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DIETARY LYSINE REQUIREMENT OF JAVA BARB (*Puntius javanicus* BLEEKER, 1855) FINGERLINGS TO OPTIMIZE FEED EFFICIENCY, GROWTH, AND NUTRIENT CONTENTS

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ABSTRACT

The slow growth of the Java barb (*Puntius javanicus* Bleeker, 1855) fingerlings is due to the feeding of plant-based protein which is usually lack of lysine. The lack of it, can hinder the feed efficiency and growth. The best way to solve the problem is by adding lysine in the diet. The objectives of the study were to determine optimal needs of lysine to increase feed efficiency, growth, and nutrient contents of Java barb. The fish samples were fingerlings of Java barb which weighed from 3.26 to 3.78 g^{-1} fish. The feed comprised 30% isoprotein and 262 Kcal isoenergy supplemented with lysine amino acid. The dosages of the lysine in the diet were 1.25% (A1), 1.5% (A2), 1.75% (A3), 2.0% (A4), 2.25% (A5), and 2.5% (A6) of dried diets. The parameters observed were relative growth rate (RGR), apparent digestibility coefficients of protein (ADCp), efficiency of feed usage (EFU), feed conversion ratio (FCR), protein efficiency ratio (PER), survival rate (SR), hepatosomalic index (HSI), viscerosomatic index (VSI) and nutrient contents of Java barb. The results of the study disclosed that the enrichment of lysine into the diet increased the feed utilization, the growth and the nutrient contents of Java barb. The needs of lysine for Java Barb (*Puntius javanicus*) fingerlings to optimize feed efficiency, growth, and nutrition contents ranged from 1.58% to-1.70% dried diet (5.3 to 5.6 % of protein diet).

Keywords: *Puntius javanicus*, lysine, growth, efficiency, diet

INTRODUCTION

Java barb (*Puntius javanicus* Bleeker, 1855) is one of native fish species in Indonesia that is easily cultured. It also has a high economic value and potential that do not need rigid requirements to culture (Rachmawati *et al*., 2019). Java barb until recently has slowly growth, so it does not attract farmers to intensively cultivate the fish. The slow growth was thought because of lack of lysine amino acid in the diet that mostly comprised of plant-based protein. This is in line with the finding of Small and Soares, (2000) that plant-based protein in the feed has lysine deficiency. The complete essential amino acids have to present in the diet to support good growth (Li *et al*, 2009). The deficiency of amino acid decreases efficiency of feed utilization and growth (NRC, 2011). One of the efforts to solve the problem is by adding lysine into the diet.

Xie *et al*. (2012) described that the lysine in the diet was easily and fastly to be utilized in metabolism compared to other amino acids. Moreover, Fang Deng *et al*. (2010) suggested that the availability of lysine in the intestine would easily be used to synthesize protein to boost fish growth. Some other studies in the lysine incorporated diet on several species were reported by other researchers. Zhang *et al*. (2008) found that by inclusion 2.5% lysine in every kg diet can increase protein efficiency utilization in Large yellow croaker (*Pseudosciaena crocea*). Meanwhile, Zhou *et al.* (2010) disclosed that sea bream (*Sparus macrocephalus*) needs 3.3% lysine kg^{-1} diet. Luo *et al.* (2006) found that the optimal dosage of lysine in the diet for the *Epinephelus coioides* growth was 2.8%/ kg diet. Moreover, Ebeneezar *et al*. (2019) advised that the lysine supplement in the diet of 2,21%/kg diet resulted in the maximum WG, SGR and PER on the Silver pompano juvenile (*Trachinotus blochii*). Xie *et al*. (2012) recommended that the optimal dosage of lysine supplementation for SGR in the Pacific white shrimp juvenile (*Litopenaeus vannamei*) was 2.05%/kg diet. Fang Deng *et al*. (2010) recorded that the Pacific threadfin juvenile (*Polydactylus sexfilis*) given the feed with 2.23-2.43% lysine supplementation in every kg diet brought about the highest SGR, FEU, PER and protein retention. Those values were much higher than those given the lysine-incorporated diet with the lysine dosage of 1.69% kg^{-1} diet. Ahmed and Khan, (2004) expressed that the lysine dosage of 2.30 g /100 g diet given to Indian major carp fingerlings (*Cirrhinus mrigala)* lead to the optimal growth and FEU. Farhat and Khan (2013) found that the lysine needs for stinging catfish fingerlings (*Heteropneustes fossilis*) to get optimal growth, FCR and protein deposit were between 2.0% and 2.3% in the dried diet. The information in the optimal need of lysine for Java barb so far has not been reported. The study was to identify the optimal dosages of lysine supplementation in the diet to obtain the best of feed efficiency, the growth, and the nutrient contents in the Java barb fingerlings.

