxygen (DO), pH, water temperature, alkalinity and ammonia. Measurement and analysis of water quality was carried out at 08.00 am. Temperature was measured using maximum-minimum thermometer, pH was measured by a pH meter, DO and alkalinity were measured by a DO meter and ammonia was obtained by titration.

Data analysis. All of the data were analyzed with a one-way variant analysis (ANOVA) using SPSS version 20. Duncan's Multiple Range Test was used to compare significant differences between treatments. The treatment effects were considered with a significance level of $p < 0.05$.

Results and Discussion

Effect of fermentation on nutrient content of Moringa leaves meal. The MLM fermentation used mixture of T2A (*Bacillus* sp.), T3P1 (*Bacillus* sp.), and JAL11 (*Lactococcus raffinolactis*) and produced a various nutrient content profile. The FMLM moisture content was $13.33 \pm 1.32\%$; similar research was reported by Moyo et al (2011), Steven et al (2015), and Nweze et al (2014). The results indicated that the increase in moisture was due to the dry matter loss during fermentation process. Fermentation significantly increased the ash content ($p < 0.05$) from 9.70 ± 0.21 to $14.05 \pm 0.25\%$. The high ash level indicates that the leaves contain substantial inorganic elements (Biel et al 2017), because the ash represents the mineral content in plants (Ihedioha & Okoye 2011). Valdez-Solana et al (2015) mentioned that a high ash content in leaves demonstrates that the elements are good inorganic mineral sources. The increase in the ash content is due to the loss of dry material in the fermentation, which causes a relative increase in the components which do not change from the fermented product, especially the fiber and ash content (Imelda et al 2008).

Fermentation increased the protein content from 25.77 ± 0.08 to $34.77 \pm 0.09\%$. The significant increase ($p < 0.05$) in the protein content in MLM during the fermentation is possibly related with the microorganism activity, since there is efficient bioconversion in the polymerized carbohydrates to become microbial protein and produce various kinds of enzymes that are proteins in nature (Vijayakumar 2003; Bhatnagar 2004). Fermentation also increased the total lipid content from 4.80 ± 0.52 to $6.25 \pm 0.13\%$. This total lipid content is lower than previous research that recorded 7.90% (Richter et al 2003), 6.50% (Moyo et al 2011), and 8.50% (Ganzon-Naret 2014). Fermentation process decreased the MLM crude fiber to be $8.24 \pm 0.56\%$. This is due to the ability of mixture bacteria to degrade the crude fiber as plant cell wall components, even though there is secretion in several enzymes (Hassaan et al 2015). Fermentation reduces the NFE content from 39.09 ± 0.18 to $23.36 \pm 0.47\%$. This reduction is due to NFE utilizing for growing bacteria. In other words, the microbial cell masses change NFE feed to become protein to be a total protein content in FMLM (Table 1).

FMLM contained calcium (Ca) and phosphorous (P) in a high amount that is appropriate to be used as feed ingredient. The Ca and P content (Table 1) demonstrates that FMLM is higher than the Ca and P in unfermented MLM. MLM is reported to have a high mineral content, especially Ca comparing to other plants (Nkafamiya et al 2010; Moyo et al 2011). Mbailao et al (2014) claimed that Ca and Mg are the primary mineral components in MLM, while P finds in a smaller portion, as well as K and Na. Ca/P (> 2.00) is indicating potential nutritional benefits of the Moringa leaves (Ijarotimi et al 2013). In general, Ca from feed ingredient which originates from nature supplies enough Ca to fulfill most of the needs of finfish.

Effect of fermentation on antinutrient content of Moringa leaves meal. The fermentation process was able to reduce the anti-nutrient substances in MLM. The phenol, tannin, phytic acid, and HCN content in FMLM were lower than in unfermented MLM (Table 1). The analysis results revealed that fermentation with bacteria (a mixture between T2A (*Bacillus* sp.), T3P1 (*Bacillus* sp.), and JAL11 (*Lactococcus raffinolactis*) was able to reduce the anti-nutrient substances in MLM. It is known that the concentration of antinutritional factor/ANF (anti-nutrient) is generally reduced after processing, particularly using fermentation and germination techniques (Nkafamiya et al 2010).

The presence of anti-nutrients in MLM revealed that bacteria was able to degrade anti-nutrients. Previous publications reported that boiling, soaking, fermenting, and roasting are able to reduce the anti-nutrient content in Moringa leaves (Steven et al 2015). Therefore, using plant materials in livestock feed must consider the toxicous chemical materials that are contained in the basic materials for animal feed (Siddhuraju et al 2000). The phytic acid content in FMLM is lower than fish feed that originates from supplementary plant harvests like soybean meal (1.00 to 1.50%), rapeseed meal (5.00 to 5.75%), and sesame meal (2.40%) (Francis et al 2001). A phytic acid level in fish feed that has a concentration about 0.5% is disadvantageous for fish growth (Francis et al 2001). The total phenol, tannin, and HCN are significantly higher in FMLM, but the levels are still much lower than *Gliricidia sepium* leaves meal, which has the highest phenol (7.71%) and tannin (7.26%) levels (Mutayoba et al 2011). The high phenol total needs to be further evaluated, because it could possibly have an effect on animal health (Mutayoba et al 2011). In contrast, the absolute limit for cyanide toxicity for fish has not been recorded, and further research is needed to determine the level of cyanide tolerance for fish (Francis et al 2001).

