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The growth rate of Nile tilapia *Oreochromis niloticus* fry fed on fermented *Lemna* sp. meal

Laju pertumbuhan benih ikan nila *Oreochromis niloticus* yang diberi pakan mengandung tepung *Lemna* sp. fermentasi

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ABSTRACT

This research aimed to evaluate the supplementation of fermented *Lemna* sp. meal (FLM) in the artificial diet on the growth rate of Nile tilapia *Oreochromis niloticus*. The research design used was complete randomized design consisting of four treatments, namely A (0% FLM), B (20% FLM), C (30% FLM), and D (40% FLM), performed in triplicates. Experimental fish used was Nila tilapia at the initial size of 5−7 cm, fed with feed containing 18% protein content. The parameters observed were the nutritional composition of FLM, specific growth rate (SGR), feed conversion ratio (FCR), survival, and water quality. Fermentation with Effective Microorganism 4 (EM4) has increased crude protein and crude lipid of *Lemna* sp. meal, respectively, by 5.60% and 5.76%. However it decreased its crude fiber content down to 15.27%. The result suggested that supplementation of FLM 0–40% in the artificial diet could give SGR 0.81–1.20%/day, FCR 2.48–2.97; and survival 72%–84% in Nile tilapia. The addition of 40% LFM showed the best result among all treatments.

Key words: *Lemna* meal, EM4, fermentation, Nile tilapia fry

ABSTRAK

Tujuan dari penelitian ini adalah untuk mengevaluasi penggunaan tepung *Lemna* sp. fermentasi (TLF) pada pakan buatan terhadap laju pertumbuhan ikan nila *Oreochromis niloticus*. Rancangan penelitian yang digunakan adalah rancangan acak lengkap terdiri atas empat perlakuan, yaitu A (0% TLF), B (20% TLF), C (30% TLF), dan D (40% TLF) dengan masing-masing tiga ulangan. Ikan uji yang digunakan adalah ikan nila dengan ukuran awal 5−7 cm yang diberi pakan mengandung protein 18%. Parameter yang diamati dalam penelitian ini adalah nilai nutrisi TLF, laju pertumbuhan spesifik (LPS) ikan nila, rasio konversi pakan (RKP), sintasan (SR), dan kualitas air. Fermentasi dengan *Effective Microorganism* 4 (EM4) meningkatkan protein kasar dan lemak kasar tepung *Lemna* sp. sebesar masing-masing 5,60% dan 5,76%, akan tetapi menurunkan serat kasar hingga 15,27%. Hasil penelitian menunjukkan bahwa pemberian TLF 0–40% dalam pakan komersial menghasilkan LPS 0,81–1,20%/hari; RKP 2,48–2,97; dan kelangsungan hidup 72%–84%. Pemberian TLF sebanyak 40% merupakan perlakuan yang menunjukkan hasil terbaik.

Kata kunci: tepung *Lemna*, EM4, fermentasi, benih ikan nila

INTRODUCTION

One of the critical success factors in aquaculture is the growth rate of the fish. The growth is defined as the increasing size, weight, and length within a certain time. Growth occurs when there is surplus input of energy and amino acids (proteins) derived from feed. Acquired energy is used for basic metabolic maintenance (i.e. respiration, specific, and dynamic action), excreted out of the body, or available as surplus energy. This excess energy afterward is allocated for gaining weight (somatic growth), gonadal maturation and reproduction, or being stored in muscle, liver, and mesenteric fat (McBride *et al*., 2015). Feed remains one of important factors in fry culture. Nutritional composition of feed given during fry rearing has a significant influence to the fish growth in the following phases.

Feeding cost in aquaculture accounts for over 50% of the total production cost (Rostika, 2009; Yousefian & Amiri, 2009). Reducing fish meal is an important issue in the aquaculture industry because of its increased price (Lim & Lee, 2011). Replacing the amount of fish meal in diet formulation with alternative local feed material is one of solutions (Kader *et al*., 2012). Fish feed components on one hand are suggested to be more affordable and not causing competition to human consumption need, while in the other hand have to contain sufficient nutrition in fine quality for fish growth.

Duckweeds are small floating aquatic plants (Talukdar *et al*., 2012; Kabir *et al*., 2009; Uddin *et al*., 2007). This plant may benefit for the fishery business as a chep and abundant source of feed component. Ansal *et al*. (2010) reported that the productivity of *Lemna* sp. planted in an effective planting system reach to 12−38 tons of dry weight/ha/year. *Lemna* sp. has high protein content (38%), however it also rich in crude fiber content. Unfortunately, this may inhibit the digestive and absorbing abilities in the fish's digestive system.

Effective Microorganism 4 (EM4) was a mixed culture of various beneficial microorganisms that includes fermented and synthetic microorganisms containing lactic acid, photosynthetic bacteria, *Actinomycetes* sp., *Streptomycertes* sp., yeast, and cellulose decomposing fungus (Ariyanto, 2013; Nur *et al.,* 2016). Fermentation process using EM4 can reduce the amount of crude fiber of a feed material (Sihotang, 2012). Fermentation of *Lemna* meal is expected to lessen its crude fiber content thus making *Lemna* potential for being a fish feed compound. The aim of this study was to evaluate the supplementation of fermented *Lemna* meal (FLM) in commercial fish feed on the growth rate of Nile tilapia *Oreochromis niloticus*.

