Growth Performance and Survival Rates of Milkfish (*Chanos chanos*) Fed Diets Supplemented with Pineapple Extract

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ABSTRACT

Milkfish (*Chanos chanos*) is one of commodity which is strategic to fulfill necessity of protein that cheap enough and delighted by customer in Indonesian. The low feed efficiency is one of the obstacles in milkfish intensive farming. This can be overcome by the addition of the exogenous enzyme that an important role to digestion of feed, among exogenous enzyme is bromelain enzyme contained in pineapple extract. Enzyme bromelain is one of enzyme protease, which has the function to dissolve the protein with hydrolyzing of peptide in acid amino. This experiment has purpose to know the influence giving pineapple extract on artificial feed to feed utilization efficiency, growth, and survival rate of milkfish (*C. chanos*). Fish samples used 42-day old *C. chanos* with an average weight of 3.54 ± 0.03 g. This research used completely randomized experimental and design methods (RAL) with 4 treatments and 3 replications, respectively. The used treatments were treatment A (0 ml), B (2.5 ml), C (5 ml), and D (7.5 ml). The results showed that the proportion of pineapple extract was very significant influence (P<0.01) to Feed Utilization Efficiency Ratio (PER) and Relative Growth Rate(RGR), but not significant influence (P>0.01) to Total Feed Consumed(TKP) and Survival Rate(SR). The optimum dose of pineapple extract is 4.50 ml can be Produce EPP maximum about 52.73%. The dose optimum pineapple extract about 4.26 ml can be produce per in maximum 1.55% and the optimum dose of pineapple extract 4.39 ml can produce RGR about 3.22%.

Keywords: Milkfish; Pineapple extract; Feed utilization; Growth

INTRODUCTION

Milkfish (*Chanos chanos*) is one of strategic commodity to fulfill the necessity of protein which is relatively cheap and delighted by Indonesian customer. Milkfish is also one of the export commodities in the form of bait milkfish and consumption. Some features and characteristics of this one marine fish are milkfish can live in brackish or fresh water; able to deal with very large changes in salinity (Eurihalin); species are successful and have been widely cultivated; migrate to the brackish area; can live at a higher density level; and resistant to disease [1].

Intensive milkfish cultivation is always constrained by a number of problems, one of them is the utilization of feed on milkfish that is not optimal, and feed becomes the biggest variable in feed costs. Feed on aquaculture activities is generally commercial feed issued for cultivation activities spending around 60-70% of the total production costs [2]. In addition, the growth of milkfish seeds which are slow also affects milkfish production because it takes a long time to harvest. Therefore, the feed provided must be efficient and effective so that it can be utilized optimally for growth. Efficiency can be done by adding proteolytic enzymes, one of them is the bormelin enzyme contained in pineapple [3].

The content of bromelin is very scrimpy in young pineapple, especially in the sticky parts. The middle part of the stem contains more bromelain than that of the edges [4]. The bromelin enzyme is one of the protease enzyme groups. Protease enzyme is an enzyme that functions to break down proteins by hydrolyzing peptide bond in amino acids [3]. The bromelain enzyme acts as an exogenous enzyme. The addition of bromelain enzymes helps to produce more amino acids so that the food consumed can be utilized more efficiently, because the feed given is more usable especially for the growth [5]. The addition of bromelain enzyme in pineapple extract in feed proved to be able to increase protein digestibility and growth in some fish species, including the results of the research [6], that the use of pineapple powder in feed amounted to 10,39% is the best dose to increase digestibility of protein and growth.

The addition of pineapple extract in artificial feed has been proven

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to improve the efficiency of protein utilization, growth, and survival in several fish species. Research on the addition of pineapple extract as a source of bromelain enzyme in catfish feed (Clarias gariepinus) was carried out on goldfish seeds(Cyprinus carpio) with the tested concentration is 0.75% - 2.25% [7,8]. The best results from both studies were at a concentration of 2.25%. The results of the study, that the use of pineapple powder in feed amounted to 10.39% is the best dose to increase protein digestibility and growth [6]. The results of [4], the best concentration of pineapple stem extract on parrotfish (Ananas testudineus) is 5% resulted in the highest digestibility of 86.7%, feed efficiency 40.222 ± 6.11, and specific growth rate of 0.974 ± 0.028%/day. Milkfish is chosen for research because all this time the use of pineapple extract is used for freshwater fish and there has been no research on the administration of pineapple extract for sea water and brackish fish, especially for milkfish. This has led to the importance of research on "The performance, feed utilization efficiency, growth, and Survival rate of Milkfish (C. chanos) through the Additions of Pineapple Extract on Artificial Feed".

