The growth, protein content, and fatty acid of catfish meat (pangasius sp.) With the addition of different lysine doses in commercial feed

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Abstract. Pangasius catfish are not able to synthesize amino acids in their bodies so that Pangasius catfish need a supply of amino acids such as lysine in their feed. This study aims to find out the effect of lysine addition in commercial feed towards the growth, protein content, and fatty acid of Pangasius catfish meat. Total doses of lysine added to the commercial feed are: P0 (0%), P1 (1.2%), P2 (2.2%), dan P3 (3.2%). The results showed that the addition of lysine essential amino acids in commercial feed showed a significantly different effect (P<0.05) on the specific growth rate, feed efficiency, feed conversion ratio, Saturated Fatty Acid (SFA), and Monounsaturated Fatty Acid (MUFA) of Pangasius catfish meat. On the other hand, Polyunsaturated Fatty Acid (PUFA) showed a very significant difference (P<0.01) with lysine addition dose of 3.2% and no significant effect (P>0.05) on SR. The results of this study showed that lysine addition dose of 1.2% could increase SGR, feed efficiency, and FCR, and lysine addition dose of 2.2% can increase the protein content of Pangasius catfish meat also lysine addition dose of 3.2% provides the best performance for fatty acids of Pangasius catfish meat.

1. Introduction
One of the potential freshwater fish to be cultivated in Indonesia is catfish (Pangasius sp.). In 2012 the production of catfish in Indonesia increased up to 651,000 tons [1];[2];[3];[4];[5]. One of the ways to increase the production of catfish is through input production in the form of fulfillment of the nutritional needs of fish feed. Nutritional needs of fish feed is the most important element that can be used for growth and survival of fish [6];[7];[8];[9];[10];[11]. The quality of the feed is said to be low if the essential amino acid content also low. Essential amino acids cannot be formed or synthesized by fish or shrimp bodies, so it should be available within the feed [12]. (Hermiastuti 2013). The addition of supplements to feed is to spur growth, to increase
the protein of catfish, to decrease the fat content in fish, and to increase the efficiency of production [13]. One of the supplements that can be given to the feed is lysine. Lysine is one of the essential amino acids.

Lysine has a role to form carnitine, which has a function as a growth promoter, to protect against ammonia toxicity, and to enhance the defense of the body against extreme temperature changes [14]. The addition of lysine within the feed can increase protein synthesis in the fish body, so the protein content in fish meat will increase and affect the process of fish growth and their survival as well [15]. Lysine also has a function as the basic ingredient of blood antibodies, to strengthen the circulatory system, and to maintain the growth of normal cells [16]. Therefore, the addition of lysine into the feed is expected to increase the formation of carnitine that has a role in the oxidation of fatty acids. Fatty acids are differentiated into saturated fatty acids and unsaturated fatty acids. The composition of fatty acids in freshwater fish is different from the content of fatty acids in saltwater fish. The effects of saturated fatty acids cause an increase in LDL (Low-Density Lipoprotein) and also can decrease HDL (High-Density Lipoprotein). Lack of unsaturated fatty acids will cause disruption to fish health including reduced fecundity, ability to form embryos and abnormal growth [17].

Since scientific research regarding the addition of lysine to commercial catfish feeds on growth, protein, and fatty acids of catfish have not been conducted, so it is necessary to conduct research that discusses about the addition of lysine in commercial feed to evaluate the growth potential, protein content, and fatty acids in catfish meat (*Pangasius* sp.).

2. **Material and methods**

2.1. **Place and Time of the Research**

The research was conducted in April-May 2017 at the Laboratory of Faculty of Fisheries and Marine Airlangga University, Surabaya and Animal Feed Laboratory Faculty of Veterinary Medicine, Airlangga University, Surabaya.

2.2. **Preparation**

The initial step of the research starts by preparing and cleaning the tools that are going to be used. The tools used are including 20 aquariums measuring 30 cm x 30 cm x 40 cm, aerator, aeration hose, 20 aeration stones, sliding term, water tank, fishing net, and chiffon tool. The tools that have been cleaned are chlorinated and soaked for 24 hours, then rinsed and dried. After that fill the aquarium with fresh water that has been deposited from the tank as much as 22.5 liters/aquarium and install the aeration system. The process of preparing aquarium and water filling is done two days before the fishes are stocked. Fishes used as experimental material are catfish derived from fish farmers in Lamongan, East Java measuring 6-8 cm. Fish that will be stocked in the aquarium undergo acclimatization process or adaptation for approximately 30 minutes with a density of 10 fishes/aquarium. Fish stocking is done in the morning or afternoon.