MATERIALS AND METHODS

Fermented *Lemna* meal (FLM)

Lemna sp. was yielded from culture pond, and sun-dried for one day. Next step was drying *Lemna* in an oven at the temperature of 60 °C. *Lemna* was then formed into meal, and was analyzed for its nutritional composition (proximate analysis) based on method carried by Takeuchi (1988).

Fermentation solution was made by diluting EM4 in water at the ratio of 1:100. EM-4 used contained *Lactobacillus casei*, *Saccharomyces cerevisiae*, and *Rhodopseudomonas palustris* and produced by PT. Songgolangit Persada. Solution then was mixed with *Lemna* meal at the ratio of 3:10, and well mixed. The mixture was then stored in a plastic bag at the temperature of 29 °C for seven days according to method performed by Handajani (2011). Sour smell was an indicator that *Lemna* meal has been fermented. The FLM was then sun-dried and analyzed for its proximate composition.

FLM was added to commercial feed at a level to the treatment dose. The commercial feed produced by PT Central Pertiwi at protein content of 18% was used as experimental feed. As a binder, albumin of chicken egg was used. Mixture was then formed into pellets at suitable size for experimental fish and was sampled for proximate analysis.

Fish rearing

The experimental fish used in this study was taken from Sumedang, West Java, at the initial size of 5−7 cm. At the first step, aquariums were washed with soap and potassium permanganate (PK), and were then rinsed, and dried. Water was filled into the aquariums and was kept for several days. Experimental fish were then acclimatized in a particular tank with no feed was given for one day. The initial weight of fish was measured before they were moved into aquariums.

Fish were fed three times in a day at 06:00 am, 12:00 pm, 16:00 pm at satiation. Faeces and other waste were taken by daily siphoning. Fish weight was monitored every seven days. Water was changed every week. Fish were reared for 42 days.

Research design

Present study was conducted using complete randomized design, consisted of four treatments in three replications, as follows:

A: commercial feed (control)
B: commercial feed 80% and FLM 20%
C: commercial feed 70% and FLM 30%
D: commercial feed 60% and FLM 40 %

Evaluating parameters

Nutritional content of expereimental feed

Nutritional contents of FLM and experimental feed used in this experiment were obtained based on proximate analysis.

Survival

Survival was calculated using formula as follows :

$$\text{Survival (\%)} = Nt / No \times 100$$

Specific growth rate

Specific growth rate of fish was counted as follows:

$$\text{SGR (\%/day)} = (\ln Wt - \ln Wo) / t \times 100$$

Feed conversion ratio

Feed conversion ratio was measured with formula as follows:

$$FCR = F / [(Wt + D) - Wo] \times 100$$

Note:

Survival = fish survival (%)

Nt = number of shrimp at the end of the experiment (individual)

No = number of shrimp at the beginning of the experiment (individual)

SGR = specific growth rate (%/day)

Wt = the average weight of the fish at the end of the study (g)

Wo = the average weigth of the fish at the beginning of the study T_1 (g)

t = rearing period

F = total of feed given during rearing period (g)

D = biomass of dead fish (g)

Data analysis

Data were collected from periodical samplings and analyzed using oneway ANOVA and using a statistical software SPSS 17.

RESULTS AND DISCUSSION

The process of fermentation using EM4 has caused the conversion of nutritional value of the *Lemna* meal involving the increase of the moisture, crude protein, and crude lipid as much as 91.15%, 5.60%, and 5.76% respectively (Table 1). However the fermentation process decreased the ash content for 3.93% and the crude fiber at the percentage of 15.27%.

The increase of crude protein level in present research was less compared to Warasto *et al*. (2014) that has successfully reached percentage of 60% in fermentation of kiambang *Salvinia molesta* with EM4. This result was predicted as caused by the difference content of EM4 and plants used. Warasto *et al*. (2014) used EM4 which contained photosynthetic bacteria, lactic acid bacteria, yeasts, and fungi, meanwhile EM4 this study did not contain fungi.

However similar result was found in Bairaqi *et al*. (2002) who has FLM with the help of *Bacillus* sp. A decrease in crude fiber, crude protein, and tannins content were obtained as the results. According Sitohang (2012) the different proximate compositition of a material after fermentation was caused by the activity of cellulase enzymes contained in microbs. In previous study Santoso and Aryani (2007) stated that EM4 produce large amounts of crude fiber digesting enzymes such as cellulases and mannase (Solfan & Rosmaina, 2010; Muhakka *et al.,* 2015).

Cellulolytic microbes using cellulose as a source of carbon and energy by producing cellulase enzymes that can break down, and degrade the cellulose component and its derivatives into glucose. Bairaqi *et al*. (2002) stated that fermentation of *Lemna* meal produces a decline in crude fiber and crude protein content by 31% and 38% respectively. This suggested that fermentation is an efective way to reduce the content of crude fiber in a plant-derived feed material.

