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## Addition of lemuru fish oil in the diet on the fat retention and fatty acid profile of silver barb (*Rasbora argyrotaenia*)

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**Abstract.** One important factor in cultivation is feed. The addition of fish oil to feed affects the activity of the protein and cell synthesis so that it affects the growth rate, feed conversion ratio, and protein and fat contents of the fish body. This study aimed to determine the effect of the addition of lemuru fish oil in feed on fat retention and fatty acid profiles of silver barb (*Rasbora argyrotaenia*) fish. This study was used as an experimental method with a completely randomized design consisting of five treatments and four replications for each treatment. The lemuru fish oil with different concentrations, namely 0, 2, 4, 6, and 8% were added to commercial feed of silver barb. The parameters of fat retention and fatty acid profile were observed. Data were analyzed statistically using a variant of analysis followed by Duncan's multiple range test. The results showed that the addition of lemuru fish oil in the fish feed for 40 days of fish rearing had a significant effect ( $p < 0.05$ ) on fat retention and increased fatty acid profile of silver barb. The 2% treatment has the best of fat retention (53.36%). A fat concentration of 9% in the diet increase unsaturated fatty acids content of silver barb (66.32%).

### 1. Introduction

Silver barb (*rasbora Argyrotaenia*) is a native Indonesian fish spread on the island of Sumatra, Java, Bali, Nusa Tenggara, Kalimantan, and Sulawesi [1]. In some regions in Indonesia, the silver barb was *overfishing* so that the cultivation of silver barb to kept awake the population [2]. One of the important factors in cultivation is feed. The nutritional value of fish feed is generally seen from the composition of nutrients, such as protein, fats, carbohydrates, vitamins, and minerals [3].

The addition of fish oil in feed may affect the activity of lipogenic enzymes to synthesize fatty acids. And also influence on the activity of the enzymes in the cell membranes to the protein and cells, to influence the rate of growth, convection of feed, water content in the flesh, the content of proteins and body fat, as well as fish liver weight [4]. Based on that, the ability of lipogenic enzymes will affect the fatty acid profile on the feed with different quality and quantity. The wild silver barb has omega 3 high content of  $20.31\% \pm 0.64$  and omega 6 for  $16.40\% \pm 1.42$  [5].

Fish Oil results in the body of the Lemuru fish containing EPA 2.76%, DHA 12.97%. Addition of essential fatty acids from lemuru fish oil as a step increase in the fatty acid profile of the silver barb cultivated [6]. This is due to the cultivation of silver barb using commercial feed can change the



content of fatty acids. The purpose of this research is to increase the influence of the addition of the lemuru fish oil in the feed against the fat retention and fatty acid profile of the silver barb, as well as to know the concentration of lemuru fish oil that can provide fat retention and fatty acid profiles of silver barb. The benefit of this research is to provide information on the influence of the addition of the lemuru fish oil in feed to fat retention and fatty acid profile so that It can be applied to the cultivation to grow fast and optimally.

## 2. Materials and methods

This study was conducted in the Laboratory of the Faculty of Fisheries and Marine, Universitas Airlangga and Saraswanti Indo Genetech Laboratory, Bogor, West Java, Indonesia.

### 2.1 Test organisms

In this study, the observed fish were Silver barb 30-40 mm in size with a  $0.49 \pm 0.01$  g in average weight, from the Installation of Freshwater Aquaculture Umbulan, Pasuruan, East Java, Indonesia.

### 2.2 Test feed production

Commercial feed in this research contained 36.95% protein and 5.29% fat, feed diameter was 0.7-1.0 mm. lemuru fish oil was added in the decided amount of concentration (0 %/ as control, 2 %, 4 %, 6 %, and 8 %) and mixed with 2 mL/ 100 g of egg white infed according to Paratiwi's method [7]. Next, added 100mL of water to the composition for every 100 g of fish feed like in Mokoginta's method [8]. Mixed commercial fish feed and lemuru fish oil evenly by spraying. The mixed feed was dried in the oven at 60 °C in 48 hours according to Saltin's method [9]. The approximate analysis of feed results was presented in Table 1.

**Table 1.** Proximate analysis of feed

Proximate analysis	Lemuru Fish Oil Concentration (%)				
	0	2	4	6	8
Protein (%)	38,37	38,39	38,41	38,26	38,61
Lipid (%)	7,63	7,66	9,46	10,16	12,64

### 2.3 Fish rearing

The silver barb was reared in a 20 L volume aquarium at a density of 3 fish/L for each treatment. The fish fasted for twenty-four hours before the treatment was done. Water was replaced every 2-3 days about 25% of the volume of the treatment unit.

