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Maggot Meal (*Hermetia illucens*) Substitution on Fish Meal to Growth Performance, and Nutrient Content of Milkfish (*Chanos chanos*)

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ABSTRACT

Maggot has a high protein content for increasing fish growth through artificial feed. This study aimed to find the best feed formulation for fish meal substitution with maggot meal on growth, feed utilization efficiency, and survival rate of milkfish (*Chanos chanos*). A completely randomized design was used with five treatments and three replications. The treatments which had been done were fish meal substitution with maggot meal as follows: A (0%), B (25%), C (50%), D (75%), and E (100%). The research parameters included total feed consumption (TFC), feed utilization efficiency (FUE), protein efficiency ratio (PER), relative growth rate (RGR), survival rate (SR), and water quality. The results showed that the fish meal substitution with maggot meal had a significant effect ($p<0.05$) on FUE, PER, RGR and had no significant effect ($p>0.05$) on TFC and SR. The best treatment of each treatment is in treatment C with a composition of 50% maggot meal substitution on fish meal which resulted in TFC value of 40.17±4.58, FUE of 27.51±0.77, PER of 0.83±0.03%, and RGR of 2.34±0.10. The highest nutrient content is in the same treatment, namely lysine 10.95% and linoleic fatty acid 8.06%.

1. Introduction

Maggot is one of the abundant wastes and widely available in the community, but cannot be utilized maximally. Maggot is a black soldier fly larvae (*H. illucens*) egg, which has not been utilized optimally. Maggot meal comes from maggot in the pre-pupa phase because in this phase, maggot has a high protein content. Maggot meal has advantages over other artificial feed ingredients, which is rich in protein and amino acid nutrients (Cummins *et al.* 2017). Maggot meal (*H. illucens*) has higher level nutrient contents such as 40-50% of protein content, 30% of fat, and amino acids such as cystine, histidine, tryptophan, and tyrosine than fish meal (Ajani *et al.* 2004). In addition, the price of maggot meal is relatively cheap and it is easily cultivated through the bioconversion process of organic ingredients, namely palm oil cake (Cummins *et al.* 2017). Therefore, maggot meal can be used to substitute fish meal as an ingredient in feed making formulation for milkfish.

Milkfish (*C. chanos*) is one of the leading local fish commodities with high economic value. Milkfish production needs which tend to increase every year is the main problem of milkfish production (Kurnia *et al.* 2017). The sale of milkfish (*C. chanos*) at the retail level is still relatively profitable at a price range of Rp. 19,000-30,000 /kg. Milkfish production in Indonesia amounted to 212,883 tons in 2006 then increased to 421,757 tons in 2010. The total production of milkfish in 2017 reached 631,125 tons or 14.74% of the total aquaculture fish production. The increase in milkfish production from 2011 to 2017 reached an average of 20.84%. The increase of milkfish production is directly proportional to the increasing price of milkfish. This is caused by

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increased production costs, especially for the supply of feed and the high use of expensive fish meal as an artificial feed ingredient. Muliantara (2012) explained that the price of fish meal was relatively expensive and Indonesia relied on imports of fish meal as much as 70,000 tons worth Rp 1.17 trillion or equivalent to Rp 17,000 /kg, while the use of local fish meal was only 30,000 tons.

Production costs in fish farming activities around 60-70% are used to purchase feed. The high cost of fulfilling feed needs in industrial activities of fish farming is directly proportional to production cost (Haryati 2011). The high price of feed is due to fish meal as the main ingredient for feed still relies on imports (Priyadi *et al.* 2009). The effort that can be done to reduce the cost of purchasing feed is to replace fish meal with alternative ingredients. One of alternative ingredients for fish meal substitution is maggot meal. Research on the utilization of maggot meal as a substitute for fish meal has been carried out on several types of freshwater fish such as catfish, pomfret (Kardana *et al.* 2012), bala shark fish (Priyadi *et al.* 2009), and milkfish which are only limited to nutritional retention (Haryati 2011). Based on the research that has been done, the purpose of this study was to find the effect and composition of the best feed formulations from the utilization of maggot meal as a substitute for fish meal on growth, efficiency of feed utilization and survival rate in milkfish (*C. chanos*). The results of the study can provide information to fish farmers in choosing the use of feed technology as an alternative that is able to meet the nutritional needs of milkfish (*C. chanos*) to encourage its growth and efficiency of utilization. Based on this, it is necessary to do research on the effect of fish meal substitution with maggot meal in feed for growth performance and nutrient content of milkfish. This research was conducted in March-August 2018 at the Jepara Brackish Water Fisheries Center (BBPBAP) and Aquaculture Laboratory, Diponegoro University, Semarang.

2. Materials and Methods

2.1. Materials

The tested fish which were used in this study were milkfish with an average weight of 0.62±0.01 g/fish. The tested fish which were used amounted to 300 fish and were obtained from the Jepara Brackish Water Fisheries Center (BBPBAP). The reference for determining the tested animals in this study is based on the research conducted by Haryati (2011) which stated that to determine protein requirements for the growth of milkfish seeds with weights ranged from 0.5 to 0.8 grams, they need about 30-40% protein content in their feed. Stocking density in bucket containers with a volume of 20 liters equipped with recirculation pumps is 20 or equal to 1 fish/liter according to FAO (2008). Before starts too keep the fish, the tested fish is acclimatized so that they are not stressed after moved to the new environment. Feed is given as much as 5% of fish biomass/day with the frequency of feeding three times a day at 07.00, 12.00, and 17.00 WIB (Western Indonesian Time) with fix feeding rate method.

