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Utilization of Ambon Banana (*Musa acuminata*) Peel Flour as a Prebiotic in the Nile tilapia (*Oreochromis niloticus)*

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ABSTRACT

The Banana peel is a waste with quite a high nutritional content but rarely utilized, so it needs to be tested for fish farming. This study aimed to evaluate the potential of the Ambon banana (*Musa acuminata*) peel flour (ABPF) as a prebiotic in the Nile tilapia (*Oreochromis niloticus*). Fifteen Nile tilapia fry with an average weight of 2.11 ± 0.18 g were reared for 28 days in a volume of 30 L. This study used a completely randomized design with four treatments and three replications. Experimental fish were fed feed with the addition of ABPF of 0% (control), 1%, 2%, and 3% kg^{-1} of feed, twice a day at satiation. The data obtained in this study were: hematological parameters, survival rate, growth performance, feed utilization, the number of lactic acid bacteria (LAB) in the intestine, and digestive enzyme activity statistically analyzed using a one-way ANOVA test and followed with Duncan's test. The results showed that adding ABPF to Nile tilapia feed significantly affected fish health, especially the levels of hematocytes, total leukocytes, monocytes, and neutrophils were higher than the control treatment to maintain a better survival rate. The final weight and absolute growth of Nile tilapia were significantly different from the control. The amount of LAB and digestive enzyme activity of Nile tilapia fed with the addition of ABPF were higher than the control. These results indicate that ABPF may be prebiotic in the Nile tilapia.

INTRODUCTION

At the present time, the science of nutrition for livestock is growing. Additives with high nutritional value in feed are known to bring positive benefits to health and growth. Prebiotics are one of the additives that are nutraceutical and functional and have been extensively studied for their effects. Prebiotic is a non-digestible and selectively fermented material, which allows for certain changes in both the composition and activity of the microbiota in the digestive tract, which in turn provides benefits to the host, including improving their health **(Synytsya *et al*., 2009)**. In addition, prebiotics serve as food for probiotics. According to **Abatenh *et al*. (2018)**, probiotics are microbes that are able to live in the digestive tract; if it is given in sufficient quantities, it will support health and increase the utilization of host nutrients.

Bananas, which belong to the Musaceae family, are one of the fruits commonly found in tropical and sub-tropical regions. They are preferred because of their sweet taste and high nutritional content. The Central Bureau of Statistics (2015) stated that banana production is increasing yearly in Indonesia. Banana production in 2014 reached 6,862,588 tons. High banana production was also accompanied with an increase in the banana processing industry, but the waste from the banana processing industry has not optimally been used. The banana peel is rarely used in large enough quantities, which is about 1/3 of a whole banana and has the potential to become organic waste. In contrast, the nutritional content is not inferior to the fruit's flesh. According to **Emaga *et al*. (2007)**, banana peel is a food source rich in starch (3%), crude protein (6-9%), crude fat (3.8-11%), total fiber (43.2-49.7%), and polyunsaturated fats, especially nutrient acids and linolenic acid, pectin, essential amino acids (leucine, valine, phenylalanine, threonine, and tryptophan), and macronutrients (K, P, Ca, Mg). The most common antioxidant components in banana peels are ascorbic acid, phenolic groups, tocopherols, ,beta-carotene, and gallocatechols **(Bankar *et al*., 2010)**. Banana peel is rich in phytate, alkaloids, oxalate, and hydrogen cyanide. However, according to the researchers, the concentration was within safe bounds, and industrial use is advised **(Abou-Arab & Abu-Salem, 2018; Oyeyinka & Afolayan, 2019)**.

