Combination of Enzymes Papain and Phytase on Commercial Feed toward Growth Rate, Survival Rate, Feed Conversion Ratio and Feed Efficiency of Tilapia (Oreochromisniloticus)

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Abstract: ***Tilapia fish (Oreochromis niloticus) was one of freshwater fishery product that people are interested in. Higher price in good quality was problem in the cultivation. Alternatives such as adding papain and phytase enzyme had been introduced in solving this problem. This study aimed to establish effect and best combination of papain and phytase dose toward growth rate, survival rate, feed conversion ratio (FCR) and feed efficiency of tilapia fish. This study used entirely randomized form with four treatments and five reproductions. The treatment used were P0 (control), P1 (papain 5% and phytase 0.15%), P2 (papain 3% and 0.1% phytase) and P3 (papain 5% and phytase 0.15%). The data were analyzed using ANOVA and Duncan's multiple range test. The result showed combination of papain and phytase enzymes in commercial feed affected growth rate (weight), FCR and feed efficiency but no any influence in length and survival rate. The dose of papain 3% and 0.1% of phytase enzymes (P2) was optimum dose to increase growth rate, feed efficiency and decreased FCR.***

I. INTRODUCTION

Tilapia fish (*Oreochromis niloticus*) is one of freshwater fishery products that people are interested in. Tilapia production from 2010 to 2014 has increased significantly with an average increase of 19.03%. Aquaculture business consists of several specific aspects that are very important to note for the cultivated commodities to achieve optimal and maximum results, one of the aspects is the selection of good quality feed. Feed is one of several factors in the cultivation of fish that requires high cost, reaching 60-70% of the total production cost [1]. The effort taken to overcome the high price of feed is the use of some vegetable materials. Unfortunately, vegetable materials have crude fiber which is hard to be digested and hard cell walls that are difficult to break [2]. Another concern on the vegetable material is that there is an anti-nutrient substance called phytic acid. An effort that can be done to overcome this concern is by adding exogenous enzymes such as papain and phytase. Papain is an enzyme of papaya fruit which is a protease enzyme. Protease is an enzyme that is proteolytic and capable to hydrolyze complex compounds of protein's into simpler element (amino acids) so that it can be easily digested optimally by the body of Nile Tilapia [3]. Phytase enzymes is one of exogenous enzymes that is expected to elaborate anti-nutritional substances particularly phytic acid so that it will increase the performance of the feed use and fish growth [1].

Based on the above portrayal it is probable that these two enzymes combination can be complementary. Papain enzyme has the capacity to break down proteins into quiet components so that the feed proteins absorption rate increases while the phytase enzymes has ability to hydrolyze phytic acid contained in the feed material into inositol compounds and phosphoric acid with organic phosphor. The purpose of this study are to study aimed to increase growth rate, survival rate, feed efficiency and decrease feed conversion ratio of tilapia (*Oreochromis niloticus*) and best doses of grouping of papain enzymes and phytase in commercial feed.

II. METHODOLOGY

The study was conducted at the Wet Laboratory of the Faculty of Fisheries and Marine Airlangga University Surabaya, from February to August 2018. The equipment were 20 aquariums measuring 40 cm x 30 cm x 30 cm, aeration stone, strainer, ruler, spray bottle, spoon, paper labels, thermometers, pH, DO meters and analytical scales.

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Materials needed in this study were tilapia fish seed (*Oreochromis niloticus*) from BBI Wlingi, Blitar measuring 7-10 cm with body weight between 7-13 grams of 200 tails. The feed used was pellet-shaped commercial food under trademark "HI-PRO-VIT". Feed treatment added by papain enzyme with brand "NEWZIME" from BBPBAP Jepara and phytase enzyme from "SMIZYME" brand from China.

The media and experimental materials used were considered uniform, distinguishing only the administration of papain and phytase enzymes consisted of four treatments with different doses in each treatment. The experimental design was Completely Randomized Design (RAL). The treatments used were P0 (control), P1 (papain 1% and phytase 0.05%), P2 (papain 3% and 0.1% phytase), and P3 (papain 5% and phytase 0.15%).

The aquariums were cleaned used soap until clean and washed again with chlorine and then dried for 2-3 days. After the aquarium was cleaned and filled with clean water as high as 15 cm from the bottom of the aquarium and given a hose and aeration stones in each aquarium. Prior to the treatments, Tilapia seeds were acclimatized to the new water temperatures and conditions, as well as familiarized with the commercial feed used.

The commercial feed used was dried pellet. The papain and phytase enzymes were in form of powder. Enzymes were weighed as much as percentage of the feed weight accorded to each treatment. In additions, 1% tapioca from the feed weight that served as the glue was added. Enzymes and tapioca were put on a spray bottle and then mixed with aquadest as much as 10% of feed weight until evenly distributed. The commercial feed was placed in a container and sprayed by the enzyme that had been mixed with aquadest until evenly distributed. The feed that had been mixed with the enzyme was then dried for 1 hour. After dry the feed was given.

The data obtained from this study results were analyzed using ANOVA statistical test to determine whether there were differences between treatments. The design used was Completely Randomized Design (RAL).

III. RESULT AND DISCUSSION

The average specific growth rate (weight), specific growth rate (length), survival rate, feed conversion ratio and feed efficiency of tilapia could be seen in Table 1.