2.1.1. Test Feed

The test feed used in this study is pellet-shaped artificial feed with a protein content of 33-34%, Haryati (2011) stated that protein requirement for the growth of milkfish seeds with weights ranging from 0.5-0.8 g is range from 30- 40% protein content in feed. Feed was made using different doses of fish meal and maggot meal, treatment A (0% of maggot meal), B (25% of maggot meal), C (50% of maggot meal), D (75% of maggot meal), and E (100% of maggot meal). The stages before making test feed were preparing all ingredients, analyzed proximate of ingredients, and calculating feed formulations to be used. After the results of the analysis of each ingredient are known, it is used for calculating the feed formulation. The composition and proximate analysis of feed ingredients are presented in Table 1.

The feed used in this study was feed with protein content of 35%. Nutritional content of proximate analysis results was used to calculate feed formulations. The composition of the feed used in the study is presented in Table 2.

The amino acid content of milkfish feed (*C. chanos*) in the study is presented in Table 3.

The fatty acid content of milkfish (*C. chanos*) feed in the study is presented in Table 4.

2.2. Methods

The method of this research is the experimental method using Completely Randomized Design (CRD) with 5 treatments and 3 replications. The treatments are as follows:

Treatment A: test feed with 100% fish meal and 0% maggot meal substitution
Treatment B: test feed with 75% fish meal and 25% maggot meal substitution
Treatment C: test feed with 50% fish meal and 50% maggot meal substitution
Treatment D: test feed with 25% fish meal and 75% maggot meal substitution
Treatment E: test feed with 0% fish meal and 100% maggot meal substitution

The treatment of the research conducted refers to the research of Haryati (2011) about the substitution of fish meal with maggot meal on nutritional retention, body chemical composition, fat and energy retention, and milkfish feed efficiency ratio.

2.2.1. Data Collection

Data variables observed in this study included total feed consumption (TFC), relative growth rate (RGR), feed utilization efficiency (FUE), protein

Table 1. Proximate analysis of feed ingredients (in % of dry weight)

Ingredients	Protein	Nitrogen-free extract	Fat	Crude Fiber	Ash
Fish meal	58.01±0.15	3.29±0.10	5.53±0.02	3.15±0.80	30.12±0.12
Maggot meal	44.89±0.32	9.87±0.21	14.67±0.23	9.82±0.50	20.75±0.25
Soybean meal	50.53±0.20	37.36±0.17	0.94±0.25	3.72±0.35	5.28±0.22
Corn gluten meal	9.67±0.22	84.41±0.12	0.34±0.24	3.99±0.29	1.59±0.28
Bran meal	11.29±0.13	49.36±0.03	8.03±0.18	21.08±0.32	10.24±0.16
Wheat flour	10.57±0.24	86.45±0.09	1.06±0.12	1.20±0.13	0.72±0.17

Table 2. Composition and proximate analysis of feed used during experiment (*)

Ingredients	Test feed (g/100 g of feed)				
	A (0%)	B (25%)	C (50%)	D (75%)	E (100%)
Fish meal	28.00	21.00	14.00	7.00	0.00
Maggot meal	0.00	9.05	18.09	27.14	36.18
Soybean meal	25.00	25.95	25.90	26.85	26.81
Corn gluten meal	2.00	2.00	2.00	2.00	2.00
Bran meal	17.00	18.00	19.00	18.00	17.00
Wheat flour	21.00	17.00	14.00	12.00	11.00
Fish oil	3.00	3.00	3.00	3.00	3.00
Corn oil	2.00	2.00	2.00	2.00	2.00
Vit-min mix	1.00	1.00	1.00	1.00	1.00
CMC	1.00	1.00	1.00	1.00	1.00
Total (%)	100.00	100.00	100.00	100.00	100.00
Proximate analysis results					
Protein (%)	33.16	33.33	33.10	33.25	33.09
Nitrogen-free extract (%)	38.49	36.55	35.10	33.90	33.18
Fat (%)	8.37	9.36	10.34	11.19	12.04
Energy (kcal)	280.14	283.85	287.42	291.81	296.05
E/P ratio**	8.44	8.51	8.68	8.77	8.96

**E/P value for optimal fish growth ranged between 8-9 kcal/g (Watanabe 1988)

Table 3. Amino acid content of milkfish feed (*C. chanos*) during experiment

Essential amino acids	Requirements*	A (0%)	B (25%)	C (50%)	D (75%)	E (100%)
Arginine	5.2	5.36±0.03	5.64±0.15	6.08±0.15	5.72±0.12	5.60±0.05
Histidine	2.0	2.40±0.06	2.42±0.13	2.85±0.17	2.45±0.22	2.47±0.09
Isoleucine	4.0	4.07±0.09	3.93±0.07	4.95±0.11	3.65±0.28	3.51±0.08
Lysine	4.0	5.70±0.02	5.47±0.08	5.84±0.21	5.01±0.08	4.77±0.12
Methionine	3.2	2.38±0.10	4.25±0.26	4.72±0.35	3.99±0.06	3.86±0.08
Phenylalanine	5.2	3.99±0.17	4.90±0.25	6.51±0.13	5.72±0.05	5.63±0.05
Threonine	4.5	3.40±0.20	4.28±0.23	5.97±0.25	4.06±0.13	4.94±0.23
Tryptophan	0.6	0.98±0.23	1.96±0.09	2.95±0.23	1.93±0.21	1.91±0.05
Valine	3.6	3.63±0.25	3.98±0.10	4.89±0.22	4.12±0.25	4.95±0.10
Leucine	5.1	4.31±0.26	5.16±0.30	7.01±0.20	5.75±0.21	5.33±0.23