Research on the use of banana peels in aquatic biota has been carried out and has shown positive results. Adding 2% banana peel flour in *Etroplus suratensis* fish feed could increase its specific growth rate **(Sreeja *et al*., 2013)**. A dosage of 5% banana peel flour gave the best final weight and specific growth rate for rohu fish (*Labeo rohita*) compared to control, the most significant increase in immune parameters such as lysozyme, alternative complement pathway, phagocytosis of leukocytes, superoxide dismutase, and catalase activity, as well as the level of the highest survival of 70% after being challenged with *A. hydrophila* bacteria. These results indicate that the banana peel flour diet at a dose of 5% could improve growth performance and strengthen immunity in *L. rohita* **(Giri *et al*., 2016)**. Another study showed that a 1% concentration of banana peel extract was able to produce a survival rate for catfish (*Pangasius hypophthalmus*) larvae of 86.96%, which is higher than that of 1.25% with a survival rate of 75.53**% (Haetami *et al*., 2019)**.

Research on the potential of banana peel flour as a prebiotic for the Nile tilapia (*O. niloticus*) needs to be done considering the increasing cultivation activities. Increased cultivation activities have the potential to face problems related to disease which result in decreased production. Ambon banana is widely grown in the city of Samarinda, East Kalimantan Province, and it is preferred because of its sweet taste. Ambon banana peels are thrown away as trash and no longer used. Based on this, the authors are interested in conducting a test by adding Ambon banana peel flour (*Musa acuminata*) in tilapia feed with doses ranging from 0-3% to determine the effect on health or hematological aspects, survival rate, growth performance, and feed utilization, amount lactic acid bacteria, and digestive enzyme activity of the Nile tilapia.

MATERIALS AND METHODS

Preparation of ambon banana peel flour and fish

Ambon banana peels were obtained from banana sellers in Samarinda City, East Kalimantan Province, Indonesia. Ripe Ambon bananas were peeled, cleaned, and dried at 60± 5ºC for 12 hours in an oven, ground or floured using a blender, and stored in an airtight bag for use in further tests **(Gupta *et al*., 2020)**. Nile tilapia specimens with an average weight of 2.11± 0.18g were obtained from a hatchery in Samarinda City. The water used was aerated and filtered in a plastic tub with a diameter of 1m which is then used to accommodate the fish during adaptation. Fish were given commercial feed twice a day till apparent satiation, and fish were then fasted for one day before being given treatment.

Diet preparation

The feed used in this study is commercial feed with a protein content of 39%. Mixing feed with Ambon banana peel flour (ABPF) was done after the feed was mashed. Mixing ABPF with respective doses of 0% (control), 1%, 2%, and 3% per kg of feed was carried out until it was homogeneous, then water was added as much as 20% of the weight of the feed. The ingredients have been mixed, then molded using a pellet machine, and dried in the oven at 50^{o}C for 2 hours. The dry pellets were then cooled at room temperature and stored in a container.

Treatment design, feeding and culture system

This study used a completely randomized design (CRD), consisting of four treatments and three replications: P0 = 0% ABPF kg feed^{-1}; P1 = 1% ABPF kg feed^{-1}; P2 = 2% ABPF kg feed^{-1} and P3 = 3% ABPF kg feed^{-1}. Fish were reared as many as 15 in each aquarium with a volume of 30L for 28 days and fed twice a day in the morning and evening till apparent satiation. The amount of feed given to the fish during this study was weighed for use in calculating the feed conversion ratio and protein efficiency ratio. The water quality parameters values for the rearing tanks were as follows: temperature ranged from 26.3-27.8^{o}C; pH ranged from 6.9-7.2; dissolved oxygen ranged from 5.32-5.67 mgL$^{-1,}$ and total ammonia nitrogen ranged from 0.06-0.08 mgL^{-1}.

Hematology assay

Fish hematological parameters were observed at the beginning and end of the study. The first step was taking a blood sample from the caudal vein using a syringe. Blood parameters were assessed for hemoglobin levels with the Sahli method using a hemometer as described by **Wedemeyer and Yasutake (1977)**. Moreover, hematocrit was assessed according to the method described by **Anderson and Siwicki (1995)**. In addition, for the analysis of red blood cells and white blood cells, the procedure of **Blaxhall and Daisley (1973)** was followed. The leukocyte differential was measured according to the method implemented by **Anderson and Siwicki (1995)**.

Survival rate

The survival rate of Nile tilapia was calculated using the number of survivors divided by the total number of initial fish multiplied by 100%.

