JPPIPA 9(6) (2023)

Jurnal Penelitian Pendidikan IPA

Journal of Research in Science Education

http://jppipa.unram.ac.id/index.php/jppipa/index

Response of Growth, Albumin, and Blood Glucose of Snakehead (*Channa Striata*) Juvenile Feed with the Addition of Different Animal Protein Sources

Novalina Serdiati[1*], Muhammad Safir[1], Umul Rezkiyah[1], R Adharyan Islamy[2]

[1] Aquaculture Study Program, Department of Fisheries and Marine Affairs, Faculty of Animal Husbandry and Fisheries, Tadulako University, Central Sulawesi, Indoensia.
[2] Departement of Aquaculture, Faculty of Fisheries and Marine Science, Brawijaya University, East Java, Indonesia.

Received: April 8, 2023
Revised: May 3, 2023
Accepted: June 25, 2023
Published: June 30, 2023

Corresponding Author:
Novalina Serdiati
novalinaserdiati@untad.ac.id

DOI: 10.29303/jppipa.v9i6.3618

Abstract: Snakehead fish or *Channa Striata* is a type of freshwater fish that has high economic value in Indonesia. Aims of this study was to evaluating the effect of adding different protein sources to snakehead fish feed on fish growth, blood glucose levels, and albumin content of snakehead fish (*Channa Striata*). This study consisted of 3 treatments and each was given 6 replications. Treatment A: Use of feed made from nilem fish flour. Treatment B: Use of feed made from golden snail flour and Treatment C: Use of feed made from earthworm flour. The best daily growth was obtained by using feed containing animal protein sources from earthworm flour of 1.85 g. The albumin level of snakehead fish at the end of rearing was highest in the treatment using feed made from earthworm flour (5.106%). followed by golden snail flour (4.802%) and nilem fish meal (4.492%). Snakehead fish blood glucose levels 2 hours after consuming the feed increased in all treatments, with the highest value in the treatment of golden snail flour and earthworms (112 mg/dL). Followed by nilem fish meal (105 mg/dL). However blood glucose levels 4 hours after consuming the feed decreased in all treatments. With the highest reduction occurring in the treatment using earthworm flour (38 mg/dL). Followed by golden snail flour (58 mg/dL) and nilem fish meal (92 mg/dL).

Keywords: Albumin; Blood glucose; *Channa striata*; Protein; Specific growth rate

Introduction

Snakehead fish or *Channa Striata* is a type of freshwater fish that has high economic value in Indonesia. They have great potential in the development of fish farming in Indonesia (Saputra et al., 2021). This fish is widely cultivated because of its delicious meat and increasing market demand (Bich et al., 2020). In addition, snakehead fish also has benefits in the health sector because of the nutritional content found in its meat. Increasing the production of snakehead fish needs to be done to meet the increasing market demand. Research on snakehead fish feed formulations continues to be carried out to improve feed quality in supporting the productivity and nutritional value of snakehead fish (Anjani et al., 2021; Permatasari et al., 2021; Rakhmawati et al., 2021). Several studies were also conducted to evaluate the potential of using animal protein sources in snakehead fish feed to increase the growth and quality of snakehead fish (Prastari et al., 2017). In this case, recent research showed that adding a source of animal protein to snakehead fish feed could increase the growth and nutrition of snakehead fish (Prastari et al., 2017).

According to previous research conducted by Ng et al. (2017), protein is an important component in fish feed, because protein is needed for the growth and repair of body tissues (Jia et al., 2022). Previous research has shown that increasing the protein content in fish feed can improve the growth and quality of fish (Adewumi, 2018; Tilami & Sampels, 2018; Hua et al.,

How to Cite:
Serdiati, N., Safir, M., Rezkiyah, U., & Islamy, R. A. (2023). Response of Growth, Albumin, and Blood Glucose of Snakehead (Channa Striata) Juvenile Feed with the Addition of Different Animal Protein Sources. *Jurnal Penelitian Pendidikan IPA*, *9*(6), 4685–4692. https://doi.org/10.29303/jppipa.v9i6.3618

2019; Lin et al., 2022). In addition, protein in fish feed can also affect blood glucose and fish albumin levels. Optimal blood glucose levels are important to ensure healthy carbohydrate metabolism and to avoid oxidative stress in fish (Hemre et al., 2002; Polakof et al., 2012; Soeprijanto & Islamy, 2023). Snakehead fish contains albumin protein (Pratama, et al 2020), which is the main protein in fish blood plasma, also has an important role in maintaining fish homeostasis and health (Belinskaia et al., 2020; Dwijayanti et al., 2016).