RESEARCH METHODS

Research Design

The study was done at the Center for Hatchery and Culture of Freshwater Fish, Muntilan, Jawa Tengah, Indonesia during March to April of 2020. The animal samples used in the study were Java barb fingerlings obtained from natural hatcheries in the center. Before the experiments were done, the fingerlings have been acclimated to the cultivating media and the feed for one week. During acclimation, the fingerings were stocked into the 500 L fiber container with dimension of 1.20 mx 0.85 mx 0.95 m and fed with the manufactured diet in satiation every morning and afternoon. Java barb fingerling samples were selected based on the size homogeneity, healthiness and free potential diseases (Rachmawati *et al.*, 2017). Then, the fish was let to fast for one day in order the fish to excrete the metabolic waste.

The experiments were started by weighing the fingerlings. Their average weights were 3.52±0.26 g^{-1} fish, then the fingerling were stocked with the density of 18 fingerlings per aquarium with the dimension of 80 cm x 60 cmx 75 cm. Each aquarium was equipped with the pass-through water flow system at the debit of 1 - 1.5 L per minute. The feeding was given at satiation method two times daily, in the morning at 08:00 and in the afternoon at 17:00. The weight gain was measured every week for 8 (eight) weeks.

Test Diet Preparation

The diet used in the study was the diet that comprised of 30% isoprotein and 262 Kcal isoenergy, based on the method of Rachmawati *et al*. (2019), and supplemented with various dosages of lysine, namely 1.25% (A1), 1.5% (A2), 1.75% (A3), 2.0% (A4), 2.25% (A5), 2.5% (A6) of g/100 g dried diet (Table 1). The lysine dosages used in the study were modified dosages of the Ahmed and Khan (2004) advised that the lysine supplement in the diet of 2.30 g/100 g dried diet the same to 5.75 g/100 g protein diet for optimal growth and efficient use of diet of fingerling Indian major carp (*Cirrhinus mrigala*). Essential amino acids were added to balance the composition of amino acids. Casein-gelatin Ratio (4:1) assisted to maintain the least possible provisiont for lysine and the maximum provision for other amino acids. Cristal lysine was weighed according the treatments and then diluted evenly with hot water (80°C). The mix was expected to have neutral pH with the NaOH 6 N solvent (Nose *et al.*, 1979). The gelatin was separately soften with the water and constantly heated and stirred, then it was mixed with the amino acid mix (Abidi and Khan, 2007). The next step was to mix all diet materials with lysine and gelatin. The diet mixture was then produced to become pellets with the 2 mm pellet machine. The wet pellets were dried to get water content of 10% in the oven at 45°C for 24 hours. The pellets were then stored in cold storage at 4 ° C.

Table 1. Ingredient Composition of Experimental Diets

Ingredients (g/100 g dry weight)	Diets					
	A1	A2	A3	A4	A5	A6
Casein[a]	16	16	16	16	16	16
Gelatin[b]	4	4	4	4	4	4
Amino acid mix[c]	21.35	22.16	23.24	23.24	23.24	23.24
Dextrin	31.84	31.78	30.45	30.20	29.95	29.70
Corn oil	2	2	2	2	2	2
Fish oil	4	4	4	4	4	4
Mineral mix[d]	4	4	4	4	4	4
Vitamin mix[e]	4	4	4	4	4	4
Carboxymethyl cellulose	11	11	11	11	11	11
α- cellulose	0.06	0.06	0.06	0.06	0.06	0.06
Lysine	1.25	1.5	1.75	2.0	2.25	2.5
Cr_2O_3	0.5	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100	100
Analyzed lysine	1.27	1.52	1.78	2.02	2.27	2.52
Proximate composition						
Crude protein (%)	30.21	30.62	31.57	31.13	31.48	30.49
Crude Lipid (%)	6.86	6.85	6.76	6.84	6.79	6.73
Ash	7.27	7.36	7.28	7.32	7.30	7.33
Gross energy (kcal/100 g, dry diet)	262.23	262.74	263.14	263.25	262.28	262.23