Growth performance and feed utilization

Parameters of growth performance and feed utilization include parameters of final weight, absolute weight gain, daily growth rate, feed conversion ratio, and protein efficiency ratio which were measured based on the study of **Morais *et al*. (2001)**. Body proximate analysis was carried out at the beginning and end of the study; it was used to determine the nutrient composition of the fish as mentioned by **Takeuchi (1988)**.

Abundance of lactic acid bacteria and total bacteria from the intestines of test fish

Parameters of the abundance of lactic acid bacteria and total bacteria in the intestines of Nile tilapia were calculated based on the total number of bacterial colonies on MRSA and NA media, based on the total plate count method followed by **Lay (1994)**.

Digestive enzyme activity

Parameters of enzyme activity including α-amylase, protease, and lipase activity. Measurement of α-amylase enzyme activity were determined following the method of **Worthington (1993)**. Moreover, the measurement of protease enzyme activity followed the method of **Bergmeyer and Grassi (1983)**, and the measurement of lipase enzyme activity followed the method of **Borlongan (1990)**.

Data analysis

Observational data such as hematology parameters, survival rate, growth performance, feed utilization, and digestive enzyme activity were analyzed for diversity using ANOVA (analysis of variance) in the SPSS application at a 95% confidence level, and Duncan's follow-up test was carried out to determine differences between treatments. Fish body composition was analyzed descriptively in tabular form.

RESULTS

1. Hematology and survival rate of the Nile tilapia

Maintenance for 28 days by adding ambon banana peel flour (ABPF) with different doses had an effect on the parameters or hematological features (Table 1) and the survival rate of Nile tilapia (Fig. 1). The hematocrit level of Nile tilapia with the addition of ABPF was 2% higher than the control ($P < 0.05$), which indicates that the nutrients given to the fish are met and utilized efficiently to support their growth. Parameters of total leukocytes, monocytes, and neutrophils showed significant differences compared to the control ($P < 0.05$).

The survival rate of the Nile tilapia until the 28th day did not show a significant difference in all treatments, 93.33% in the control treatment and 3% ABPF; 95.56% at 1% and 2% ABPF treatment, respectively. Fish mortality was higher in the control treatment, and the addition of ABPF was 3% compared to 1% and 2%.

Table 1. The hematological features of Nile tilapia with a diet containing different levels of ABPF

Parameter and day	0% ABPF	1% ABPF	2% ABPF	3% ABPF
Hb 0 (gdL^{-1})	3,09±0,16[a]	3,09±0,16[a]	3,09±0,16[a]	3,09±0,16[a]
Hb 28 (gdL^{-1})	4,20±0,53[a]	5,73±0,81[a]	4,60±0,53[a]	6,13±1,60[a]
Hct 0 (%)	27,26±5,31[a]	27,26±5,31[a]	27,26±5,31[a]	27,26±5,31[a]
Hct 28 (%)	30,55±4,81[a]	36,51±5,50[ab]	41,54±2,29[b]	39,68±5,50[ab]
RBC 0 ($x10^6 mm^{3-1}$)	3,17±0,45[a]	3,17±0,45[a]	3,17±0,45[a]	3,17±0,45[a]
RBC 28 ($x10^6 mm^{3\ -1}$)	4,67±0,79[a]	5,22±1,26[a]	4,89±1,05[a]	4,64±0,41[a]
WBC 0 ($x10^4 mm^{3\ -1}$)	1,75±0,18[a]	1,75±0,18[a]	1,75±0,18[a]	1,75±0,18[a]
WBC 28 ($x\ 10^4 mm^{3\ -1}$)	1,75±0,01[a]	2,22±0,39[ab]	3,15±0,32[c]	2,44±0,28[b]
Lymphocytes 0 (%)	87,26±4,00[a]	87,26±4,00[a]	87,26±4,00[a]	87,26±4,00[a]
Lymphocytes 28 (%)	87,44±3,67[c]	66,31±4,99[ab]	69,51±0,84[b]	60,03±4,13[a]
Monocyte 0 (%)	8,45±2,88[a]	8,45±2,88[a]	8,45±2,88[a]	8,45±2,88[a]
Monocyte 28 (%)	8,87±3,00[a]	26,63±5,22[b]	25,75±1,66[b]	36,19±4,68[c]
Neutrophil 0 (%)	4,28±1,13[a]	4,28±1,13[a]	4,28±1,13[a]	4,28±1,13[a]
Neutrophil 28 (%)	3,68±0,69[a]	7,06±1,82[b]	4,72±1,12[a]	3,78±0,58[a]