Basically, research on the effect of feeding from different protein sources on growth response, blood glucose levels, and albumin content of snakehead fish (*Channa Striata*) is very important to do in order to optimize the nutritional quality of cultured snakehead fish. Therefore, this study was conducted with the aim of evaluating the effect of adding different protein sources to snakehead fish feed on fish growth, blood glucose levels, and albumin content of snakehead fish (*Channa Striata*).

Method

This research was conducted from April to May 2022. The research took place at the Laboratory of Water Quality and Aquatic Biology, Faculty of Animal Husbandry and Fisheries, Tadulako University.

Preparation of the Test Fish

The test organisms used in the study were 18 snakehead fish seeds measuring 9-11 cm/head. Seeds were obtained from local residents in Mantikulore District, Palu City, Central Sulawesi. The snakehead fish used is acclimatized first by placing the fish in a test aquarium and leaving it for several days (usually 3-7 days) to adapt to the new environment. During the acclimatization period, the snakehead fish are fed regularly and the water quality in the aquarium is kept good. The acclimatization stage was carried out referring to the method of published journals (Armando et al., 2021; Islamy et al., 2017, 2023; Kilawati & Islamy, 2019).

Fish Feed Formulations

This study consisted of 3 treatments and each was given 6 replications: (a) Treatment A: Use of feed made from nilem fish meal, (b) Treatment B: Use of feed made from golden snail flour, (c) Treatment C: Use of feed made from earthworm flour.

The feed used is feed with a protein content of 32% which is formulated according to the ingredients to be used. Feed formulations are prepared with the same protein and energy content in the same feed (Table 1).

Table 1. Formulation of Snakehead Fish Feed (*Channa Striata*)

Feed Ingredients	Feed Formulation (g/100)		
	Treatment A	Treatment B	Treatment C
Sardinella flour	7.00	7.00	7.00
Worm flour	0.00	0.00	28.33
Golden snail flour	0.00	40.30	0.00
Nilem fish flour	50.00	0.00	0.00
Soybean Flour	18.00	12.70	4.00
Bran Flour	7.50	15.00	25.67
Corn starch	7.50	15.00	25.00
Vegetable oil	2.50	2.50	2.50
Fish oil	2.00	2.00	2.00
Tapioca flour	3.00	3.00	3.00
Mixed vitamins & minerals	2.50	2.50	2.50

Table 2. Proximate Test Results for Snakehead Fish (*Channa Striata*) Feed

Nutritional Content of Feed	Feed Proximate Test Results		
	Treatment A	Treatment B	Treatment C
Crude protein	40.95	36.66	25.76
Crude fat	10.67	9.12	7.92
Water content	8.66	2.97	7.73
Coarse fiber	5.16	3.79	4.96
Ash content	13.42	9.75	10.95

The feed used is artificial feed with feed raw materials as shown in Table 1. The procedure for making feed refers to Safir et al. (2020) namely starting from the weighing stage of feed raw materials, raw material mixing, printing and drying. The procedure for making flour from each raw material, namely for nilem fish, starts with cleaning the fish in running water and drying it in the sun for 7 days. While the manufacture of raw materials for golden snail feed begins with cleaning the golden snail from dirt and shells. Then dried in the sun for 7 days. Meanwhile, earthworms are first cleaned of dirt and then dried in the sun.

When all the feed raw materials have dried, then the feed raw materials are milled separately between the nilem fish. Golden snail and worms. After that, sifting all feed raw materials is carried out separately, including soy flour, corn flour and bran.

Next, weigh the raw materials according to the percentage of raw material use (Table 1). Fine feed raw materials are mixed with other feed raw materials. after all the ingredients are mixed evenly then steamed for ± 15 minutes. and after steaming, the mixing of liquid ingredients, including fish oil, is carried out. coconut oil and water. then printing using a pellet printer. The feed that has been printed is then cut according to the mouth size of the snakehead fish to be used in the study. Last stage. i.e. feed that has been printed is dried in the sun.

Fish Treatment

Snakehead fish seeds were kept in containers (20 cm x 18 cm in size) with a density of 1 fish/container. Each container with a volume of 2 liters of water is equipped with an aeration stone. The container used previously has been washed and dried in the sun.