### 2.4 Treatment

This study employed a complete randomized design (CRD) as an experimental method with five treatments of lemuru fish oil concentration and four replications respectively. The concentration of Lemuru fish oil addition in the fish feed based on cyprinid fish fat requirement is 5-15% and the optimum fat level in fish feed is 12% according to Kaushik's method [10]. The treatments were done through the addition of lemuru fish oil in the feed (0 %/ as control, 2 %, 4 %, 6 %, and 8 %) for 42 days. Feeding frequency was three times a day as in Masitoh's method [11] for 4 % biomass weight according to Mukti's method [12].

### 2.5 Observation result

Fat retention is calculated using this following formula [13]:

$$\text{Fat retention (\%)} = \frac{\text{final fat (g)} - \text{initial fat (g)}}{\text{fat feed (g)}} \times 100\%$$

$$\text{Initial fat (g)} = \frac{\text{initial fish fat (\%)}}{100\%} \times \text{initial fish weight (g)}$$

$$\text{Final fat (g)} = \frac{\text{final fat fish (\%)}}{100\%} \times \text{final fish weight (g)}$$

$$\text{Fat feed (g)} = \frac{\text{fat feed content (\%)}}{100\%} \times \text{amount of feed consumed (g)}$$

The profile of fatty acids can be known by analyzing silver barb using *Gas Chromatography Spectrometry Mass* (GC-MS).

### 2.6 Data analysis

The fat retention data obtained is processed using *Analysis of Variance* (ANOVA) to determine the effect of the treatment given. The additional treatment of the Lemuru fish oil on the commercial feed which shows significant then the calculations continued with a double distance test Duncan (Dunn's Multiple Range Test). Data on fatty acids profiles are analyzed using a descriptive method.

## 3. Result and discussion

### 3.1 Result

*Analysis of Variant* (ANOVA) statistic test result shows that the addition of the lemuru fish oil in the feed with different concentrations has an absolute influence on fat retention of silver barb ( $P < 0.05$ ). The highest fat retention is generated from the addition of 4% lemuru fish oil at 63.55%. The addition of 8% of lemuru Fish oil produces the lowest fat retention of 28.49%. The fat retention results of silver barb are presented in Table 2.

**Table 2.** Fat Retention of Silver Barb With The Addition of Lemuru Fish Oil on Feed

Lemuru Fish Oil Concentration (%)	Fat Retention $\pm$ SD
0	44.32 <sup>c</sup> $\pm$ 1,162
2	53.36 <sup>b</sup> $\pm$ 2,209
4	63.55 <sup>a</sup> $\pm$ 2,976
6	34.83 <sup>d</sup> $\pm$ 1,153
8	28.49 <sup>e</sup> $\pm$ 2,261

Results of the addition of lemuru fish oil in the feed shows the fatty acid profiles of Silver barb. The fatty acid profiles of Silver barb are presented in Table 3.

**Table 3.** Fatty Acid Profiles of Silver Barb (%) With The Addition of The Lemuru Fish Oil on Feed

Fatty Acid	Lemuru Fish Oil (%)	Lemuru Fish Oil Concentration (%)					Wild Silver barb* (%)
		0	2	4	6	8	
C12:0	0.16	0.2	0.15	0.18	0.17	0.11	n.d
C14:0	5.32	2.89	3.21	3.15	2.64	3.09	1.89
C15:0	0.39	0.52	0.47	0.56	0.36	0.51	1.33
C16:0	29.71	24.95	24.13	24.55	23.27	23.76	22.75
C17:0	0.33	1.06	0.79	0.94	0.58	1.21	n.d
C18:0	5.68	10.53	9.40	9.42	9.25	9.03	11.48
C20:0	0.53	0.66	0.49	0.60	0.62	0.59	0.33
C22:0	0.18	0.96	0.91	0.75	1.03	0.77	0.31
C24:0	0.11	0.36	0.29	0.30	0.30	0.19	n.d