Hb: Hemoglobin, Hct: Hematocrit, RBC: Red blood cell, WBC: White blood cell. The average number followed by the same letter in the same row is not significantly different ($P > 0.05$)

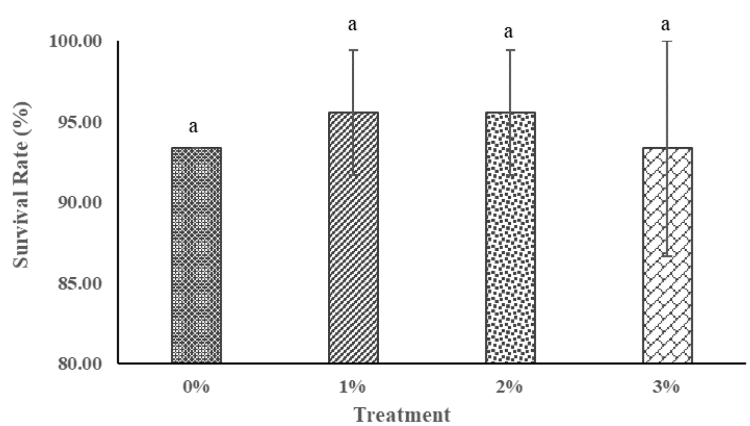

Fig. 1. The survival rate of Nile tilapia with different diets of ABPF levels

2. Growth performance and feed utilization

Table (2) shows that adding ABPF significantly affected several parameters of the growth performance of the Nile tilapia. The final weight parameter, absolute growth, was higher in the ABPF treatment by 1% and 2% compared to the control ($P < 0.05$). In contrast to growth performance, the addition of ABPF in feed at different doses did not show a significant difference in feed utilization for Nile tilapia fish compared to controls ($P > 0.05$), as shown in Table (2).

Table 2. Growth performance and feed utilization of Nile tilapia with diet containing different levels of ABPF

Parameter	0% ABFP	1% ABFP	2% ABFP	3% ABFP
Initial weight	31.60±5.20[a]	30.80±0.35[a]	30.90±0.35[a]	33.13±1.88[a]
Final weight	85.90±1.87[a]	96.80±2.62[b]	97.50±1.71[b]	92.93±7.10[ab]
Weight gain	54.3±6.56[a]	66.00±4.18[b]	66.60±2.03[b]	59.8±5.70[ab]
SGR	3.60±0.65[a]	4.09±0.28[a]	4.10±0.10[a]	3.68±0.17[a]
FCR	1.50±0.26[a]	1.30±0.16[a]	1.24±0.03[a]	1.41±0.09[a]
PER	1.74±0.29[a]	1.99±0.23[a]	2.06±0.05[a]	1.82±0.11[a]

SGR: Specific growth rate; FCR: Feed conversion ratio; PER: Protein efficiency ratio. The average number followed by the same letter in the same row is not significantly different (P> 0.05)

During 28 days of rearing, the body composition of Nile tilapia changed, both in the control group and with the addition of ABPF (Table 3). The addition of ABPF in feed tends to contain ash and nitrogen-free extract, while protein increases at a dose of 2% and decreases at a dose of 3% ABPF. Lipids increased at a dose of 1% ABPF and then decreased with decreasing ABPF doses in the feed.