Information:
[a]Crude protein (76%)
[b]Crude protein (96%)
[c]Amino acid mixture (g 100 g-1) alanine 1.23; proline 0.952; valine 1.67; glycine variable; ; leucine 1.89; aspartic acid 0.09; methionine 1.03; glutamic acid 0.00; cystine 0.85; serine 0.00; phenylalanine 1.55; lysine variable; tyrosine 0.94; arginine 1.20; threonine 1.02; histidine 0.37; tryptophan 0.45; isoleucine 2.15.
[d]Mineral mixture (g 100 g-1) cuprous chloride 0.010; sodium chloride 4.35; calcium lactate 32.69;magnesium sulphate 13.20; zinc sulphated 7H2O 0.40; sodium biphosphate 8.72; potassium phosphate 23.98; magnus sulphated H2O 0.080; aluminum chloride 6H2O 0.0154; cobalt chloride 6H2O 0.10; ferric citrate 2.97; potassium iodide 0.015; calcium biphosphate 13.57 (Halver, 2002).
[e]Vitamin mixture calcium pantothenate 0.05; vitamin B12 0.00001; pyridoxine hydrochloride 0.005; menadione 0.004; ascorbic acid 0.10; niacin 0.075; thiamin

hydrochloride 0.005; riboflavin 0.02; folic acid 0.0015; vitamin E as α-tocopherol 0.04 biotin 0.0005; cellulose to 3 g 100 g-1; inositol 0.20; choline chloride 0.5 g 100 g-1 (Loba Chemic India)

Parameters Observation

The following formulas were used to calculate the observed parameters in the study of the Java barb (*P. javanicus*) in response to various dosages of lysine-enriched diet.

$$AWG\ (\%) = 100 \times (\text{final body weight} - \text{initial body weight}) \quad (1)$$

$$ADCp\ (\%) = 100 - \{100 \times \frac{\text{Cr2O3 in the feed x \% protein in the feces}}{\text{\% Cr2O3 in the feces x \% protein in the feed}}\} \quad (2)$$

$$EFU\ (\%) = 100 \ x\ \frac{(\text{Final weight} - \text{initial weight})}{\text{diet weight consumed}} \quad (3)$$

$$FCR = \frac{\text{feed fed (g, dry weight)}}{\text{body weight gain (g)}} \quad (4)$$

$$PER = \frac{\text{body weight gain (g)}}{\text{protein intake (g)}} \quad (5)$$

$$RGR\ (\%) = 100\ x\ \frac{(\text{Final weight} - \text{initial weight})}{(\text{times of experiment x initial weight})} \quad (6)$$

$$HIS\ (\%) = \frac{\text{liver weight (g)}}{\text{body weight (g)}}\ x\ 100\% \quad (7)$$

$$VSI\ (\%) = \frac{\text{viscera weight (g)}}{\text{body weight (g)}}\ x\ 100\% \quad (8)$$

$$SR\ (\%) = \frac{\text{end count (tails)}}{\text{starting count(tails)}}\ x100.\% \quad (9)$$

Chemical Analyses

Proximate analyses on the feed and fish body consisted of crude protein and fat, and ash analyses using AOAC method (2005). Crude lipid was calculated using Soxhlet extract method (FOSS Soxtec2043). Where crude protein (N × 6.25) was measured using Kjeldahl semi-automatic system method (FOSS Kjeltec 2300). The ash was obtained by burning the sampled fish in the burner at 550 ° C for 24 hours. Hydrolysis of 0.3 mg sample in the 1 ml HCL 6 N for 22 hours was done to analyze amino acids in the feed and initial and final carcasses of the Java barb fish. Then the samples were diluted with 0.02 N HCl and put into Automatic Analyzer for Amino Acid (Hitachi L-880). The analyses of amino acids of test diets were presented in the Table 2. The hepatosomalic index (HSI) and viscerosomatik index (VSI) calculations were obtained from 9 (nine) fish samples in each treatment (n = 3 x 3). Each fish of nine samples was measured the length, while its liver and viscera were weighed. The measurement of nutrient content in the final Java barb carcasses was computed using proximate analysis in 3 (three) fish samples from each repetition treatment.