MATERIALS AND METHODS

The procedure in this study consists of preliminary tests, preparation, and implementation stages. Preliminary tests were carried out to determine the dose of pineapple extract used for research. The dose of pineapple extract for preliminary tests refers [4] namely the levels of pineapple extract 0 ml, 2.5 ml, 5 ml and 7.5 ml for milkfish (*C. chanos*). Preliminary test was carried out for \pm 21 days (7 days of acclimatization and 14 days of maintenance) using *C. chanos* fish obtained from Bulusan Pond, Demak, commercial feed with 35% protein, and 12 containers of plastic buckets. The results of the preliminary test that provides the best response are at a dose of 5 ml/kg.

The preparation stage of the study includes preparation of animals, feed, containers, and test media. Preparation of test animals is done by selecting healthy fish and having a uniform size. Test animals were acclimatized for one week and giving commercial feed without the addition of pineapple extract. Test animals were selected on day 7 with weight measurements using digital scales. There were 20 test animals were inserted into each maintenance container. The test feed that will be used is from the trademark of Hi-Provite Feed Fish Floating Seed FF-999 protein 35%. Test feed used were added with pineapple extract by diluting every 1 kg with 1 liter of water for each treatment. Feeding during the study was carried out using the fix feeding rate method (5% of the weight of biomass) and the frequency of feed given twice a day [1]. The test container used was a 25 liter of a plastic bucket with 12 pieces. Maternity live media is 15.4 ppt (SNI, 1999) consisting of a mixture of fresh water and sea water (deposited for 24 hours). Make a medium with the desired salinity using the formula V_1 . $N_1 = V_2$. N_2 [9].

This research was carried out for 42 days. The test animals used in this study were milkfish seeds C. *chanos* with an average weight were 3.54 ± 0.03 grams/head, 1 head/L. The dose of pineapple extract used is a modification of the preliminary test, where the best preliminary test results are 5ml/kg. The amount of feed given is 5% of the weight of biomass with the frequency of feeding 2 times a day at 08.00 and 17.00. Water quality measurements such as temperature, pH, and salinity are carried out every day, DO measurements are carried out once a week and for ammonia (NH₃) is measured at the beginning, middle and end of the study. The experimental design used in this study was a Completely Randomized Design (CRD) with 4 treatments and 3 repetitions. The treatment of this research is:

Treatment A: feed added pineapple extract dose 0 ml/kg

Treatment B: feed added pineapple extract dose 2.5 ml/kg

Treatment C: feed added pineapple extract dose of 5 ml/kg

Treatment D: feed added pineapple extract dose of 7.5 ml/kg

Data collection

Data collection conducted in this study included data on Total Feed Consumption (TKP), Feed Utilization Efficiency (EPP), Relative Growth Rate (RGR), Survival Rate (SR) and water quality.

- TKP = F₁ F₂, where TKP: Total feed consumption, F₁: Stock of feed provided during the study (g), F₂: Stock of feed left over from use during the study (g).
- EPP = (W_t W₀/F) × 100%, where EPP: Efficiency of Feed Utilization (%), W_t: Total weight of test fish at the end of the study (g) W₀: Total weight of test fish at the beginning of the study (g), F: The amount of food consumed during the study (g).
- PER = (W_t W₀/P_i) × 100%, where PER: Protein Efficiency Ratio (%), W_t: Total weight of test fish at the end of the study (g) W₀: Total weight of test fish at the beginning of the study (g), P_i: Weight of feed consumed × % of feed protein.
- RGR = (W_t W₀/W₀ xt) × 100%, where RGR: Relative growth rate (%), W_t: Total weight of test fish at the end of the study (g) W₀: Total weight of test fish at the beginning of the study (g), t: maintenance time (days).
- SR = $(N_{1}/N_{0}) \times 100\%$, where SR: fish survival rate (%), N_t: Number of fishes at the end of the study (tail), N₀: Number of fishes at the beginning of the study (tail).

Data analysis

The data in the form of percentage includes Total Feed Consumption (TKP), Efficiency of Feed Utilization (EPP), Protein Efficiency Ratio (PER), Feed Conversion Ratio (FCR), Relative Growth Rate (RGR) and Survival (SR) which were obtained then analyzed using analysis of variance (ANOVA) at 95% and 99% of confidence levels to see the effect of treatment. If in the analysis of variance was found to have a significant effect (P<0.05) or very significant effect (P<0.01), then the double Duncan's area was tested to determine the differences between treatments. Water quality data were analyzed descriptively and compared with the value of water quality feasibility to support the growth of fish [10]. Estimating the optimal dose of pineapple extract in the feed was carried out by orthogonal polynomial analysis with Maple software 2016.