2.3. **Provision of Experimental Feed**

The feed used in the study is commercial feed pellet-shaped 781-1. Other materials are commercial liquefaction and tapioca flour. The materials to be used are analyzed proximate in advance. The results can be seen in Table 1.

After the proximate test, the determination of lysine dose and tapioca flour that added to the feed for each treatment is conducted. First, pellets are milled and then the ingredients are mixed from the smallest amount to the larger amount. After the mixing process, water is added to homogeneous material as much as 20-40% then mixed until homogeneous and can be fisted. The dough mixture is steamed for 20 minutes and printed in a pellet machine according to the size of the fish's mouth opening. The mold is accommodated on a tray and the pellet is dried in an oven with a temperature of 60 °C for 12-24 hours [18].
Table 1. Result of commercial feed proximate analysis (P0), commercial lysine, and tapioca flour

| Nutrition Composition | Commercial Feed (P0) | Commercial Lysine | Tapioca Flour |
|-----------------------|----------------------|-------------------|---------------|
| Dry Matter %          | 91.3125              | 95.8199           | 89.5180       |
| Ash %                 | 8.7339               | 0.5890            | 0.1596        |
| Crude Protein %       | 23.5577              | 67.5913           | 1.7361        |
| Crude Fat %           | 5.991                | 2.4807            | 0.5738        |
| Crude Fiber%          | 5.6859               | 0.5853            | 0.5809        |
| BETN %                | 47.3440              | 24.5736           | 86.4676       |
| ME (kcal/kg)          | 2971.4907            | 3336.7985         | 3281.1442     |

Note BETN (Extracts Material without Nitrogen) and ME (Energy Metabolism).

The study consisted of four treatments and five replications as follows:
P0: Commercial feed 100%
P1: Commercial feed 100% + lysine 1.2% + tapioca flour 1%
P2: Commercial feed 100% + lysine 2.2% + tapioca flour 1%
P3: Commercial feed 100% + lysine 3.2% + tapioca flour 1%

2.4. Maintenance of Catfish
At the beginning of maintenance, fish is fasted for 24 hours, then weighed to get initial weight. Fish maintenance is conducted for 30 days. Feeding as much as 5% of body weight with twice-daily frequency at 08:00 and 16:00 pm. The amount of feed given is determined every 7 days, the calculation of biomass by sampling catfish as much as 5 fishes per aquarium.

At the end of the maintenance, fish is fasted again for 24 hours, then calculated and weighed to determine the final weight. During maintenance the water quality parameters measured are the temperature about 27.4-28.7°C, pH 7.0, dissolved oxygen ranged between 4.47 - 7.08 mg / l, and ammonia of 0.003 mg / l.

2.5. Parameters of the Research
The observed parameters during the research were Specific Growth Rate (SGR), Feed Efficiency (FE), Feed Conversion Ratio (FCR), Survival Rate (SR), Protein Content, and the content of saturated fatty acids and unsaturated catfish meat (Pangasius sp.). Here’s how the calculations used in this research:

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SGR = \frac{\ln Wt - \ln We}{t} \times 100\% \quad [15]
\]

Description:
SGR = specific growth rate (% / day)
Wt = average weight of fish at the beginning of maintenance (g)
Wo = average weight of fish at the start of maintenance (g)
t = maintenance time (day)

\[
FE = \frac{(Wt+D)-We}{F} \times 100\% \quad [19]
\]

Description:
FE = feed efficiency (%)
Wt = weight of fish at end of maintenance (g)
Wo = weight of fish at the beginning of maintenance (g)
D = the weight of dead fish during maintenance (g)
F = amount of feed consumed (g)
FCR = \frac{F}{(Wt+D) - W_0} \quad [15]

Description:
FCR = feed conversion ratio  
F = amount of feed consumed (g)  
W = weight of fish at end of maintenance (g)  
Wo = weight of fish at the beginning of maintenance (g)  
D = the weight of dead fish during maintenance (g)

SR = \frac{N_t}{N_0} \times 100\% \quad [15]

Description:
SR = survival rate of fish (%)  
N_t = number of fish at the end of maintenance (tail)  
N_0 = number of fish at the beginning of maintenance (tail)

2.6. The Analysis of Data
Analysis of data used is Analyze of Variance (ANOVA) to find out the effect of treatment given. If there is a significant difference between treatments, then proceed with Duncan's Multiple Range Test.

3. Result and discussion
The results of the statistical analysis can be seen in Table 2. The addition of different lysine doses on commercial feeds has a significant effect (P<0.05) on specific growth rates, feed efficiency, and feed conversion ratio of catfish. On the other hand, the feed treatment did not give significant effect (P>0.05) on the survival rate of the pangasius catfish.