Table 2. Amino Acid Contents of Test Diets (% Dried Matter)

Amino Acid Essential	A1	A2	A3	A4	A5	A6
Threonine	1.68	1.66	1.70	1.69	1.68	1.70
Lysine	1.27	1.52	1.78	2.02	2.27	2.52
Phenylalanine	1.87	1.86	1.87	1.85	1.86	1.86
Arginine	1.39	1.37	1.41	1.35	1.36	1.40
Methionine	1.07	1.07	1.09	1.09	1.08	1.07
Leucine	1.67	1.63	1.71	1.64	1.65	1.64
Isoleucine	0.76	0.77	0.78	0.76	0.75	0.76
Histidine	1.83	1.83	1.84	1.82	1.82	1.84
Valine	0.62	0.62	0.64	0.63	0.62	0.63

Protein digestibility analysis

The measurement of protein digestibility was indirectly done by adding Cr_2O_3 0.5% as digestibility indicator (Watanabe, 1995). The feed containing chromium was adapted for 7 (seven) days. Starting at the eighth day, the feed for the experiment was given until 58 days and the Java Barb feces were collected by siphoning every morning during that time. The collected feces were put in the bottle and stored in the freezer. After the feces collection was done, then the feces were dehydrated in the oven at 6° for 24 hours. Then, protein deposit and Cr_2O_3 were analyzed from the dried feces (NRC, 2011). The measurement of Cr_2O_3 was conducting using spectrophotometer with 350 nm wave length (Watanabe, 1995).

Statistical Analysis

The statistical analyses used in the study included analysis of variance (NOVA), Duncan double test and Polynomial Orthogonal test to determine optimal dosage for lysine amino acid in the diet (Stell *et al*., 1996).

RESULTS AND DISCUSSION

Results

The results on growth performance, feed utilization, and indexes of Java barb condition were presented in the Table 3.

Growth Performance and Feed Utilization

The ANOVA test of the lysine supplemented diet were highly significant ($P<0.01$) on ADCp, EFU, RGR, FCR, PER, except on SR of Java barb (Table 3). The relation between the supplementation of lysine and ADCp was calculated using the Polynomial Orthogonal test was a quadratic pattern as Y= -13,991x2 + 46,84x + 30,059 R^2 = 0,856 (Figure 1), the optimal dosage of lysine was 1.67 % dried diet. Meanwhile, EFU was also the relation was a quadratic pattern as Y= -13.129x2 + 42.808x + 32.706, R^2 = 0.853 (Figure 2), the optimal dosage of the lysine was 1.63 % dried diet. RGR also had a quadratic pattern as Y = -2.06x2 + 7.0005x – 2.0863, R^2 = 0.943(Figure 3), the optimal dosage of the lysine was 1.70 % dried diet. FCR resulted in the quadratic relation as Y = 0.68x2 -2.2023x + 3.8209 (Figure 4), R^2 = 0.685, the optimal dosage of the lysine was 1.62 % dried diet. PER was also a quadratic pattern as Y = -1.4286x2 + 4.5263x- 0.7985, R^2 = 0.917 (Figure 5), the optimal dosage of lysine was 1.58 % dried diet.

Table 3. Growth Performance, Feed Utilization, and Indexes of Java Barb Condition

Parameters	Dietary lysine levels (%)					
	1.27	1.52	1.78	2.02	2.27	2.52
Initial Weight (g)	3.56±0.28	3.58±0.35	3.55±0.29	3.57±0.29		3.52±0.26
Final Weight (g)	25.93±1.1	43.97±1.04	32.97±1.1	27.96±1.03	23.96±1.02	15.95±1.03
AWG (g)	22.37±1.24	40.39±1.24	29.42±1.21	24.39±1.21	20.44±1.21	12.43±1.24
ADCp (%)	65.72±0.28[c]	71.46±0.23[a]	68.19±0.32[b]	67.82±0.27[b]	62.82±0.27[d]	60.15±0.30[e]
EFU (%)	64.19±0.07[c]	70.78±0.02[a]	66.35±0.04[b]	65.63±0.08[b]	60.35±0.03[d]	58.43±0.06[e]
RGR (%/day)	3.37±0.03[c]	4.03±0.09[a]	3.78±0.01[b]	3.57±0.06[c]	3.18±0.03[d]	2.51±0.02[e]
FCR	2.24±0.03[c]	1.78±0.0[a]	2.16±0.05[b]	2.22±0.05[c]	2.32±0.05[c]	2.56±0.01[d]
PER	2.52±0.02[b]	3.01±0.17[a]	2.76±0.06[b]	2.33±0.11[bc]	2.13±0.11[c]	1.58±0.04[d]
HIS (%)	1.4±0.02	1.3±0.02	1.2±0.03	1.1±0.01	1.0±0.02	1.0±0.01
VSI (%)	1.5±0.47	1.5±0.26	1.5±0.02	1.5±0.04	1.5±0.03	1.5±0.09
SR (%)	93.85±2.47[a]	93.85±2.47[a]	100±0.00[a]	100±0.00 [a]	100±0.00[c]	93.85±2.47[a]