Table 4. The fatty acid content of milkfish (*C. chanos*)

Saturated fatty acids	Sample					
	Require ments*	A (0%)	B (25%)	C (50%)	D (75%)	E (100%)
Methyl butyrate	<0.1	0.88±0.06	1.35 ± 0.05	2.66 ± 0.02	0.88 ± 0.07	0.58 ± 0.09
Methyl hexanoate	<0.1	0.59±0.07	1.45 ± 0.04	2.02 ± 0.07	1.89 ± 0.06	1.09 ± 0.04
Methyl undecanoate	<0.1	0.09±0.03	1.25 ± 0.03	2.47 ± 0.03	3.09 ± 0.04	2.19 ± 0.05
Methyl laurate	0.23	0.73±0.01	1.90 ± 0.06	2.52 ± 0.04	1.83 ± 0.03	1.33 ± 0.02
Methyl tridecanoate	0.89	1.52±0.05	1.65 ± 0.07	3.58 ± 0.03	3.02 ± 0.04	2.82 ± 0.08
Methyl pentadecanoate	2.27	2.36±0.04	2.55 ± 0.08	3.99 ± 0.01	2.86 ± 0.05	2.17 ± 0.02
Methyl palmitate	0.73	2.35±0.03	2.93 ± 0.05	4.09 ± 0.03	3.85 ± 0.03	3.35 ± 0.01
Methyl heptadecanoate	0.97	1.18±0.07	1.80 ± 0.02	2.15 ±0.05	1.28 ± 0.05	1.23 ± 0.09
Methyl arachidate	4.75	4.73±0.06	4.95 ±0.05	5.65 ±0.02	4.37 ± 0.02	3.37 ± 0.03
Methyl tricosanoate	1.26	1.25±0.01	1.93 ± 0.08	2.59 ± 0.03	1.85 ± 0.06	1.05 ± 0.06
Unsaturated fatty acids						
Linoleic	<0.1	1.08±0.04	1.97±0.05	2.81±0.03	2.06±0.01	2.06±0.02
Linolenic	<0.1	1.54±0.03	2.08±0.04	2.76±0.02	1.74±0.05	1.74±0.05
Erucate	2.93	1.33±0.01	1.62±0.03	2.05±0.02	1.83±0.02	1.94±0.02
Eicosapentaenoic	0.93	1.18±0.06	2.07±0.02	2.67±0.05	1.98±0.04	1.65±0.04
Docosahexaenoic	<0.1	1.52±0.02	2.15±0.01	2.59±0.08	2.22±0.02	1.12±0.02

efficiency ratio (PER), survival rate (SR) and water quality parameters.

2.2.1.1. Total Feed Consumption (TFC)

According to Pereira *et al.* (2007), calculation of daily total feed consumption value is calculated using the following formula:

$$TFC = F1 + F2 ... + Fn$$

Description:
TFC : feed consumption (g)
F1 : the amount of feed on first day (g)
F2 : the amount of feed on second day (g)
F3 : the amount of feed on n day (g)

2.2.1.2. Relative Growth Rate (RGR)

Based on Yustianti *et al.* (2013), the relative growth rate (Relative Growth Rate) of fish is calculated using the formula:

$$RGR = \frac{W_t - W_0}{W_0 \times t} \times 100\%$$

Description:
RGR : relative growth rate (%/day)
W_t : biomass weight of tested fish at the end of the experiment (g)
W_o : biomass weight of tested fish at the beginning of the experiment (g)
t : length of experiment (day)

2.2.1.3. Feed Utilization Efficiency (FUE)

According to Watanabe (1988), the efficiency of feed utilization can be calculated using following formula:

$$FUE = \frac{W_t - W_0}{F} \times 100\%$$

Description:
FUE : feed Utilization Efficiency (%)
W_t : biomass weight of tested fish at the end of the experiment (g)
W_0 : biomass weight of tested fish at the beginning of the experiment (g)
F : the amount of fish feed given during the experiment (g)

2.2.1.4. Protein Efficiency Ratio (PER)

Protein efficiency ratio can be determined using the formula by Tacon (1987), as follows:

$$PER = \frac{W_t - W_0}{Pi} \times 100\%$$

Description:
PER : protein efficiency ratio (%)
W_0 : biomass weight of tested fish at the beginning of the experiment (g)
W_t : biomass weight of tested fish at the end of the experiment (g)
Pi : the amount of test feed consumed is multiplied by the protein content of the test feed

2.2.1.5. Survival Rate (SR)

Survival or survival rate (SR) is calculated to determine the mortality rate of test fish during the study, survival rate can be calculated using the formula Yustianti *et al.* (2013), as follows:

$$SR = \frac{N_t}{N_0} \times 100\%$$

Description:

SR : survival Rate (%)

N_0 : number of test fish at the beginning of the experiment (fish)

N_t : number of test fish at the end of the experiment (fish)

2.2.1.6. Water Quality Parameters

Observations of water quality include temperature, dissolved oxygen (DO), acidity level (pH) and ammonia (NH_3) content. Observation of water quality consists of ammonia (NH_3) content was carried out in the middle and end of the study. Observation of water temperature, dissolved oxygen (DO), and pH is carried out every day in the morning, afternoon, and evening. Measurement of ammonia content was analyzed at PT. Central Proteinaprima (Shrimp Feed Marketing Laboratory) Purworejo, temperature measurement was using a thermometer, while pH measurement was done using a pH meter by dipping the pH meter in the fishkeeping container and seeing the results on the pH meter. Measurement of dissolved oxygen (DO) used DO meter by dipping the tip of the indicator (probe) into the water then waiting until constant and recording its value.