RESULTS

Based on the weekly growth pattern of *C. chanos* for each treatment during the study, a graph can be made which can be seen in Figure 1.

Based on Figure 1 it can be seen that the pattern of weight gain of milkfish (C. *chanos*) increases every week exponentially. Treatment

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C gave the highest weight gain pattern, which was 95.97 g, then followed by treatment B at 84.27 g, treatment D was 80.47 g and treatment A was 66.40 g.

Based on the study of the effect of pineapple extract on artificial feed on the utilization of *C. chanos* feed, covering the values of TKP, EPP, PER, RGR and SR were presented in Table 1.

Based on the table above, it can be seen that the total average value of feed consumption (TKP) in each treatment from the highest to the lowest is treatment C which is 181.73 g, treatment B is 177.07 g, treatment D amounted 176, 83 g and treatment A is 167.20 g. The average value of Feed Utilization Efficiency (EPP) in each treatment from the highest to the lowest was treatment C at 52.73%, treatment B was 47.54%, treatment D was 45.50% and treatment A was 39.63%. The average value of Protein Efficiency Ratio (PER) in each treatment from the highest to the lowest was treatment C at 1.54%, treatment B at 1.48%, treatment D at 1.33% and treatment A at 1, 17%. The average Relative Growth Rate (RGR) in each treatment from the highest to the lowest was treatment C of 3.33%/day, treatment B of 2.93%/day, treatment D of 2.67%/day and treatment A of 2.20%/day. The average value of Survival Rate (SR) in each treatment from the highest to the lowest was treatment C at 86.67%, treatment B at 78.33%, treatment D at 76.67% and treatment A was 73.33%.

The results of orthogonal polynomial graph of feed utilization efficiency and protein efficiency ratio in *C. chanos* during the study are presented in Figure 2.

Based on the above Figures 2a and 2b above it can be seen that the optimum dose of pineapple fruit extract is 4.50% capable

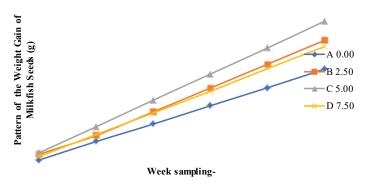


Figure 1: Weekly pattern of the weight gain of Milkfish (*Chanos chanos*) for each treatment during the study.

Table 1: The values of TKP, EPP, PER, RGR, and SR of Milkfish (C. *chanos*) during the study.

Treatment	Observed Variables						
	TKP (g)	EPP (%)	PER (%)	RGR (%/ hari)	SR (%)		
А	167,20 ±	39.63 ±	1.17 ±	2,20 ±	73,33 ±		
	9,36ª	1,32 ^b	0,04 ^b	0,10 ^b	5,77ª		
В	177,07 ±	47,54 ±	1.48 ±	2,93 ±	78,33 ±		
	7,61ª	1,88 ^b	0,11 ^b	0,06 ^b	5,77ª		
С	181,73 ± 7,66ª	52.73 ± 1,47 ^a	1.54 ± 0,04ª	3,33 ± 0,15ª	86,67 ± 2,89ª		
D	176,83 ±	45.50 ±	1.33 ±	2,67 ±	76,67 ±		
	4,81ª	1,97 ^b	0,06 ^b	0,15 ^b	7,64ª		

Information: Average values with different superscript show significant differences (P>0.01).

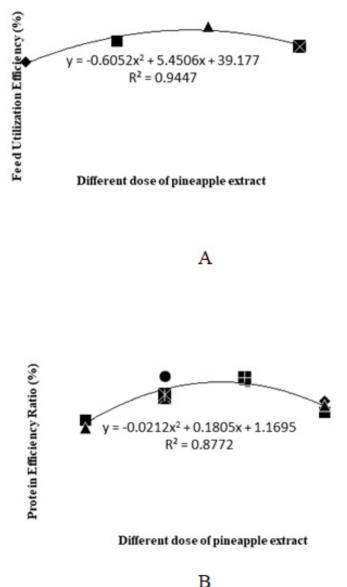


Figure 2: Orthogonal Polynomial Graph (A) Feed utilization efficiency and (B) Protein Efficiency Ratio (PER) in *C. chanos* during the study.

for maximum EPP production at 52.73%, the optimum dose of pineapple fruit extract is 4.26% is able to produce a maximum PER at 1.55% and the optimum dose Pineapple fruit extract of 4.39% can produce a maximum RGR at 3.22%/day. The results of the water quality measurement in the maintenance media which can be seen in Table 2 shows that the water quality in the maintenance medium is still in the library range, so it is still safe for the medium of milkfish rearing (*C. chanos*).