Table 2. Specific growth rates (SGR), feed efficiency (FE), feed conversion ratio (FCR), and survival rate (SR) of pangasius catfish.

| Parameters             | P0 (0%)   | Treatment of Lysine Dose on the Feed | P3 (3.2%)   |
|------------------------|-----------|--------------------------------------|-------------|
| SGR (%BW/day)          | 2.129±0.579\textsuperscript{a} | 2.977±0.415\textsuperscript{b} | 3.028±0.134\textsuperscript{b} | 2.856±0.472\textsuperscript{b} |
| EP (%)                 | 46.948±12.044\textsuperscript{a} | 73.937±13.012\textsuperscript{b} | 76.077±6.825\textsuperscript{b} | 72.109±10.949\textsuperscript{b} |
| FCR                    | 2.278±0.736\textsuperscript{a} | 1.388±0.251\textsuperscript{b} | 1.323±0.121\textsuperscript{b} | 1.411±0.204\textsuperscript{b} |
| SR (%)                 | 88.00±8.367\textsuperscript{a} | 94.00±5.477\textsuperscript{a} | 98.00±4.472\textsuperscript{a} | 86.00±13.416\textsuperscript{a} |

Note: Different superscript on the same line indicates inter-treatment has a significant difference (P<0.05). The value shown is the average value and standard deviation.

Treatment P0 produced the specific growth rate and the lowest feed efficiency compared to P1, P2, and P3 treatment ie 2.129±0.579% / day and 46.948±12.044%. The treatment of P0 resulted in the highest conversion ratio of catfish feed compared to P1, P2, and P3 treatment of 2.278±0.736. However, P1, P2, and P3 treatment showed that the specific growth rate, feed efficiency, and feed conversion ratio of catfish were not significantly different (P>0.05). The degree of survival of catfish during 30 days of maintenance between 86-98%.

The addition of lysine in the commercial diet showed significant differences (P<0.05) on protein content, Saturated Fatty Acid (SFA), Monounsaturated Fatty Acid (MUFA), while Polyunsaturated Fatty Acid (PUFA) in catfish meat showed a high difference result (P<0.01). The results can be seen in Table 3.
Table 3. Protein content, Saturated Fatty Acids (SFA), Monounsaturated Fatty Acids (MUFA), and Polyunsaturated Fatty Acids (PUFAs) of catfish.

| Description | Treatment of Lysine Dose on the Feed (%) |
|-------------|----------------------------------------|
| Protein (%) | P0 (0%)  | P1 (1.2%)  | P2 (2.2%)  | P3 (3.2%)  |
| SFA (%)     | 13.288±0.203 | 13.754±0.261 | 14.532±1.136 | 13.730±0.434 |
| MUFA (%)    | 4.966±1.4542 | 5.686±1.2616 | 5.403±0.6675 | 3.588±0.9282 |
| PUFAs (%)   | 7.246±1.1659 | 5.712±0.3942 | 5.963±0.7992 | 5.568±0.8716 |

Description: Different superscript on the same line indicates inter-treatment has a significant difference (P<0.05). The value shown is the average value and standard deviation.

The statistical analysis showed that the treatment with the highest protein content was 14.532±1.316% found in treatment of P2, the highest Saturated Fatty Acid (SFA) was 5.686±1.2616% in treatment of P1, the highest unsaturated fatty acid of MUFA was 7.246±1.1659% found in the treatment of P0, and the highest unsaturated fatty acid of PUFAs is 23.108±3.4405% found in treatment P3.

The results of this research found that the Specific Growth Rate (SGR) of pangasius catfish was the lowest in treatment P0 (control) which equals to 2.129% /day. This is due to the amino acid composition in the tested commercial feed unbalanced. The limited availability of lysine can cause other amino acid content to be inactivated by the body for protein synthesis, thus causing protein stores in the body to below and will cause decreased growth rate [13].

Specific Grow Rate (SGR) of catfish at P1 treatment was 2.977% BW/day and P2 treatment was 3.028% /day. The increased growth of fish by the addition of lysine in the feed caused by lysine which is one of the essential amino acids that have a role as a substrate to produce carnitine. Carnitine serves as a growth promoter [14]. Lysine also serves to maintain the growth of normal cells [16]. P3 treatment decreased the specific growth rate of 2.856% /day. Increased intake of lysine will affect the metabolism that is antagonistic with arginine. Inhibition of arginine absorption occurs so that it can lead to decreased growth rate [20].