Table. 1 Average GR (Weight), GR (Length), SR, FCR and FE Nile Fish

Treatment		P0	P1	P2	P3
Growth Rate (Weight)	Average (%) ± SD	$1,66^{a}$±0,43	$1,69^{a}$±0,359	$2,29^{b}$±0,11	$1,92^{ab}$±0,21
	Transforms (√y+0,5) ±SD	1.46±0,15	1.47±0,12	1.67±0,03	1.55±0,07
Growth Rate (Length)	Average(%) ± SD	0,49±0,17	0,52±0,18	0,61±0,14	0,59±0,65
	Transforms (√y+0,5) ±SD	0,99±0,09	1,01±0,08	1,05±0,07	1,05±0,03
SR	Average(%) ± SD	72±4,47	72±4,47	82±10,95	74±5.48
	Transforms (√y+0,5) ±SD	8.51±0,26	8.51±0,26	9.07±0,59	8.63±0,31
FCR	Average(%) ± SD	$1,41^{a}$±0,050	$1,37^{ab}$±0,02	$1,20^{c}$±0,14	$1,29^{cb}$±0,05
	Transforms (√y+0,5) ±SD	1,38±0,02	1,37±0,01	1,31±0,05	1,34±0,02
FE	Average(%) ± SD	$69,55^{a}$±1,68	$72,25^{ab}$±1,28	$83,34^{c}$±9,53	76,94±2,89
	Transforms (√y+0,5) ±SD	8,37±0,10	8,53±0,01	9,14±0,52	8,80±0,17

The data about the water quality in this study can be seen in Table 2.

Table. 2 The water quality range as the maintenance of tilapia fish for 30 days.

Parameters	Range
Temperature (^{0}C)	26-28,9^{0}C
pH	7
Dissolved Oxygen (mg/l)	3,73-5,67 mg/l
Ammonia (mg/l)	0,01

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Discussion

The result showed best growth rate value was the P2 (papain 3% and 0.1% phytase). This result indicated that specific doses in given feed could be absorbed properly in the fish's digestive system. In the digestive system, papain enzyme with a dose of 3% was able to optimally hydrolyze the protein into a simpler compound, amino acids. Syahputra et al. (2016) found hydrolyze proteins into amino acids had increased absorption rate of feed [4]. The phytase enzyme with a dose of 0.1% also worked to decomposition phytic acid so that metabolic processes such as the breakdown of proteins and minerals complex in the body works well. The phytases was catalyzed the hydrolysis of phytate (myo-inositol hexakis phosphate) into myo-inositol and inorganic phosphate. The presence of two enzymes could help maximized nutrient absorption. Blood would bring amino acids to every cell of the fish's body.

The tilapia growth was closely related to the feed amount given during the magnification process. Papain enzyme worked actively on vegetable protein. Papain was able to hydrolyze proteins into peptides and amino acids. More proteins hydrolyzed into amino acids, the more amino acids could be absorbed by the fish body for growth [5]. The phytase enzyme in the feed was able to decrease and decompose phytic acid and break the bond between phytic acid, protein and mineral complex which affected the protein-breaking enzymes in decomposing protein to amino acid [1].

The survival rate was fish number that still survived during maintenance. The result showed combination of 30% papain and 0.1% phytase was best optimum dose to increase growth rate, feed efficiency and decreased FCR. High fish death was occurred in early week of maintenance due to stress. The stress was due adaption phase for new environment and low DO level. The fish survival was highly depended on stocking density and optimal water quality to support their growth [6].

The fish survival rate was affected by several factors such as feed protein. The lack of protein contributed weight loss, weakness, muscle tissue shrinkage and edema [7]. The papain enzyme in the diet help in breaking protein into amino acids. Feed with optimal protein content could produce maximum growth. The phytase had increased mineral concentration such as magnesium, phosphorus, calcium, manganese and zinc in plasma, bone and whole body [8]. The feed conversion ratio also included by feed protein. The feed protein which fit nutritional requirement resulted more feeding efficiency [9].

In this study, high feed efficiency was observed in P2 which equal to 88.27%. High feed efficiency value was assumed that feed given to the fish could be utilized or absorbed by fish body well led to high efficiency level. Meanwhile, growth rate was very influential in getting good feed efficiency value. The papain and phytase enzymes was a treatment which affected on feed efficiency. The temperature ranges in this study was between 26°C and 28.9°C that suitable for tilapia environment. Optimum water temperature for best growth performance in juvenile tilapia was ranged between 28°C and 30°C.

The result of dissolved oxygen measurements (DO) was ranged from 3.7 mg/l to 5.67 mg/l. During the first week maintenance, average DO value in all aquariums was 3.7 mg / lcaused some fish to die but in the following week the value of DO had improved, ranging from 4 mg/l to 5.67 mg/l. The ammonia was 0.01 mg/l which ideal ammonia level for tilapia need below than 0.2 mg/l.

IV. CONCLUSION

In conclusions, combination of papain and phytase enzyme in commercial feed of tilapia fish influenced growth rate, feed conversion ratio and feed efficiency of tilapia. These combinations had no effect on tilapia life (*Oreochromis niloticus).* The best combination was 3% of papain and 0.1% of phytase enzymes (P2) was optimal dose to increase growth rate of tilapia (*Oreochromis niloticus).*

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