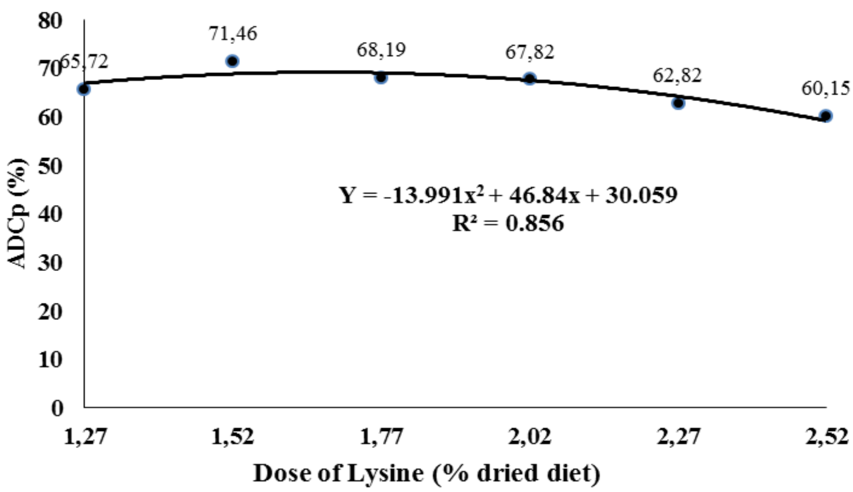

Figure 1. Relationship of Lysine in the Diet and ADCp

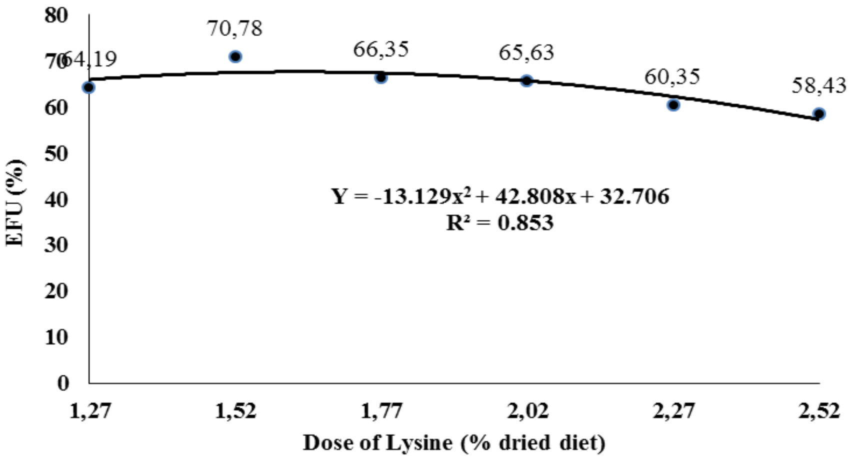

Figure 2. Relationship of Lysine in the Diet and EFU

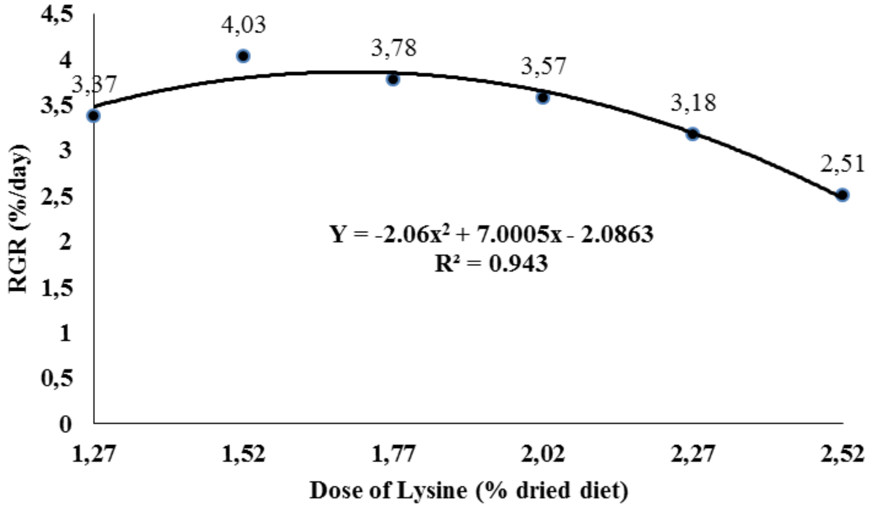

Figure 3. Relationship of Lysine in the Diet and RGR

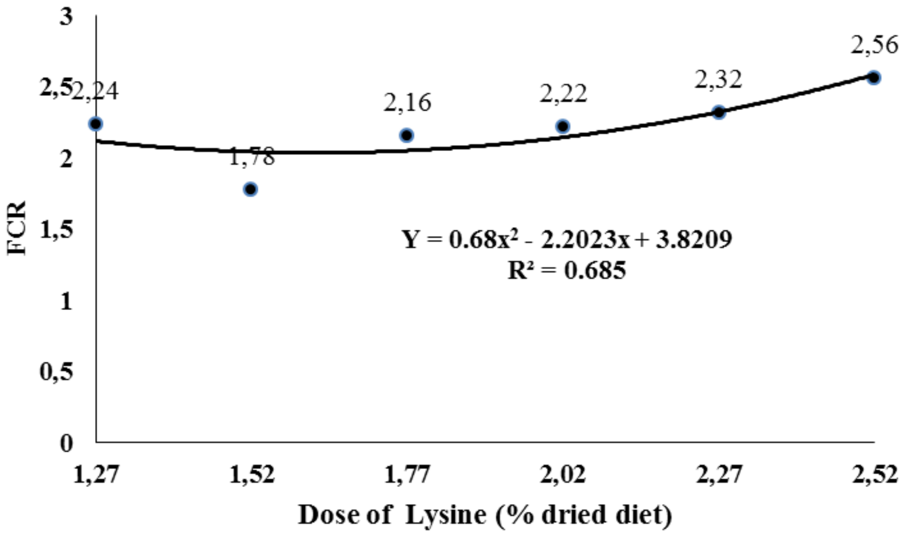

Figure 4. Relationship of Lysine in the Diet and FCR