Effect of fermentation on nutrient content of fish diets. The variance analysis results showed that replacing fish meal with FMLM in diets have significant results ($p < 0.05$) for protein, total lipid, ash, crude fiber, and NFE levels, while the moisture content results were deemed to be insignificant ($p > 0.05$) (Table 2). The protein, total lipid, ash, and crude fiber feed produced were in accordance with Indonesian National Standard 01-7242/2006, which is protein content $\geq 25\%$, total lipid content $\geq 5\%$, an ash content $\leq 15\%$, and crude fiber content $\leq 6\%$. The feed moisture content has a range between 13 and 15%, which is higher than the fish feed standard of 12%. The higher percentage of replacing fish meal with FMLM in feed actually reduces the moisture content, even though the value was still above 12%.

Growth performance of red tilapia. The weight of the red tilapia in all treatments increased in line with the cultivation time period (Figure 1). During 15 days of feeding trial, it was seen that the red tilapia started to show a weight increase. From the 30th until the 60th days, the weights of the red tilapia at 0, 10, and 30% were more dominant compared to the treatment of 20%. Replacing fish meal with FMLM in feed at 0% actually produced the highest weight in red tilapia followed by the weight of tilapia fed with 10% treatment. This depicts that replacing fish meal with FMLM in feed influenced slightly the growth of red tilapia.

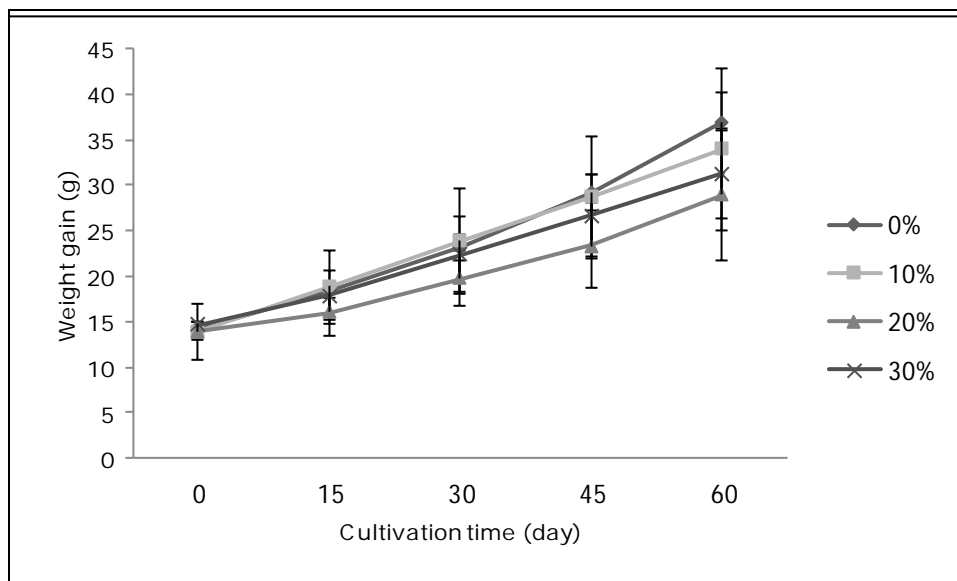


Figure 1. The weight gain of red tilapia for 60 days cultivation.

Although the nutrient levels already match with the red tilapia needs, the variance analysis results convey that replacing fish meal with FMLM in feed did not significantly influenced ($p > 0.05$) the specific growth rate, feed conversion ratio, feed efficiency, protein efficiency ratio and fish survival of red tilapia. The growth performance parameters of red tilapia were shown in Table 3.