Table 3. Body composition of Nile tilapia with a diet containing different levels of ABPF

Parameter	0th day	28th day			
		0% ABPF	1% ABPF	2% ABPF	3% ABPF
Ash (%)	19.30±0.44	16.16±0.39	16.52±0.40	15.39±0.64	15.83±0.65
Protein (%)	62.64±0.73	60.36±0.84	61.47±0.51	63.63±2.90	60.63±2.01
Lipid (%)	16.58±0.55	19.53±0.61	19.88±0.40	19.76±0.55	19.48±0.39
Fiber (%)	0.22±0.04	0.58±0.20	0.46±0.25	0.74±0.44	0.37±0.23
Nitrogen free Extract (%)	1.26±0.37	3.38±0.96	1.68±0.20	1.07±0.11	2.10±0.44

3. Abundance of lactic acid bacteria and total bacteria from the intestine of the Nile tilapia

The treatment of ABPF for 28 days on Nile tilapia fry was able to affect the abundance of lactic acid bacteria and total bacteria in the intestine (Table 4). Increasing the ABPF dose in this study showed a significant difference in the abundance of LAB and total gut bacteria of tilapia fry compared to the control (P< 0.05).

Table 4. Abundance of autochthonous lactic acid bacteria and total culturable autochthonous bacteria of Nile tilapia with a diet containing different levels of ABPF

Parameter and day	0.0% ABPF	1.0% ABPF	2.0% ABPF	3% ABPF
LAB 0 (x10^3 CFUmL^{-1})	1.53±0.29[a]	1.53±0.29[a]	1.53±0.29[a]	1.53±0.29[a]
LAB 28 (x10^3 CFUmL^{-1})	2.09±0.39[a]	2.27±0.72[a]	5.52±0.61[b]	7.24±1.70[b]
TB0 x10^6 CFU mL^{-1})	2.28±0.43[a]	2.28±0.43[a]	2.28±0.43[a]	2.28±0.43[a]
TB28 (x10^6CFU mL^{-1})	2.81±0.47[a]	7.31±1.66[b]	7.57±1.99[b]	8.40±1.33[b]

LAB: Lactic Acid Bacteria, TB: Total Bacteria. The average number followed by the same letter in the same row is not significantly different (P> 0.05)

4. Digestive enzyme activity

The digestive enzyme activity of tilapia seeds in this study, including α-amylase, lipase, and protease enzymes, showed significant differences with ABPF administration compared to the control ($P < 0.05$). The activity of the three enzymes increased with increasing doses of ABPF (Fig. 2). The activity of the α-amylase enzyme on day 28 ranged from 3.61-6.38 $IUmL^{-1}$, while the activity of the lipase enzyme ranged from 0.16 - 0.21 $IUmL^{-1}$. The protease enzyme activity ranged from 0.19 - 0.36 $IUmL^{-1}$.