The decrease in the ash content to 3.39% in the FLM was closely related to the decrease in the crude fiber content of the feed. The crude fiber content in the FLM reduced 15.27% as a result of the activity of the cellulose enzyme contained in *Saccharomyces cereviseae* (Sihotang, 2012). The

Table 1. Results from proximate analysis of fermented *Lemna* meal (FLM)

Nutrient	Before fermentation (%)	After fermentation (%)	Difference (%)
Moisture	6.49	13.61	+91.15
Ash	14.48	13.91	-3.93
Crude protein	24.00	25.35	+5.60
Crude lipid	7.81	8.26	+5.76
Crude fiber	15.19	12.87	-15.27

amount of crude fat in FLM increased up to 5.76% after fermentation. The increase of fat content was caused by the microorganism's capability to produce lipid / fat during fermentation process. Microorganisms, as other life cell systems, produce lipid or fat.

The SGR value was not influenced by the treatment of FLM (Table 2). It means that FLM could produce the same growth rate as the control feed (without supplementation of FLM) did. The most problem, a plant and algae-derived alternative feed material commonly faces are the crude fiber and anti-nutritional content (Suarez *et al*., 2009; Ncha *et al*., 2015; Suprayudi *et al*., 2015). There factors are the major causes for the low level of feed digestibility in fish which eventually could lower fish growth performance. Similar to this statement, Bairagi *et al*. (2002) proposed that factors that reduce the fish growth rate when they are fed with fermented meal content in the feed was the amount of crude fiber and other anti nutritions in the feed. It was because the fish do not have cellulose enzyme to hydrolyze the cellulose.

In analysis of FCR, no significant difference was found in all treatments (Table 2). The FCR level in this study varied from 2.48±1.91 in treatment C to 5.20±2.19 in control treatment. Even it was not markedly different with other treatments, the FCR shown in treatment control was tremendously higher than study conducted by Ayuningtyas *et al*. (2015) 1.53 and Vinasyiam *et al*. (2016) 1.72. This could be explained as the result of the lower SGR than other Nile tilapia in some studies or because of other reasons which were not further analyzed at present experiment i.e. fish appetite, feed texture and taste, slow digestion and absorption process rate in gut.

Determination of best treatment needed a further approach for instance the feed cost analysis (Table 3). Based on the mean value of feed conversion ratio, the minimal cost to produce 1 kg of fish was obtained in treatment C which was the addition of 30% FLM in commercial feed. Regarding to the feed cost analysis, it could be stated that the most efficient treatment was C which showed the minimum cost (Rp21,824,-) to produce the similar SGR and FCR. However treatment C surprisingly had the least survival (72%) among all treatments (72–84%) (Figure 1), thus making treatment D with feed cost Rp25,452,- and survival (77%) became the best treatment in this study. No sufficient data was obtained to explain the reason of survival result. Further study was needed to investigate the relation of addition FLM at particular level to the mortality of fish.

CONCLUSION

Present study concluded that the fermentation of *Lemna* meal using EM4 reduced the content of crude fiber to 15.27%, increases the crude protein to 5.6%, and increases the crude fat to 5.76%. The supplementary FLM to 40% in commercial feeding resulted the best treatment among all treatments because it showed the highest survival, lower feed cost, while did not

Table 2. The specific growth rate (SGR) and feed conversion ratio of Nile tilapia fry

Treatment	Specific growth rate (%/day)	Feed conversion ratio
A	0.81±0.13a	2.97±2.19a
B	1.20±0.48a	3.15±1.04a
C	1.10±0.58a	2.48±1.91a
D	0.83±0.60a	3.03±1.76a

Note: different letter in the same column indicates significant difference.

Table 3. Feed cost analysis

Treatment	Feed price per kg	Feed cost*
A	Rp10,000,-	Rp29,700,-
B	Rp9,200,-	Rp28,980,-
C	Rp8,800,-	Rp21,824,-
D	Rp8,400,-	Rp25,452,-

Note: *= total feed cost to produce 1 kg of fish based on mean value of feed conversion ratio.

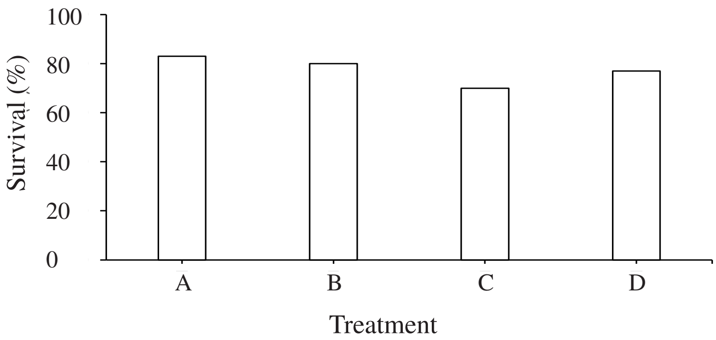

Figure 1. The average of survival Nile tilapia fry. Note: A=0% FLM, B=20% FLM, C=30% FLM, and D=40% FLM)

show significant difference of daily growth rate and feed conversion ratio from control (without fermented *Lemna* meal).

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