2.2.2. Data Analysis

The data obtained were then analyzed using analysis of variance (ANOVA) the confidence interval used was 95%. Before ANOVA is carried out, the data will first be tested for normality, homogeneity test, and addivity test to find out that the data is normal, homogeneous and additive for further testing, namely analysis of variance. After analysis of variance is carried out, if found the treatment was significantly different ($p<0.05$), then the testing was carried out using the Duncan's multiple range test, to find out the difference in middle values between treatments.

Proximate analysis. The proximate chemical composition of the samples was determined using a standard procedure (AOAC 2005; Herawati *et al.* 2018). The crude protein content was calculated by multiplying the total nitrogen factor. The carbohydrate content was estimated by the difference.

Essential amino acid profile. The amino acid composition of the sample was determined using High Performance Liquid Chromatography (HPLC) (Shimadzu LC-6A) (AOAC 2005; Herawati *et al.* 2018).

Fatty acid profile. The fatty acid composition of the sample was determined using a gas cromatograph (Shimadzu) (AOAC 2005; Herawati *et al.* 2018).

3. Results

The results of the study were the value of feed consumption level, feed utilization efficiency (FUE), feed conversion ratio (FCR), protein efficiency ratio (PER), relative growth rate (RGR), and survival rate (SR) are presented in Table 5.

The highest TFC value was in 100% of maggot meal substitution (treatment E) that was 47.20±2.51 g and the lowest was in 50% of maggot meal substitution (treatment C) that was 40.17±4.58 g. The highest feed utilization efficiency (FUE) was 50% of maggot meal substitution (treatment C) of 27.51±0.77% and the lowest was in 0% of maggot meal substitution (treatment A) that was 22.53±0.34%. The highest protein efficiency ratio (PER) value was in 50% of maggot meal substitution (treatment C) that was 0.83±0.03% and the lowest was in 0% of maggot meal substitution (treatment A) that was 0.68±0.01%. The highest relative growth rate (RGR) value was in 50% of maggot meal substitution (treatment C)

Table 5. Value of feed consumption level, feed utilization efficiency, feed conversion ratio (FCR), protein efficiency ratio (PER), relative growth rate (RGR), and survival rate (SR) during experiment

Treatments	Observed variables				
	TFC (g)	FUE (%)	PER (%)	RGR (%)	SR (%)
A (0%)	43.50±5.01[a]	22.53±0.34[e]	0.68±0.01[e]	1.93±0.24[d]	80±5.00[a]
B (25%)	42.49±3.12[a]	26.60±0.72[b]	0.80±0.02[b]	2.21±0.16[ab]	80±5.00[a]
C (50%)	40.17±4.58[a]	27.51±0.77[a]	0.83±0.03[a]	2.34±0.10[c]	80±5.00[a]
D (75%)	41.29±1.26[a]	25.57±1.06[c]	0.77±0.03[d]	2.26±0.10[e]	80±5.00[a]
E (100%)	47.20±2.51[a]	24.96±0.84[d]	0.76±0.03[c]	2.16±0.03[a]	80±5.00[a]

of 2.34±0.10% and the lowest was in 0% of maggot meal substitution (treatment A) of 1.93±0.24%. The survival rate (SR) value in each treatment got the same value of 80.00±5.00.

3.1. Feed Utilization Efficiency (FUE)

Based on FUE data of milkfish (*C. chanos*) during the study, orthogonal polynomials can be made and it is presented in Figure 1.

Based on the Polynomial Orthogonal test, it was obtained a quadratic pattern relationship (y=-0.0014x^2 + 0.1549x + 22.923) and R^2=0.8448 with an optimum point of 55.3%. The dose of fish meal substitution with maggot meal obtained from this equation was 55.3% capable of producing FUE 27.21%. The value of R^2 indicates that 84.48% FUE is influenced by the substitution of fish meal with maggot meal in artificial feed and 15.52% is influenced by other factors.

3.2. Protein Efficiency Ratio (PER)

Based on PER data of milkfish (*C. chanos*) during the study, orthogonal polynomials can be made and it is presented in Figure 2.

Based on the Polynomial Orthogonal test obtained a quadratic pattern relationship (y=-4E-05x^2 + 0.0046x + 0.6909) and R^2=0.8289 with an optimum point of 57.5%. The dose of fish meal substitution with maggot meal obtained from this equation is 57.5% which can produce a 0.82% PER. The R^2 value indicates that 82.89% of PER is influenced by the substitution of fish meal with maggot meal in artificial feed and 17.11% is influenced by other factors.

3.3. Relative Growth Rate (RGR)

Based on milkfish RGR data (*C. chanos*) during the study, orthogonal polynomials can be made and it is presented in Figure 3.

Based on the Polynomial Orthogonal test obtained a quadratic pattern relationship (y=-0.0001x^2 + 0.0134x + 1.9366) and R^2 = 0.976 with an optimum point of 67%. The dose of fish meal substitution with maggot meal obtained from this equation is 67% capable of producing RGR of 2.38%. The value of R^2 indicates that 97.6% FUE is influenced by the substitution of fish meal with maggot meal in artificial feed and 0.4% is influenced by other factors. The amino acid content of milkfish after being fed using fish meal substituted by maggot meal is presented in Table 6.