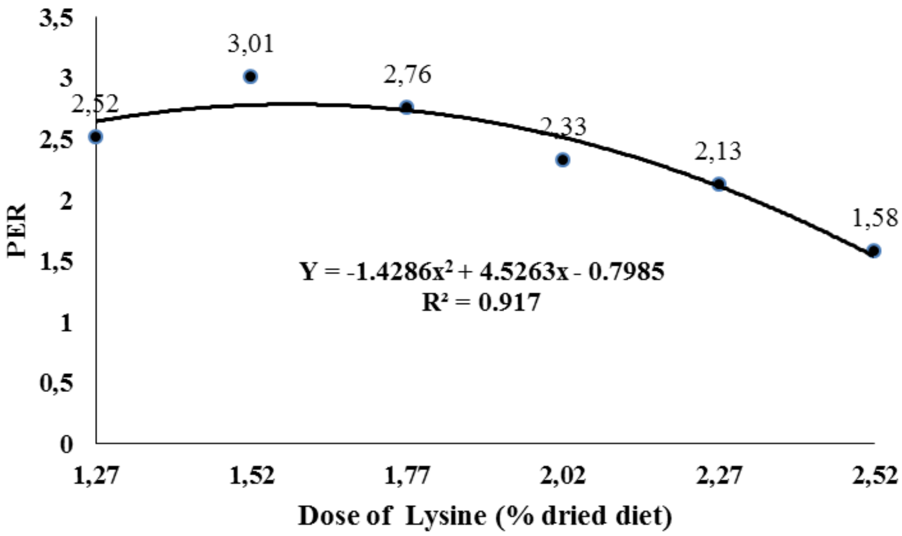

Figure 5. Relationship of Lysine in the Diet and PER

Somatic Indices (HIS and VSl)

The results show that the Java barb fingerlings given lysine supplemented diet with the low dosage of 1.27% (A1) generated the highest value of HSI (Table 3) which would decrease with higher lysine doses. Meanwhile, the various dosages of lysine in the feed did not affect on the values of VSI.