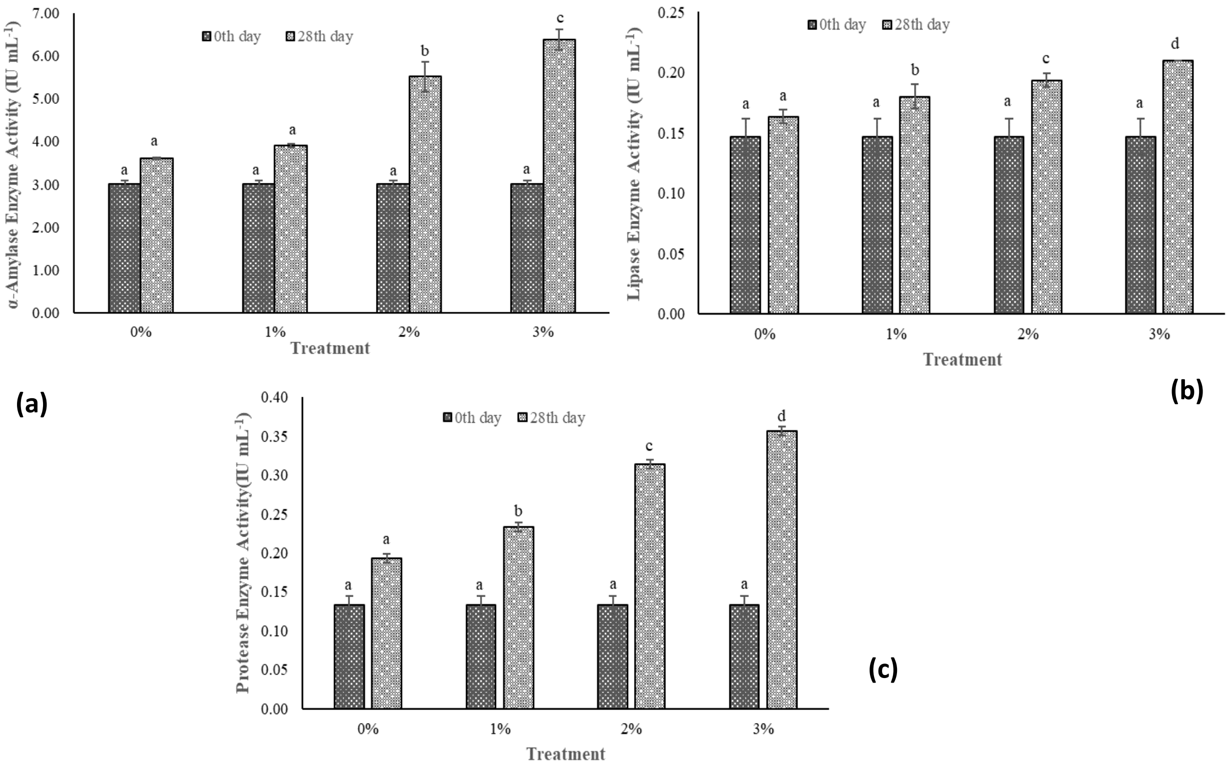

Fig. 2. Digestive enzyme activity of Nile tilapia with different diets of ABPF levels showing: **(a)** α-amylase enzyme activity; **(b)** Lipase enzyme activity, and **(c)** Protease enzyme activity

DISCUSSION

Hematological parameters are very important for determining physiological and pathological changes in fish. Blood parameters are important criteria that indicate physiological changes in fish farming activities and can provide important information for disease diagnosis and prognosis. Economic losses due to disease attacks in aquaculture systems can be reduced by early disease diagnosis through this hematological method **(Fazio, 2019)**. Changes in blood pictures and blood chemistry, both qualitatively and quantitatively, can determine the condition of the fish or its health status **(Baghizadeh & Khara, 2015)**. In general, the hematological parameters of Nile tilapia in this study were still within the normal range. This shows that adding ABPF in the Nile tilapia feed for 28 days did not interfere with its health. **Salasia *et al.* (2001)** stated that there was a correlation between the number of erythrocytes, hemoglobin, and hematocrit related to the health status, nutrition, and growth of fish. Parameters of total leukocytes, monocytes, and neutrophils are related to the non-specific immune response in fish. According to **Roberts (2012)**, monocytes as a type of leukocyte cell act as macrophages, which will phagocytize antigens that enter the fish's body. This indicates that adding ABPF, in addition to improving the health status of fish, also has the potential to increase the immune response of Nile tilapia.

In this study, it was found that adding ABPF to the Nile tilapia seed feed did not affect the fish's health, as evidenced by the survival rate, indicating its ability to support the fish's survival. These results are in line with research conducted **by Giri *et al*. (2016)**, where several parameters of immunity at a dose of 5% ABPF were also better to support survival after the juvenile rohu fish (*Lobeo rohita*) was challenged with *A. hydrophila* bacteria ($P < 0.05$). Moreover, the study aimed to assess the efficiency of fish feed supplemented with ABPF by examining mortality rates. This evaluation determined the impact of ABPF addition on fish survival.. The data obtained after 28 days of rearing showed that there was 6.67% mortality in control fish that were given control feed, while those treated with inulin had a lower mortality of around 4.4%. Therefore, it can be concluded that certain supplements contained in ABPF can reduce mortality in fish. Fish mortality predominantly happens during the transition from larval to fry stages due to significant physiological and ecological changes in fish development. During this period, larvae transition from relying on internal nutrients to external sources for nutrition. Rapid replacement of old functions by new ones occurs in the body during this crucial stage **(Shifa & Mathias, 1987)**. This transition might explain the 6.67% mortality observed in the control group