The highest profiles was found in lysine, which was 10.95%. The fatty acid content of milkfish after being fed using fish meal substituted by maggot meal is presented in Table 7.

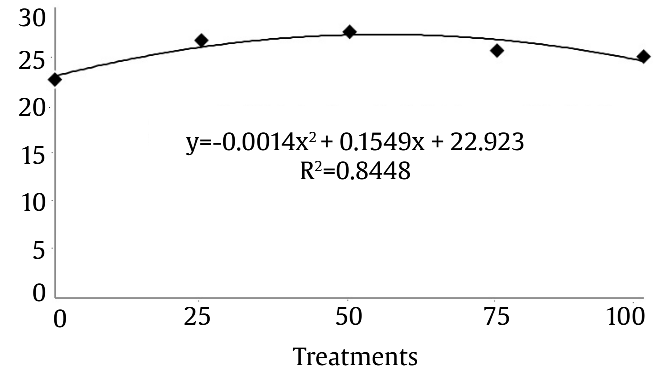

Figure 1. Polynomial orthogonal graph and curve analysis result of feed utilization efficiency (FUE) of milkfish (*C. Chanos*)

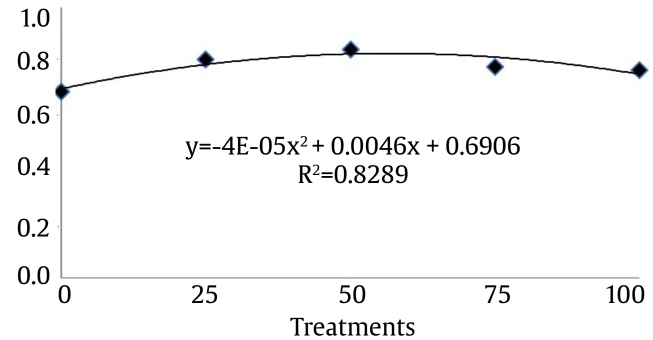

Figure 2. Polynomial orthogonal graph and curve analysis result of protein efficiency ratio (PER) of milkfish (*C. Chanos*)

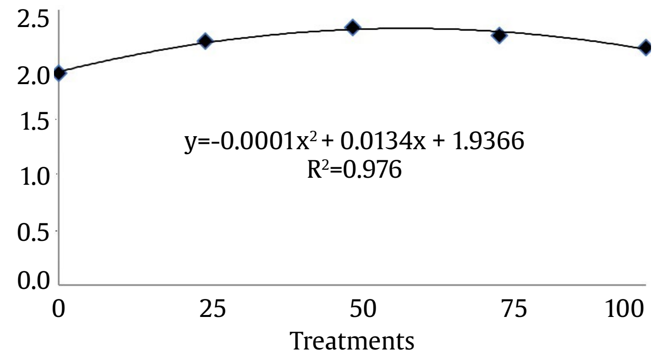

Figure 3. Polynomial orthogonal graph and curve analysis result of relative growth rate (RGR) of milkfish (*C. Chanos*)

The highest profile results of essential fatty acids were found in essential fatty acid linolenic fatty acid, which was 8.06% and non essential fatty acid palmitate fatty acid which 10.78%.

3.4. Water Quality Parameters

The results of the measurement of various parameters of water quality in milkfish keeping media (*C. chanos*) during experiment can be seen in Table 8.

Table 6. Amino acid content of milkfish after being fed using fish meal substituted by maggot meal (*)

Essential amino acids	0%	25%	50%	75%	100%
Arginine	6.07±0.03	6.17±0.04	6.98±0.07	5.75±0.03	5.78±0.02
Histidine	2.45±0.05	2.98±0.03	4.75±0.09	2.99±0.02	2.79±0.09
Isoleucine	4.52±0.04	4.05±0.02	6.23±0.02	4.17±0.05	4.63±0.11
Lysine	5.95±0.08	6.88±0.06	10.95±0.01	7.55±0.02	6.06±0.05
Methionine	2.76±0.02	4.80±0.09	6.70±0.07	4.78±0.01	3.97±0.02
Phenylalanine	4.09±0.01	5.05±0.06	7.19±0.02	5.95±0.04	5.82±0.07
Threonine	3.75±0.05	4.89±0.02	6.92±0.06	4.62±0.08	5.08±0.0
Tryptophan	2.76±0.09	2.15±0.01	3.90±0.02	2.85±0.02	2.06±0.04
Valine	3.96±0.08	4.18±0.03	5.77±0.01	4.76±0.02	5.06±0.02
Leucine	4.64±0.03	5.24±0.07	9.18±0.09	5.88±0.06	5.92±0.05

*Haryati (2011)

Table 7. The fatty acid content of milkfish after being fed using fish meal substituted by maggot meal