Carcass Composition

The supplementation of various dosages of lysine in the diet affected on the composition of carcass in the Java barb fingerlings (Table 4). The highest protein deposit in the Java barb carcass was attained when the fingerlings were fed with 1.52 % lysine supplemented diet. The higher the lysine in the diet is, the higher is the water content in the Java barb carcass. Moreover, the higher the lysine in the diet is, the lower the ash in the Java barb carcass becomes.

Table 4. Body Contents of Java Barb Fed With the Diets Containing Various Lysine Dosages

($g\ 100\ g^{-1}$ wet	Dosages of lysine (% dried diet)						
weight)	Initial	1.27	1.52	1.78	2.02	2.27	2.52
Moisture	71.6±0.3	72.5±0.4	73.4±0.2	74.3±0.3	74.9±0.2	75.7±0.3	76.3±0.4
Protein	12.8±0.2	16.2 ±0.5	18.3±0.1	17.6±0.3	16.7±0.4	15.8±0.5	14.7±0.3
Lipid	8.6±0.2	7.2±0.3	6.6±0.4	5.6±0.5	4.6±0.1	4.4±0.3	4.0±0.2
Ash	4.3±0.2	3.7±0.1	3.5±0.2	3.4±0.2	3.2±0.2	3.1±0.2	3.0±0.2

Discussion

The Java barb fingerlings that were given feed with the addition of lysine as much as 1.27% (A1) could generate lower values of AWG, RGR, ADCp, EFU, FCR, PER than those given higher dosages of lysine that ranged from 1.52% to 2.52% (A2, A3, A4, A5, A6). The diet that has lysine deficiency can lower feed palatability, growth and increase mortality of the fish (Mai *et al*., 2006). The fish in the study experienced no decrease in palatability, because the diet was supplemented with lysine. The fish was very active during feeding and has eaten all available feed in aquarium. The results show that the low growth and feed utilization in the Java barb fingerlings was not because of lacking of available feed, but it was because of lacking of lysine in the diet, therefore it hampered protein synthesis or other fish metabolism. The same discovery was reported by Abboudi *et al*. (2006).

The dosage of 1.52 % (A2) lysine in the diet was the best dosage, since it generated the highest growth and feed utilization. The lower growth and feed utilization were found at the dosage below or above 1.52% of lysine. The results, as presented in the Table 3, exhibited the decreases of ADCp, EFU, RGR and PER values in the Java barb fingerlings which were given the lysine-supplemented diet with the higher dosages (1.78-2.52%) than they needed. The results were similar to the discoveries by Walton *et al*. (1984) in the *Salmo gairdneri*, Murthy and Varghese, (1997) in the *Labeo rohita*, Xie *et al*. (2012) in the *Litopenaeus vannamei*; Fang Deng *et al.* (2010) in the *Polydactylus sexfilis*, Ebeneezar *et al*. (2019) in the *Trachinotus blochii*, Farhat and Khan, (2013) in the *Heteropneustes fossilis*.

The results of treatments A3-A6 (1.78-2.52%) that were given higher than optimal dosages demonstrated the decreases of the growth and feed utilization; however, they cannot be concluded due to the toxicities of over-supplied of lysine in the feed (Farhat and Khan, 2013). Therefore, the low growth and feed utilization were because of lysine deficiency or excess in the feed (1.27% and1.78-2.52%). Murthy and Varghese (1997) disclosed that the amino acids deficit or surplus reduced feed palatability. It was suggested that the low growth and feed utilization was due to the fish given without optimal dosages of lysine-supplemented diet (1.26% and 2.27-2.52%). Similar results were documented by Mai *et al.* (2006) on Japanese sea bass, Luo *et al.* (2006) on grouper, Walton *et al.* (1984) on rainbow trout, Nguyen and Davis, (2016) on *Rachycentron canadum*.