Prior to stocking the snakehead fish, body weight was first measured. Body weight measurements were carried out at the beginning of maintenance and every 10 days until the end of the study. The study was conducted for 30 days. Feeding as much as 10% of the fish's body weight (Nasrin et al., 2021) with a frequency of feeding three times a day, namely morning, afternoon and evening (08.00, 12.00 and 17.00 WITA respectively). Water change is done once in 2 days until the end of maintenance

Test Parameters

Daily Specific Growth Rate (LPSH)

The specific growth rate is calculated by referring to the equation used by Hendy et al (2019) which are:

$$SGR\% = \frac{LnWt - LnWo}{\Delta t} \times 100 \quad (1)$$

Where, SGR = Daily specific growth rate (%/day); LnWt = Individual weight at the time of measurement (g), LnWo = Individual weight at the start of maintenance (g) dan Δ t = Long duration of fish maintenance.

Albumin Levels

Albumin levels were measured at the beginning and end of rearing according to published method (Saputra et al., 2018). 1 fish sample was taken at the beginning of rearing (before feeding treatment) and 2 test fish were taken from each treatment at the end of rearing and had been fasted for 2 days.

Analysis of albumin levels from the test fish samples (initial and late rearing fish) was carried out in the Tadulako University Faculty of Mathematics and Natural Sciences laboratory. Sample preparation by weighing the sample, then put into Erlenmeyer. Mix 25 ml of distilled water into Erlenmeyer and shake until homogeneous for 2 hours. The sample is then filtered using filter paper until the liquid is clear. The sample is transferred to a test tube and then centrifuged. Filtrate remove the gray measuring 100 ml and adjust it with distilled water. 1 ml of sample was mixed with 3 ml of bareford reagent. Then add distilled water until the volume reaches 10 ml. The sample was allowed to stand for 30 minutes and then its albumin content was measured using UV Vis spectrophotometry at the maximum wavelength.

The equation used to produce albumin levels is as follows (Fuadi et al. 2017).

$$\text{Albumin level (\%)} = \frac{(\frac{X}{1000})\ x\ Y\ x\ Fp}{W} 100 \quad (2)$$

Where, X = Concentration of fish albumin sample (ppm), Y = Volume (L), Fp = dilution factor; W= Sample weight (g).

Blood Glucose

Blood glucose levels of snakehead fish (*Channa Striata*) were measured at the Laboratory of Water Quality and Aquatic Biology, Faculty of Animal Husbandry and Fisheries, Tadulako University. Blood glucose levels were observed at the beginning and end of the study (four weeks of maintenance). According to published articles (Dai et al., 2022; Eames et al., 2010; Pottoo, 2017), before measuring glucose levels, the fish must be fasted for 48 hours. Blood collection began at 0 hour (before feeding) and 4 hours (after the fish were fed until they were full) (Safir et al. 2020). Blood samples were taken from the fish's veins using a syringe. Then the test strip is installed in the glucometer hole. Fish blood samples were measured using a test strip. After the blood sample on the test strip is evenly distributed. And then the automatic glucometer starts measuring blood glucose levels (measurement results are shown on the display screen in a countdown).

Data Analysis

Daily specific growth rate data body weight gain analyzed using one way analysis (Anova). If the treatment has an effect then the Dancan further test is carried out. While survival data albumin levels blood glucose and water quality were analyzed descriptively and displayed in figures and tables.

Result and Discussion

Daily Specific Growth Rate of Snakehead Fish (Channa Striata)

Daily specific growth rate of snakehead fish fed using animal protein sources in the form of nilem fish meal golden snail flour and earthworm flour showed a value of 0.64 respectively 0.88 and 0.93% per day. (Results of the analysis of variance (ANOVA) showed that the treatment of artificial feed with different animal protein sources had no effect (P>0.05) on the specific growth rate of snakehead fish fry.

The provision of artificial feed made from earthworm flour gave the highest daily specific growth rate for snakehead fish seeds (0.93%). The high specific daily growth rate in this treatment is thought to be caused by the eating habits of snakehead fish in nature.

Earthworms have the fishiest aroma compared to artificial feed made from nilem fish meal. Feed is determined by physical and chemical factors like shape, size, flavour, smell, aroma and color (Arditya et al., 2019; Barrett et al., 2010).

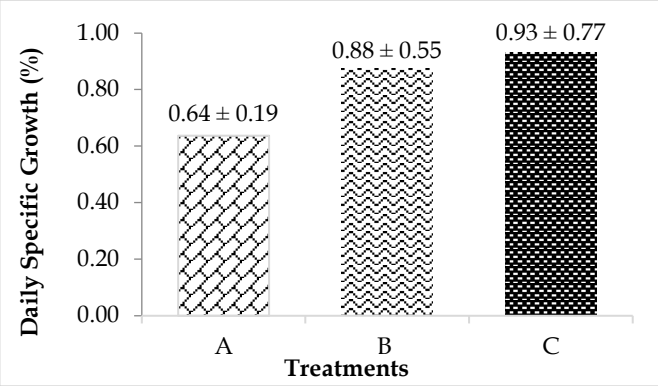

Figure 1. Daily specific growth rate of snakehead fish (*Channa Striata*) fry

Besides that the highest daily specific growth rate was supported by a high feed response or appetite during the study. This statement is in line with Susatyo et al. (2016) that the amount of feed consumed affects the growth of fish. Where fish that consume more feed will get nutrients such as sufficient protein for growth.