DISCUSSION

Feed consumption level

The average yield of Total Feed Consumption (TKP) in each treatment found the highest value in treatment C which was 181.73 g, and the lowest value was in treatment A at 167.2 g. After analyzing the variety of total feed consumption of C. *chanos* showed that the addition of pineapple extract in milkfish feed did not affect significantly toward the TKP value (P>0.01). The highest total value of feed consumption in treatment C (addition of 5ml/kg extract) this is thought that the protein energy produced was suitable for

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Treatment	Estimated Water Quality Parameter						
	Temperature (°C)	pH	DO (mg/l)	$NH_3(mg/l)$	Salinity (ppt)		
А	27-31	7-7,5	4,45-5,67	0,003-0,006	14		
В	27-31	7-7,8	4,53-5,59	0,003-0,006	15		
С	27-31	7-7,8	4,60-5,38	0,003-0,006	15		
D	27-31	07-Aug	4,55-5,46	0,003-0,005	15		
References	28-32*	7-8,5*	>5*/>3**	<1***	5-35*		

 Table 2: Results of water quality measurement during the study.

the needs of fish compared to the other treatments that affect the amount of feed consumed by fish. The total feed consumption is more influenced by the balance of protein energy in feed rather than the palatability of feed. Energy that exceeds the need would reduce the level of feed consumption so that the intake of other nutrients will also decrease [11].

The content of nutrients contained in the feed greatly affects the level of feed consumption of milkfish. The energy content of feed can determine the amount of consumption. Protein requirements in fish are influenced by the level of feeding and energy content. Digestible feed can increase the amount of food consumed by fish [12]. The low feed intake by fish can be affected by the presence of ingredients that are difficult to digest, the presence of anti-nutrition substances and the presence of materials that are difficult to eat by fish. In addition, the nutrient content that is almost similar in the feed is thought to be able to spur the response of fish on the feed given [13].

The results of TKP in this study have no significant effect [6], the highest total feed consumption in tilapia (*O. niloticus*) was 175.2 g with pineapple extract in feed as much as 15% and the lowest in 0% control was 135.8 g, and [14] the highest total feed consumption in java barb fish (*P. javanicus*) was 67.60 g with pineapple extract in feed was 0.75% and the lowest in 0% control amounted to 65.74 g.

Feed utilization efficiency

The highest EPP results in this study were found in treatment C (5ml/kg) which was 52.73% and the lowest value was in treatment A (0%) which was 39.63%. The results of treatment C have EPP values above 50% and for treatment A, B, & D are expressed deficient because the EPP value is below 50% this is presumably because the dose given to C treatment is the right dose for milkfish (C. *chanos*). The EPP greater than 50% is considered "good" growth [15].

The results of the analysis of various EPP values in C. *chanos* showed that the addition of pineapple extract on artificial feed with different doses gave a very significant effect (P<0.01) on the value of milkfish EPP C. *chanos*. Protease enzymes are responsible for breaking proteins. The breakdown of proteins into smaller molecules aims to facilitate digestion and will increase the EPP value, which will directly affect growth. The enzymes in artificial feed can increase nutrient absorption and adjust the excretion of nutrients, such as phosphorus, nitrogen, and minerals, and hydrolysis of phytic acid into inositol and phosphoric acid. Hydrolysis can destroy minerals from the complex [16].

The highest feed utilization efficiency (52.73%) was shown by fish fed with 5 ml pineapple extract. These results are still lower compared to other studies that stated that, the use of 2.25% doses

on the addition of bromalin extract to feed showed the highest efficiency of catfish feed utilization (63.80%) [17]. However, compared with the study of [4] that, the addition of pineapple extract with a dose of 5% in climbing perch fish feed shows the highest (40.22%).

Protein efficiency ratio

Protein Efficiency Ratio (PER) is a value that shows the amount of fish weight produced by each unit of protein weight in feed assuming that all proteins are used for growth. The results showed that the PER value with the addition of pineapple extract obtained the highest value in treatment C of 1.55%, then treatment B of 1.44%, treatment D of 1.33%, and the lowest value in treatment A of 1, 17%. The treatment C with the addition of 5 ml/kg pineapple extract has the highest value among other treatments, it is suspected that the dose is the appropriate dose to decompose protein and nutrient feed so that it can be utilized by fish optimally. The greater amount of enzyme added to the feed promoted the more protein that is hydrolyzed to amino acids, and thus increases protein absorption and growth of fish feed. On the other hand, if the amount of enzyme passes through the optimal point, it can respond to negative effects and the power to inhibit the growth of fish [17].