Table 3
Growth performance of red tilapia fed diets containing various levels of fermented Moringa leaves meal (FMLM) for 60 days

Parameter	FMLM levels			
	0%	10%	20%	30%
Initial weight (g)	14.20±0.81 ^a	13.87±0.31 ^a	13.93±0.92 ^a	14.71±0.27 ^a
Final weight (g)	22.79±2.82 ^a	20.07±5.47 ^a	18.32±3.42 ^a	16.53±4.67 ^a
SGR (% day ⁻¹)	1.59±0.12 ^a	1.48±0.07 ^a	1.18±0.50 ^a	1.24±0.24 ^a
Feed conversion ratio	1.33±0.06 ^a	1.38±0.06 ^a	1.41±0.03 ^a	1.32±0.08 ^a
Feed efficiency (%)	74.75±3.55 ^a	72.50±3.36 ^a	70.88±1.78 ^a	75.85±4.94 ^a
Protein efficiency ratio	1.21±0.35 ^a	1.11±0.15 ^a	1.61±1.03 ^a	1.45±0.28 ^a
Feed digestibility (%)	34.13±0.31 ^a	31.12±7.85 ^a	47.48±0.36 ^b	65.08±1.32 ^c
Survival rate (%)	99.11±1.98 ^a	100.00±0.00 ^a	100.00±0.00 ^a	98.66±1.98 ^a

Different superscripts on the same row reveal a significant difference ($p < 0.05$)

The feed treatment did not significantly influenced ($p > 0.05$) the specific growth rate. This depicts that the energy from FMLM in feed as a replacement for fish meal can be utilized by red tilapia, but it only has a small influence towards the growth of red tilapia. The results show that it is possible to replace 30% of fish meal with FMLM with only limited effect on red tilapia growth performance. This condition is suspected because of the presence of anti-nutrient substances in FMLM. Anti-nutrient substances can cause a low level of feed digestibility (Kokou & Fountoulaki 2018).

The feed conversion ratio in this study ranged from 1.32±0.08 to 1.41±0.03 (Table 3). The values are higher than that reported by Richter et al (2003) and Yuangsoi & Masumoto (2012), but still lower than that reported by Karina et al (2015). The feed conversion ratio values reveal the advantages of feed nutrients by fish. The low consumption of feed (feed intake) during fish cultivation is a result of feed adaptation. The amount of feed consumption is influenced by the feed palatability. Palatability of feed will affect the process of searching, taking and ingestion of feed by fish related to the nutrient content. The lower the feed conversion ratio values are, then the feed usage will be more efficient. This is directly proportional with the feed efficiency value.

Feed efficiency is the proportion of additional fish biomass with the amount of feed consumed by fish. In this study, the feed efficiency values ranged from 70.88 ± 1.78 to $75.85 \pm 4.94\%$. The high level of feed efficiency value depicts that feed has good quality, so that red tilapia utilized feed optimally. This feed efficiency value is higher than the feed application with fermented *Lemna minor* in *O. niloticus* (Pinandoyo et al 2019). This study resulted in the protein efficiency ratios in the range of 1.11 ± 0.15 to 1.45 ± 0.28 . The protein efficiency ratio values are influenced by the ability of red tilapia to digest feed. This ability is influenced by several factors, including the feed composition (Pinandoyo et al 2019). The present study results show that the feed digestibility values are in the range of 31.12 ± 7.85 to $65.08 \pm 1.32\%$ implying a low level of digestion. The low level of feed digestion is due to the presence of anti-nutrient substances in FMLM. Kokou & Fountoulaki (2018) stated that anti-nutrient substances reduce the level of feed digestibility. A feed is categorized to have a good digestibility if the values range from 75 to 95% (NRC 2011).

Replacing fish meal with FMLM in diets significantly influenced ($p < 0.05$) feed digestibility (Table 3). The mixture of bacteria has proteolytic, amylolytic and cellulolytic activities so that it can produce protease, amylase and cellulase enzymes that can help break down protein, carbohydrates, cellulose into simpler forms so that it can play a role in maintaining microbial balance in the fish digestive tract. The survival rate in these treatments ranged from 98.66 ± 1.98 to $100.00 \pm 0.00\%$, higher compared to research carried out by Karina et al (2015) and Bhole et al (2016) with survival rate of $93.33 \pm 5.77\%$ and 93.33% , respectively. The high level of the survival rate indicated that red tilapia was able to tolerate the diets containing various levels of FMLM.

Water quality. Water quality is one of the supporting factors for survival rate and growth. The temperature ranged from 28.00 to 28.38°C, pH ranged from 7.14 to 7.20, dissolved oxygen ranged from 4.56 to 5.08 mg L⁻¹, total alkalinity ranged from 129.79 to 131.45 mg L⁻¹, and ammonia was 0.01 mg L⁻¹. The water quality was in the desirable ranges for red tilapia cultivation in this study (El-Sayeed 2006; SNI-7550-2009; Cavalcante et al 2014).

Conclusions. Fermentation increases the protein, ash, total lipid, calcium, and phosphorous contents in Moringa leaves meal (MLM). The fermentation process actually reduced the anti-nutrient elements (phenol, tannin, phytic acid, and HCN) in MLM. The replacement of fish meal with fermented Moringa leaves meal (FMLM) in diet has a significant influence towards feed digestibility. FMLM can be used as a replacement for fish meal in diet at 30% with only limited effect on the growth performance of red tilapia.

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