Saturated fatty acids	Sample				
	0%	25%	50%	75%	100%
Methyl butyrate	0.75±0.08	1.23±0.05	4.07±0.02	2.86±0.08	2.96±0.09
Methyl hexanoate	2.10±0.05	1.77±0.02	5.18±0.07	2.09±0.04	1.95±0.03
Methyl undecanoate	1.53±0.06	2.86±0.07	5.09±0.03	3.17±0.05	3.20±0.02
Methyl laurate	1.98±0.02	1.99±0.04	3.24±0.04	2.13±0.01	2.06±0.02
Methyl tridecanoate	3.75±0.04	2.98±0.03	6.23±0.03	3.97±0.07	3.97±0.06
Methyl pentadecanoate	3.78±0.01	4.05±0.01	5.90±0.01	4.06±0.04	3.99±0.08
Methyl palmitate	7.90±0.03	7.02±0.02	10.78±0.03	7.97±0.02	7.95±0.02
Methyl heptadecanoate	1.71±0.05	2.03±0.07	3.75±0.05	2.88±0.09	2.65±0.07
Methyl arachidate	4.88±0.06	5.75±0.02	7.79±0.02	5.96±0.05	5.85±0.07
Methyl tricosanoate	1.64±0.01	2.09±0.04	3.90±0.03	2.05±0.01	2.15±0.02
Unsaturated fatty acids					
Linoleic	2.06±0.03	4.05±0.05	8.06±0.04	5.26±0.02	4.54±0.02
Linolenic	2.66±0.04	3.68±0.04	7.12±0.06	3.88±0.05	4.04±0.05
Erucate	1.45±0.01	2.99±0.02	3.77±0.01	2.97±0.02	3.03±0.02
Eicosapentaenoic	1.28±0.05	2.97±0.01	4.89±0.01	2.09±0.04	2.19±0.04
Docosahexaenoic	1.12±0.07	1.55±0.06	3.79±0.07	1.72±0.02	1.66±0.02

Table 8. Results of measurement of various water quality parameters in milkfish (*C. chanos*) keeping media during experiment

Water quality parameters	Water quality range of experiment	Water quality parameters
Temperature (ºC)	28–32	25–32[b]
pH	7.64–7.90	6.0–8.5[a]
DO (mg/l)	3.00–5.30	≥3[c]
NH_3 (mg/l)	0.00–0.045	<1[a]

[a]Haryati 2011, [b]Reksono *et al.* 2012, [c]Mardihasbullah *et al.* 2013

4. Discussion

The growth of milkfish (*C. Chanos*) can be seen through the variables observed during the study including total feed consumption (TFC), feed utilization efficiency (FUE), protein efficiency ratio (PER), relative growth rate (RGR), and survival rate (SR). Based on the results of the research that has been done, fish meal substitution with maggot meal has been shown to have a significant effect ($p<0.05$) on FUE, PER, and RGR, but did not have significant effect ($p>0.05$) on TFC and SR. Observation of feed utilization in this study consisted of calculation of total feed consumption (TFC), feed utilization efficiency (FUE), and protein efficiency ratio (PER). Nutrient content in artificial feed with substitution of maggot meal is almost the same in each treatment.

The highest result of total feed consumption was at 100% of maggot meal substitution (A) that was 47.20±2.51 g and the lowest was in 50% of maggot meal substitution (C) that was 40.17±4.58 g. Based on analysis of variance, the results showed that maggot meal substitution on fish meal in artificial feed for milkfish (*C. chanos*) did not have a significant effect ($p>0.05$) on total feed consumption (TFC). This is because the test fish are in the same stage and the test feed has the same physical properties. This is strengthened by Kardana *et al.* (2012) that fish feed consumption is influenced by a number of factors including water quality, feed quality, feed management, genetic factors and physiological characteristic of fish. High water temperature can increase body's metabolic system so that feed consumption increases.

Based on TFC and FUE results, it was found that 50% of maggot meal substitution (C) produced the lowest TFC value of 40.17±4.58 g and resulted the highest FUE value of 27.51±0.77%. These results show that the feed is well digested by fish so that the feed is absorbed by the body of the fish effectively and efficiently. This is consistent with the statement stated by Kurnia *et al.* (2017) that given feed must really consider the quantity, because if the feed is given too little will result in low fish growth, however if too much it will cause inefficient metabolism so that it is not digested well. The feed composition of 50% substitution of maggot meal (C) showed the best FUE value compared to other treatments, which amounted to 27.51%. This value is better than the FUE value of milkfish in research conducted by Haryati (2011) regarding nutritional retention of 23.48%. The increase in the efficiency of feed utilization shows that the feed consumed has a good quality, so that it can be used efficiently. This is reinforced by (Kardana *et al.* 2012; Herawati *et al.* 2020) which stated that the high and low efficiency of feed is influenced by the source of nutrients and the amount of each component of the nutrient source of feed.

The highest FUE and PER results were in 50% of maggot meal substitution (C) were 27.51±0.77% and 0.83±0.03%, respectively. The protein efficiency ratio shows the percentage of protein weight in the feed given, which is used for growth. Based on the results of the research obtained, it shows that the quality of protein is not good because with FUE 27.51±0.77% only contributes a little protein to the body of the fish that is equal to 0.83±0.03%. Based on the results of the study, 50% of maggot meal substitution (C) was able to increase the protein and energy content in feed so that to be able to meet the minimum protein and energy requirements of milkfish feed for growth. Kurnia *et al.* (2017) in stated that feed formulation had an effect on milkfish growth, increasing protein content in feed formulations would increase the value of protein efficiency ratio in fish and growth rate in milkfish. The lowest FUE and PER values were in the 0% of maggot meal substitution (A), those were 22.53±0.34% and 0.68±0.01%, respectively. The low value is assumed that in 0% of maggot meal substitution (A) in feed, there is an imbalance in nutrient content in feed so that the value of FUE and PER and growth is not maximum. Kurnia *et al.* (2017) stated that vaname shrimp (*L. vanamei*) fed with only fish meal in feed formulations would produce less maximum growth compared to feed containing additional ingredients.