The increase of lysine dosage in the diet caused significant difference on the body composition of Java barb fish. The Java barb fish given the feed containing 1.52 % of lysine had higher protein deposit in their body compared to the fish given the feed containing 1.27%, 1.78%, 2.02%, 2.25% and 2.52% of lysine in the dried diet. According Luo *et al*. (2006) the protein contentt in the fish depended on the lysine-supplemented diet availability. Moreover, Furuya and Furuya (2010) and Hamid *et al*. (2016) suggested that lysine had a role in the development of protein deposit in the fish and also the nitrogen retention increase. The highest fat content in the Java barb was found in the treatment A1 that had lowest dosage of lysine in the diet (1.27%) and kept decreasing as the dosages of lysine increasing, especially in the range dosages of 1.52%-2.52% (A2-A6). The decrease of fat content in the fish body was a response on the availability of lysine-supplemented diet (Luo *et al.* 2006). Other important roles of lysine were to synthesize carnitine (Walton *et al*., 1984), that has important function to carry long-chained fat from cytoplasm to mitochondria in order to do β-oxidation (Wu, 2013). Therefore, the fish that had lysine deficiency in the diet accumulated lipid deposit in the body, as reported by Ruchimat *et al.* (1997) on yellow tail, Walton *et al.* (1984) on rainbow trout, Xie *et al.* (2012) on large yellow croaker, Zhou *et al*. (2010) on black sea bream, Yang *et al*. (2011) on silver perch and Helland *et al*. (2011) on Atlantic salmon.

The results show that the Java barb fingerlings given lysine supplemented diet with the low dosage of 1.27% (A1) generated the highest value of HSI (Table 3). As reported by Bicudo *et al*. (2009) that the lower dosage of lysine in the feed resulted in higher HSI. The negative correlation between lysine dosages in the feed and HSI were also reported by Espe *et al*. (2007) on Atlantic salmon, Tibaldi *et al*. (1994) on European sea bass, Marcouli *et al.* (2006) on gilthead sea bream, Zhou *et al*. (2007) on cobia, Peres and Oliva-Teles, (2008) on turbot, Bicudo *et al*. (2009) on pacu, Luo *et al*. (2006) on grouper. This happened because lysine was supposed to synthesize protein instead being used to convert muscle into fat or glycogen; therefore, it increased HSI. Meanwhile, the various dosages of lysine in the feed did not affect on the values of VSI.

Until recently the manufactured diet used plantbased protein as the main protein origin in the diet. The suplementation of lysine amino acid in the diet was needed to balance amino acids contents. The lysine addition was also needed to support amino acids in order to be able to synthesize protein and to grow muscle. The results of Polynomial Orthogonal test suggested that the need of lysine amino acid for Java Barb fish ranged from 1.58% to-1.70% dried diet (5.3 to 5.6 % of protein diet). The results of this study were higher than their need as reported on rainbow trout (*O. mykiss*) as much as 1.7% (Bodin *et al*., 2009), yellow tail (*Seriola quinquiradiata*) as much as 1.66% (Ruchimat *et al*., 1997). However, the need of lysine in this study was much lower than the need for grouper fish (*Epinephelus coioides*) as much as 2.8% (Luo *et al*. 2006), gilthead sea brain (*Sparus murata*) as much as 2.15% (Marcouli *et al*., 2006), Japanese sea bass (*Loteolabrax japonicas*) as much as 2.49-2.61% (Mai *et al*., 2006), Large yellow croaker (*Pseudosciaena crocea*) as much as 2.5% (Zhang *et al*., 2008), Black sea bream (*Sparus macrocephalus*) as much as 3.3% (Zhou *et al*., 2010), fingerling stinging catfish (*Heteropneustes fossilis*) as much as 2.0% to 2.3% (Farhat and Khan, 2013), juvenile Silver pompano (*Trachinotus blochii*) as much as 2.40–2.45% (Ebeneezar *et al*., 2019). The various need of the lysine amino acid in every species were thought due to the different sources of protein (Forster and Ogata, 1998), feeding pattern, size, age, genetic, species and cultivation (Nguyen *et al*., 2013).

CONCLUSION

The needs of lysine in the diet for Java barb (*P. javanicus*) fingerlings to optimize feed utilization, growth, and nutrient contents ranged from 1.58% to-1.70% dried diet (5.3 to 5.6 % of the crude protein diet).

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