Treatment C is the best treatment, whereas in treatment D the value of PER decreases. This shows that the higher the dose of pineapple extract given will have a negative impact. The level of protein in aquaculture feed is generally an average of 18-20% [15]. The treatment of D experienced a decrease in PER value because in treatment D the protein hydrolyzed by protease enzyme exceeded the needs of *C. chanos* so that the addition of enzyme had a negative impact on the value of PER. The protease enzymes are more effective working on foods that have lower protein values when compared to high protein foods [18].

Relative growth rate

The results of the analysis of various RGR values in milkfish (C. *chanos*) showed that the addition of pineapple extract in artificial feed with different doses gave a very significant effect (P < 0.01) on the value of RGR in C. *chanos*. Growth occurs because the energy used for activity (maintenance) has been fulfilled so that nutrients such as protein will be displayed by increasing fish weight. The utilization of feed protein correlates with the energy in the feed. Energy is obtained from protein catabolism when insufficient feed energy or excess feed protein. Feed is said to be effective when eaten food is displayed in growth such as repairing and building as much tissue as possible with the smallest amount being converted into energy. The use of protein in fish can be increased by using supplements such as bromalin enzymes [17].

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Based on the research that has been done, the highest RGR value is found in treatment C (5 ml/kg). This is due to precisely the dose of pineapple extract for milkfish (*C. chanos*) so as to get good growth. Growth in fish that get periodically restricted feed intake can correlate in utilizing feed which includes the catalytic process by digestive enzymes [19]. Protein in the living body things plays a role in growth. Factors that influence growth include external and internal factors. The external factors include physical, chemical, and aquatic biology, while internal factors include inheritance, disease resistance and ability to use food. The digestive organs play a role in the absorption function of nutrients [20].

The highest growth rate in milkfish is shown in feed given pineapple extract with a dose of 5 ml which is 3.33%. The highest use of pineapple extract in catfish feed was achieved with feed added 0.75% [7]. Study showed different results with the use of the same dose in goldfish producing the highest growth rate at a dose of 1.5%. This shows that giving pineapple extract to feed given to different types of fish produces different maximum growth [8].

Survival

The results of the analysis of variance showed that the addition of pineapple extract on artificial feed had no significant effect (P>0.01) on the survival of *C. chanos*. Life is not affected directly by the feed; death is suspected to be internal and external factors such as fish being stressed during sampling a week. The survival of fish is influenced by internal and external factors. Internal factors come from the fish itself. Fish experience stress due to inadequate treatment resulting in high mortality and food competition. External influencing factors include environmental conditions such as high ammonia and or due to conditions that are less supportive in maintenance [21].

The factor of adding pineapple extract to the feed is known to have no significant effect on the survival of C. *chanos*, but the value of survival in this study is quite high, namely 73.3% - 86%. The survival rate of java barb fish was not influenced by the concentration of pineapple extract [22]. Life is influenced by water quality, especially pH. The average value of java barb fish life is 82%. The pineapple extract did not have a significant effect on the survival of tilapia (O. *niloticus*) and the highest survival value in this study was 93.3% in 0% treatment [6].

The survival of milkfish is directly affected by water quality. Qualified water quality can make the growth and continuity of the fish good, good water quality in maintenance the survival of the fish becomes great [23]. Good water quality will affect fish survival rate and fish growth [24].

CONCLUSIONS

The conclusion that can be drawn from the research results are as follows:

- 1. The use of pineapple extract on milkfish feed has a significant effect on Protein Efficiency Ratio (PER), Feed Utilization Efficiency (EPP), and Relative Growth Rate (RGR) but does not have a significant effect on total feed consumption and Survival (SR).
- 2. The optimum dose of giving pineapple extract to Total Feed Consumption (PER) was 4.26 ml, Feed Utilization Efficiency (EPP) was 4.50 ml and the Relative Growth Rate (RGR) was 4.39 ml.

RECOMMENDATIONS

The suggestion that can be given in this research is that the addition of pineapple fruit extract with a concentration of 0-7,5 ml can be applied to different cultures or for larger size milkfish for achieving total feed consumption, efficient utilization of feed and optimal growth.

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