The value of feed efficiency and feed conversion ratio showed the efficiency of nutrient utilization of feed by fish. Feed efficiency is increased when the value of the feed conversion ratio is lower. The efficiency of feed on treatment P1, P2, and P3 was higher (P<0.05) than treatment P0. The ratio of feed conversion on treatment P1, P2, and P3 was lower (P<0.05) than treatment P0. This result showed that lysine added to the feed can improve the efficiency of catfish feed. Several factors affecting feed efficiency are including fish species, fish body weight, feed type, feed quality, feeding techniques, and water quality [21].

The lysine content can improve feed efficiency and decrease feed consumption [22]. The addition of lysine in the diet tends to increase the protein, fat, and energy content in the feed. Energy content in the feed can also determine the high or low feed efficiency. Moreover, the addition of lysine in the feed can improve the digestibility of the ileum (intestine), so that nutrients can be absorbed quickly in order to make fish quickly satisfied, high growth rate, and to make feed efficiency increases. Lysine that has been given to feed can also increase the digestibility of other amino acids, one of which is the non-essential tyrosine amino acid that can regulate fish appetite and the body's response to stress [23].

The addition of lysine does not affect the degree of survival of catfish. This is because the lysine added to the feed with no lysine added to the feed has a balance or the same utility, so there is no significant difference. The likelihood of anti-body formation in the catfish body is similar to the formation of antibodies with untreated lysine, so the result has no significant difference.

Statistical analysis showed that the protein content of P2 treated fish meat was significantly
different from P0 and there was no significant difference with P1 and P3. The protein content of catfish meat 14.5% [24] with lysine 2.2% in 1,000 grams of commercial feed can maintain the amount of protein content of catfish meat. Means 2.2% of lysine added in commercial feed can maintain a meat protein content of 14.532%. Lysine added in a commercial feed of 3.2% produced protein content of 13.73%, which means that adding lysine 3.2% cannot improve the lysine performance so that the protein in the meat does not increase. The antagonistic lysine properties can suppress the metabolism of other amino acids such as the essential amino acid arginine which will lead to protein synthesis not working properly.

The content of Saturated Fatty Acid (SFA) treatment of P3 was significantly different from P1, P2, and was not significantly different from P0. The content of MUFA unsaturated fatty acids in the treatment of P0 was significantly different from the treatment of P1, P2, and P3. The content of PUFA unsaturated fatty acid in the treatment of P3 was significantly different from the treatment of P0, P1, and P2. Saturated Fatty Acids (SFA) in this research showed positive results with a decrease in percentage in each treatment given. The best treatment is in P3 with 3.2% lysine addition which can decrease SFA level to 3.5882% from the original control treatment 4.966%. This happens because of lysine containing carnitine which is able to metabolize well with fatty acids that can be used as energy. Meanwhile, MUFA obtained in treatment P0 showed higher results compared with the treatment of P1, P2, and P3. It is presumed that catfish metabolism on the treatment of P1, P2 and P3 is different from the metabolism of catfish treatment P0, so the addition of lysine essential amino acid did not show the increase of catfish meat MUFA. According to [25], the content of fatty acids in the body depends on the ability of fish in deciphering essential fatty acids both anabolic and catabolic naturally.

The value of PUFA unsaturated fatty acids in this research showed positive results in which an increase in the content of unsaturated fatty acids of PUFA at commercial feed was added with lysine amino acid by 2.2% (P2) and 3.2% (P3), but at the time of commercial feed added 1.2% (P1) showed a lower result than the control treatment (P0). The act of giving lysine can increase the content of unsaturated fatty acids of PUFA because lysine is capable of producing carnitine which is key in fatty acid metabolism in the catfish body. In line with that, saturated fatty acid can be metabolized and formed into energy for fish growth. This is similar to the statement stated by [26] that lysine essential amino acids as carnitine precursors added to commercial feed can help the metabolism of fatty acids.

Lysine essential amino acid given to commercial feed has the potential to fatty acid metabolism in the body of catfish. This can be seen from the content of saturated fatty acids contained in catfish meat is lower than the content of unsaturated fatty acids. The results showed that the provision of essential amino acids was significantly different in fatty acids to saturated fatty acids and unsaturated fatty acids of MUFA (P<0.05) and differed significantly in PUFA unsaturated fatty acids (P<0.01).

4. Conclusion

The addition of 1.2% lysine can increase the growth, feed efficiency, and feed conversion ratio of Pangasius catfish, while the addition of 2.2% lysine can increase the protein content of meat and unsaturated fatty acids of MUFA. Also, the supplementation of 3.2% lysine may increase the content of unsaturated fatty acids of PUFA but decrease saturated fatty acid content of SFA. However, the addition of lysine in commercial feed did not affect the degree of survival of catfish.

5 References

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