C18:1n-9	26.41	41.16	41.99	48.53	43.60	42.92	14.29
C20:1	0.52	4.23	3.85	3.40	3.31	3.41	n.d
C20:3n-3	0.03	0.58	0.90	1.65	1.50	1.44	0.41
C20:5n-3	7.19	4.14	4.25	4.50	4.20	4.33	2.24
C22:1n-9	0.06	0.19	0.19	0.19	0.23	0.20	0.16
C22:6n-3	5.68	7.92	7.64	7.65	7.38	7.30	10.05
C24:1	0.23	0.33	0.35	0.40	0.33	0.29	0.23
ΣSFA	42.41	42.05	39.84	40.45	38.22	39.26	38.09
ΣUsFA	40.12	58.55	59.17	66.32	60.55	59.89	27.38
Rasio SFA : UsFA	1.06	0.72	0.67	0.61	0.63	0.66	1.39

Note: n.d = *not detected*, Wild Silver barb\* [5]

### 3.2 Discussion

The research showed that the addition of the lemuru fish oil in the feed with a different concentration affects fat retention as well as increase the fatty acid profile of silver barb. The addition of 4% lemuru fish oil in the feed (total fat in feed 9.46%) shows the highest fat retention of silver barb of 63.55%. Fats have a greater energy content than proteins and carbohydrates [14]. Fats are a major component in adipose tissue, which serves as a source of energy, an acidic source – essential fatty acids, body structure forming, precursors and a flavor enhancer [15]. Fat retention is an overview of the amount of feed fats absorbed and stored in the body of the fish during maintenance [16]. High fat concentrations in the feed when not worth the need, it will be stored as fish body fat [17]. Fat retention is influenced by the amount of feed, fat type, water temperature, degree of fat saturation and length of carbon chain [18].

Fatty acids are organic acids consisting of hydrocarbon chains, one has a hydroxyl group (COOH) and the other has a methyl group (CH<sub>3</sub>) [19]. Fatty acid can be distinguished by the level of saturation, saturated fatty acids (*Saturated fatty acid*/SAFA) and unsaturated fatty acids (*Unsaturated fatty acids*) consisting of *Monounsaturated fatty acids* (MUFA) and *Polyunsaturated Fatty Acid* (PUFA) [20].

The addition of the lemuru fish oil in the feed produces a different fatty acid profiles of silver barb. Saturated fatty acids showed a decline in the addition of 0% lemuru fish oil at 42.05% to 40.45% at concentrations 4% lemuru fish oil produced lemuru fish oil. The addition of essential fatty acids can decrease the percentage of saturated fatty acids [21]. The content of palmitic acid on silver barb according to Karapanagiotidis [5] amounting to 22.75%. The cultivated silver barb with the addition of 4% palmitic acid content of 24.55%. One of the main components of saturated fatty acids is palmitic acid ranges from 53-65% of the total saturated fatty acids [22]. Gunawan [23] states, the content of fatty acids on fish is influenced by development and growth, seasonality, salinity, water temperature, fish type, and fish feed.

Unsaturated fatty acids of silver barb of 66.32% on the addition of 4% lemuru fish oil, while with the addition of lemuru fish oil 0% produce unsaturated fatty acids by 58.55%. Feed with different fat content can affect the content of fatty acids in fish [24]. The fatty acids needed by fish include EPA and DHA. EPA and DHA are an important role in the process of growth and feed efficiency [25]. The cultivated silver barb with the addition of 4% lemuru fish oil produced EPA amounting to 4.50%, while cultivated silver barb without the addition of lemuru fish oil produced EPA by 4.14%. One of the EPA's roles is as a substrate of lipolytic digestive enzymes.

DHA content of silver barb on the addition of 0% lemuru fish oil amounted to 7.92%, DHA content decreases with the addition of lemuru fish oil. The height of n-6 fatty acids can inhibit the biosynthesis of DHA from n-3 fatty acids. Decreased fatty acid composition of EPA and DHA of *Anguilla bicolor* on the addition of fish oil on feed by 10% and 15% [4]. The content of DHA wild silver barb according to Karapanagiotidis [5] amounted to 10.05%. Omega-3 fatty acids EPA and DHA long

chains can naturally be obtained from fish fats especially seawater fishes. EPA and DHA acquired are the results of the synthesis of plankton which is the main feed of fish [26]. Kandemir and Polat [27] state that fatty acid content in aquatic organisms can be influenced by the treatment of free life in nature or cultivated.

#### 4. Conclusion

The addition of a lemuru fish oil in the feed affects fat retention and fatty acid profile. Under 4% (total feed fat 9.46%) addition of lemuru fish oil can decrease fat retention and unsaturated fatty acid content, it occurs in the addition of a lemuru fish oil more than 4%.

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