Fish growth is the increase in weight and body length of fish in a certain period. Physically, there is a change in shape due to an increase in length, weight, and volume within a certain time interval individually. Individual growth is a net increase due to cell division and excess input of energy and amino acids (proteins) derived from food **(Aliyas *et al*., 2016)**. These study results are in accordance with several previous studies stating that adding banana peel flour can improve fish growth performance. **Giri *et al*. (2016)**, found that adding banana peel flour (*M. acuminata*) to rohu fish (*Labeo rohita*) feed with an average weight of 15.3g at a dose of 5% produced the best final weight and specific growth rate during 60 days of rearing. Snakehead fish (*Channa striata*) fed with the addition of synbiotics, namely 6% prebiotics from banana peel flour and probiotics with a dilution of 10^{-4} colonies, showed better survival rates and growth performance compared to the control **(Mir & Singh, 2019)**.

According to **Emaga *et al*. (2007)**, banana peel is a food source rich in starch (3%), crude protein (6-9%), crude fat (3.8-11%), total fiber (43.2-49.7%), and polyunsaturated fats, especially nutrient acids and linolenic acid, pectin, essential amino acids (leucine, valine, phenylalanine, threonine, and tryptophan), and macronutrients (K, P, Ca, Mg). The nutritional content can be utilized in metabolizing Nile tilapia seeds to produce energy which is then used for growth. Growth occurs when the food nutrients digested and absorbed by the fish's body are greater than the amount needed to maintain the body. Energy sources from fats and carbohydrates can be utilized by fish to prevent protein catabolism into energy to increase energy used for growth **(Esteban, 2013; Rakhmawati *et al.*, 2018)**. An increase in the average body weight of fish suggests that the provided feed adequately fulfills the nutritional requirements, enabling efficient digestion and absorption within the fish's body. The similarity in feed efficiency parameters between the group supplemented with ABPF and the control group, as noted by **Karaket *et al.* (2021)**, suggests that fish can efficiently digest the high fiber content in the feed, likely facilitated by enzymes present in banana peels. The addition of ABPF to the feed up to a dose of 2% is thought to be able to support the process of utilizing energy from carbohydrates and fats, so that the protein content is higher than the control.

Substances that are considered prebiotics must have the following characteristics: (a) resistant to gastric acidity and not hydrolyzed by gastrointestinal enzymes, (b) cannot be absorbed by the gastrointestinal tract, and (c) selectively stimulates beneficial bacteria in certain amounts **(Teng & Kim, 2018)**. Prebiotics are feed ingredients in the form of fiber. This fiber can trigger an increase in beneficial bacteria for livestock, such as *Bifidobacteria*, thereby improving the host's health **(Manning *et al*., 2004)**. Oligosaccharides can act as prebiotics since they cannot be digested but are able to stimulate the growth of lactic acid bacteria (LAB) such as *Lactobacillus* and *Bifidobacteria* in the digestive tract **(Manning *et al*., 2004)**. According to **Emaga *et al*. (2007)**, banana peel is a food source that contains total fiber (43.2-49.7%); this makes banana peel flour potentially used as a source of stimulating the growth of good bacteria in the intestine. Prebiotic dietary fiber is a specific microbiota-forming compound that serves as a carbon source for the growth of beneficial bacteria, thus providing specific changes that confer host health related to their metabolism **(Carlson *et al*., 2018)**.

Fructooligosaccharides (FOS) are found in various foodstuffs, such as grains, fruits, vegetables, nuts, tubers, and other plant products. Hydrolysis or enzymatic processing of polysaccharides, such as starch and crude fiber, can also be obtained from FOS. Some prebiotics, such as FOS and inulin, play a role in improving health by modifying the balance of intestinal microflora **(Chrisanti, 2012)** and selectively stimulating the growth of beneficial bacteria such as *Lactobacillus* and *Bifidobacteria* **(Cumming *et al*., 2001)**. These specific carbohydrates serve as food for beneficial bacteria **(Patterson & Burkholder, 2007)**.