The highest relative growth rate (RGR) was in 50% of maggot meal substitution (C) that was 2.34±0.10%. Increasing of RGR value is found because protein in feed can be breakdown effectively and efficiently become simple component such as amino acids that are easily absorbed by the body for growth. This is reinforced by (Kardana *et al.* 2012; Herawati *et al.* 2020) stated that high feed efficiency shows efficient use of feed, so that only a few proteins are overhauled to meet energy needs and the rest is used for growth. The lowest PER and RGR values were found at 0% of maggot meal substitution (A) and the highest 50% substitution for maggot meal (C). The low PER and RGR values at 0% of maggot meal substitution (A) due to differences in the quality of low protein and amino acid content in maggot meal have not been balanced so that a less optimum value is obtained in fish growth. While the highest PER and RGR was in 50% of maggot meal substitution (C), amino acid content in the feed is able to meet amino acid requirements of milkfish to fulfill energy needs for maintaining environmental osmoregulation system and growth. According to Yustianti *et al.* (2013) that the amino acid methionine is one type of essential amino acid which plays a role for growth in the body of fish. This is also reinforced by Pramana *et al.* (2017) that lysine is an amino acid that has an important role in growth, which has a function to stimulate appetite, help convert fatty acids into energy in fish. The content of amino acids and minerals in feed plays an important role in spending energy during the osmoregulation process in maintaining ions. The PER and RGR values obtained increased values starting from the lowest was in 0% of maggot meal substitution (A) then increasing to 50% of maggot meal substitution (C) which was the best treatment, then decreased to 100% of maggot meal substitution (E). This shows that the substitution composition of maggot meal has a balanced point in the treatment of 50% dose of maggot meal substitution for the growth of milkfish. This is because maggot meal has an anti-nutrient in the form of chitin which if too much content will inhibit growth in fish. This is in accordance with the opinion of Stamer *et al.* (2014) that in his research on substitution of maggot meal in Rainbow trout fish, that in maggot meal there are anti-nutrients that can inhibit growth if too much use of maggot meal in feed.

Based on proximate analysis of protein energy ratio (E/P ratio) in each treatment ranged from 8.44 to 8.96 kcal/g protein. These results indicate that the ratio of protein energy in feed is within the limits of good susceptibility. According to NRC (1993) that the right E/P in fish feed is in the range of 8.0-12.3 kcal/g protein. Overall, the measurement variables FUE, PER, and RGR at 50% substitution of maggot meal(C) had different effects and showed the best growth among all treatments. This is presumably because the nutritional and energy content in feed is balance that suits the needs of the cultivan makes it grow well. Aslamyah and Karim (2012) stated that protein content and nutrient levels and energy in the feed must be balanced in order to support optimal fish growth. Feed with the optimum energy per protein ratio illustrates the balance between the amount of energy needed for basal metabolism and growth. Feeds that lack energy will cause most feed proteins to be used as energy sources for fish metabolism needs.

The FUE results showed that the substitution of maggot mealin each treatment gave results that were not significantly different from each treatment, so the results of the PER and SGR variables were also not significantly different from each treatment. The PER value ranged from 0.68-0.83% and RGR which ranged from 1.93 to 2.34% in this study was still relatively low. It is assumed that feed energy in each treatment substitution of maggot mealwhich ranges from 280.14-296.05 kcal is not fully used for growth, but is used to carry out body metabolism and maintain body ions and osmoregulation systems with the environment. The amount of energy that is much needed for the activity of maintaining the osmoregulation of milkfish on its environment, causes a reduction in energy for growth, so that it will cause a low growth rate. The high and low energy to maintain the osmosis system in milkfish is closely related to environmental salinity. The higher the environmental salinity, the higher the energy used to maintain the osmotic pressure of milkfish on the environment. According to Suharyanto and Tjaronge (2009) states that salinity greatly influences the osmotic pressure of water, the higher the salinity the higher the osmotic pressure of the environment. Milkfish balances the body's osmotic pressure with its living media through an osmoregulation mechanism. The higher the media salinity, the higher the workload of milkfish to balance the osmolarity pressure (media and blood) and balance the electrolyte content (media and blood), so that energy which is wasted for osmotic performance is higher.

The optimum dose of maggot mealsubstitution in fish meal was carried out using orthogonal Polynomial test with the results of 50% substitution of mggot flour (C) as the best treatment in the study. The orthogonal polynomial test results obtained FUE values of 55.3%, PER and RGR respectively with an optimum point of 55.3%, 57.5% and 67.0%. The results of the orthogonal FUE Polynomial test obtained a quadratic pattern relationship ($y=-0.0014x^2 + 0.1549x + 22,923$) and $R^2=0.8448$ with an optimum point of 55.3%. The FUE value of the equation is 55.3% capable of producing FUE 27.21%. The results of the polynomial orthogonal PER test obtained quadratic patterned relationships ($y=-4E\text{-}05x^2 + 0.0046x + 0.6909$) and $R^2=0.8289$ with an optimum point of 57.5%. The results of the orthogonal Polynomial test on the RGR obtained a quadratic pattern relationship ($y=-0.0001x^2 + 0.0134x + 1.9366$) and $R^2=0.976$ with a maximum point of 67%. Based on these results that with an optimum dose of 67% substitution of maggot meal can produce a minimum RGR of 2.38%.