In a similar study, it was found that feed with the addition of banana peel flour of 2% +1% *Bacillus* probion in *Etroplus suratensis* fish with a maintenance period of 91 days showed an increase in growth performance, feed utilization, and an effect on microbial diversity in their digestive tract. In the intestine, *E. suratensis* fish were fed with banana peel flour and probion on 91 days of the experiment, genus such as *Bacillus* spp. higher than other genera, namely *Klebsiella* sp., *Pseudomonas* sp., *Micrococcus* sp., and *Enterobacter* sp. This indicates that the chances of good bacteria are greater in occupying the intestine by utilizing existing nutrients and reducing the number of potentially pathogenic bacteria **(Sreeja *et al*., 2016)**.

The abundance of bacteria with the addition of prebiotics was able to support the growth and utilization of feed. This was evidenced by the significant increase in body weight in *E. suratensis* fish which were fed additional food with banana peel flour by 2% and probion *Bacillus altitudinis* and *Bacillus licheniformis* each by 1% than the control, with a rearing period of 41 days. Weight percentage increased in fish with experimental feed compared to the fish with control feed. The consumption rates of the three experimental groups did not differ much, but the production rates varied significantly in the fish fed this diet. Probion given through feed can choose a place to attach to the intestine and prevent colonization by pathogens **(Sreeja *et al*., 2013)**. Giving banana peel meal via the feed did not affect the average final weight and daily growth rate of giant prawns but did affect the amount of feed consumption and feed efficiency as well as the final biomass. Banana peel meal contains FOS which are thought to play a role in the digestive process so that the amount of giant prawn feed consumption increases **(Rakhmawati *et al*., 2020)**. These prebiotics will be fermented by good bacteria, especially *Bifidobacteria* and *Lactobacillus*, and produce short-chain fatty acids that the body can use as an energy source **(Syafura *et al*., 2016)**. Ambon banana peels contain fructooligasaccharides (FOS) which have strong potential as prebiotics; they have the ability to increase the growth oflactic acid bacteria *Bifidobacterium bifidum*.

Banana peel flour contains a total phenol of about 25.1mgg^{-1} and about 9.03mgg^{-1} of flavonoids. ABPF’s prebiotic activity was tested *in vitro* against three types of bacteria *Lactobacillus plantarum*, *L. rhamnosus*, and *L. casei* **(Gupta *et al*., 2020)**. The results showed that tophianahat fructooligosaccharide isolated from banana peels could support the growth of lactic acid bacteria (*Bifidobacteria*), and it was concluded that FOS has the potential to be a source of prebiotics in livestock rations since it can support the growth of lactic acid bacteria *in vitro* **(Syahputra *et al*., 2019)**.

Fish digest nutrients in feed with the help of digestive enzymes, and then increase feed efficiency **(Widanarni *et al*., 2015)**. Digestive enzyme activity and changes related to prebiotic inclusion in a feed show different results depending on fish species. The results of this study are in line with the studies adding 2 and 4g kg^{-1} of Mannan-oligosaccharide (MOS) in *Atractosteus tropicus* fish larvae feed which increased lipase and α-amylase activity **(Maytorena-Verdugo *et al*., 2022)**. Meanwhile, digestive enzymes such as protease, amylase, and lipase were found to increase significantly ($P<$ 0.05) in fish fed with 2-10g kg^{-1} *Citrus sinensis* compared to the control **(Shabana *et al*., 2019)**.

CONCLUSION

Adding Ambon banana peel flour in Nile tilapia feed by 2% kg^{-1} feed improved health, survival, and growth performance, which was supported by an increase in the number of lactic acid bacteria and digestive enzyme activity. In contrast, feed utilization was not different from the control, with a rearing duration of 28 days. Based on this, Ambon banana peel flour has the potential as a prebiotic for Nile tilapia.

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