The growth of milkfish (*C. chanos*) is relatively fast using substitution of maggot meal in fish meal. This is presumably due to the high protein and amino acid content in maggot meal compared to fish meal which is able to meet the minimum requirements of milkfish. The optimum dose for each variable obtained from the orthogonal polynomial test was able to meet the minimum amino acid requirements of milkfish and provide a significant influence in substituting fish meal with maggot meal. Haryati (2011) state that essential amino acids in feed that have not met the amino acid requirements of milkfish will be replaced with non-essential amino acids. The need for aromatic amino acids (phenylalanine and tyrosine) for the growth of milkfish is around 5.22% of the protein content in feed and the value of tyrosine replacement for phenylalanine is around 46%. This shows that the need for phenylalanine amino acids can be replaced by tyrosine. In addition, the need for methionine amino acid in feed can also be replaced by cystine. This is reinforced in research by Haryati (2011) that the total amino acid requirements of methionine and cystine for milkfish seed growth of 3.2% from feed protein content and cystine replacement value for methionine were about 50%. All feeds used were deficient in histidine and methionine amino acids, but methionine deficiency can be replaced by cystine non-essential amino acids. Based on the description shows that the feed used has relatively the same quality and in accordance with the needs of milkfish.

The results of the analysis of variance showed that the substitution of fish meal with maggot meal in artificial feed had no significant effect ($p>0.05$) on the survival of milkfish (*C. chanos*). It is suspected that the substitution of fish meal with maggot meal in artificial feed has an influence on growth, but does not have a significant effect on the level of survival rate. The level of survival rate of milkfish in the study was classified as good, the average survival rate of each treatment is 80%. Fish life is not directly affected by feed. This is thought to be fish that die because of lack of adaptation and stress during research. According to Kardana *et al.* (2012), factors that can influence the high and low of survival are internal and external factors. Internal factors come from the fish itself. Fish experience stress due to inadvertent treatment so that the mortality is high. External factors that influence include environmental conditions.

The highest essential amino acid in the similar treatment found in lysine (10.95%). The functions of lysin amino acid is to increase the energy in body, increase the stamina, and improve the damaged muscle tissues and help in the blood in an injury (Ovie and Eze 2013). Amino acid is an important component and cannot be apart from protein and plays a role in the production of carnitine and collagen. Meanwhile, the benefit of carnitine is to stimulate the growth by increasing collagen, support the production of other protein such as enzyme, antibody and hormone. In addition, other benefits of lysine are to enhance the calcium absorption and convert the fatty acid into energy. This statement is strengthened by the previous research stating that it can act as the frame of forming vitamin B1, antivirus, and help in calcium absorption, stimulate the appetite, help in carnitine production to convert the fatty acid into energy (Ovie and Eze 2013; Herawati *et al.* 2015).

The result of the analysis on the profile of the oleic fatty acid was found highest in the substitution of fish meal with maggot meal in artificial feed (treatment C, 50%) of essential linoleic fatty acid. Linoleic acid functions as the unsaturated fatty acid of omega-6, and it is required for the function of healthy brain, skin and bone density, energy production and health of reproduction (Benjamin and Olivia 2007). Palmitate fatty acid also called as hexadecanoic acid is the main saturated fatty acid functioned as the antioxidant and the source of vitamin A. Palmitic acid is a saturated fatty acid that serves as energy storage used for SAFA or fatty acid biosynthesis. Meyer (2004) and Benjamin and Olivia (2007) mention in their experiments that palmitic fatty acid is the substrate of SAFA fatty acid biosynthesis.

Water quality during research was in optimum condition for milkfish growth. The temperature of the container during fishkeeping ranged from 28-31°C. This temperature is quite suitable for milkfish condition. This is in accordance with the opinion of Haryati (2011) that the temperature range that is good for milkfish life is 24-31°C. Significant changes in temperature will cause stress in fish. (Haryati 2011; Herawati *et al.* 2020) stated that temperature is one source of stress that can affect the physiological changes in the fish's body. High water temperature can increase the body's metabolic system so that feed consumption increases. The dissolved oxygen measured during the study showed results of 3.00-5.30 mg/l. The results of the research on water quality parameters of dissolved oxygen, salinity, pH and ammonia are still in a good range for milkfish cultivation. This is in accordance with the opinion of Budiasti and Anggoro (2015) that in milkfish cultivation the availability of dissolved oxygen in a waters should not be less than 3 mg/l. In addition, milkfish can grow well at 15-35 mg/l salinity. The pH value obtained at the time of the study was 7.64-7.90, the results of these variables are still within the feasibility limit, Salam and Darmawati (2017) stated that milkfish live in pH conditions ranging from 7.0-8.5, because it is good for the growth and reproduction of organisms. Temperature and pH are the limiting factors that influence and determine the speed of metabolic reactions in consuming feed. Low pH values can cause clots of mucus in the gills and the fish will suffocate. The results of the ammonia measurements obtained were 0.00-0.045 mg/l, which were categorized as still within the normal range.

5. Conclusion

The conclusion that can be drawn from the results of the study is that the use of maggot meal as a substitute for fish meal in artificial feed has a significant effect ($p<0.05$) on relative growth rate (RGR), feed utilization efficiency (FUE) and protein efficiency ratio (PER), but it has no significant effect on total feed consumption (TFC) and survival rate (SR). The best dose of composition of fish meal substitution with maggot meal in artificial feed was in treatment C (50% of maggot meal substitution and 50% of fish meal) which gave a relative growth rate of 2.34±0.10%, protein efficiency ratio of 0.83± 0.03% and efficiency of feed utilization of 27.51±0.77%. The highest amino acid was lysine that was 10.95% and the highest fatty acid was linoleic that was 8.06%.

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