Growth is caused by excess energy and material derived from feed. This is supported by the statement of Sastra et al. (2020) when energy intake is reduced then protein is broken down by the body to be used as an energy source so that growth is hampered because the main function of protein is to form new cells. Growth is influenced by several internal and external factors. The internal factors include heredity, resistance to disease and the ability to utilize food with external factors including physical properties, chemistry and aquatic biology (Syafirah et al., 2021).

Furthermore, the daily growth rate in nilem fish meal treatment was low (0.64%) due to decreased feed response. This can be seen during the maintenance period where the feed given is not consumed properly by snakehead fish seeds. The low feed consumed causes the availability of nutrients to support metabolism in the body to be limited and has an impact on decreased growth. It is important to provide feed that is close to or in accordance with eating habits so that it can support growth.

Snakehead Fish (Channa Striata) Albumin Levels

The results of measuring the initial albumin levels of snakehead fish were 4.20%. Furthermore, the final albumin level in each treatment (nilem fish, golden snail and earthworms) sequentially was 4.49%. 4.80% and 5.10%. This shows that the albumin level in the group of fish that were fed treatment C (Earthworms) was higher than the other treatments.

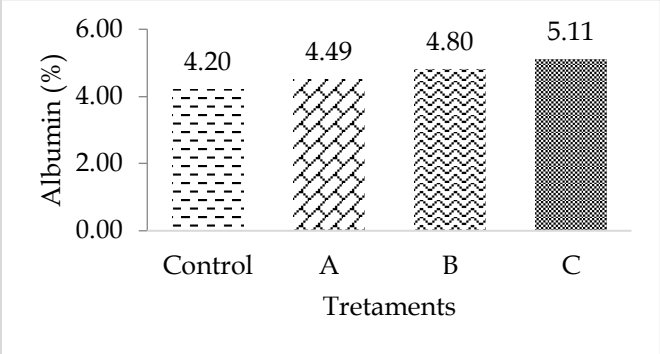

Figure 2. Albumin levels of snakehead fish (*Channa Striata*) with different feed ingredients

One of the compounds found in the snakehead fish body is albumin. Albumin levels in the treatment of earthworm flour (5.10%) compared to the treatment of feed made from golden snail flour and nilem fish meal. This is thought to be caused by the appropriate protein content for the needs of snakehead fish seeds and the amount of feed consumed from fish treated with earthworm flour is higher than the other treatments so that the source of animal protein in earthworms can be utilized by the body to trigger high levels of albumin. Consumption of feed that functions to provide high opportunities for nutrients to enter the body. This condition causes the amino acid measurement of albumin compounds to enter the body to increase. This is in line with what was stated by Wada et al. (2017) that amino acids play a very close role with albumin synthesis in tissues. Albumin synthesis is influenced by the main factor, namely the type of amino acid that enters the body through food, namely tryptophan, arginine, ornithine, lysine, phenylamine, threonine and proline (Wei et al., 2021). Further reported by Istiqomah et al. (2009) that the mino acid composition of earthworm flour includes the amino acid methionine (0.16%), lysine (0.08%), aspartate (2.59%), glutamate (0.67%), leucine (0.05%), isoleucine (0.01), serine (3.42%) and asparagine (0.05%). The different thing from the treatment of golden snail flour and nilem fish meal is because of the low feed consumption. So that the intake of amino acids for albumin accretion is lower.

Blood Glucose Level of Snakehead Fish (Channa Striata)

The results of measuring the glucose levels of the treated fish at 0 hours (before the fish ate) with the lowest glucose level of 52 mg/dL and increased 2 hours after the fish consumed the feed and then decreased after the next 2 hours (4 hours after consuming the feed).

Blood glucose levels 2 hours after the fish consumed feed for treatment B (Golden snail) was 112

mg/dL, followed by treatment A (Nilem fish) of 105 and the lowest was in treatment C (Earthworms) of 102 mg/dL)

Furthermore, blood glucose levels 4 hours after the fish consumed the feed showed the highest value in treatment A (nilem fish), namely 92 mg/dL, followed by treatment B (the golden snail) which was 58 mg/dL and the lowest was in treatment C (Earthworms) which was 38 mg/dL (Figure 3).

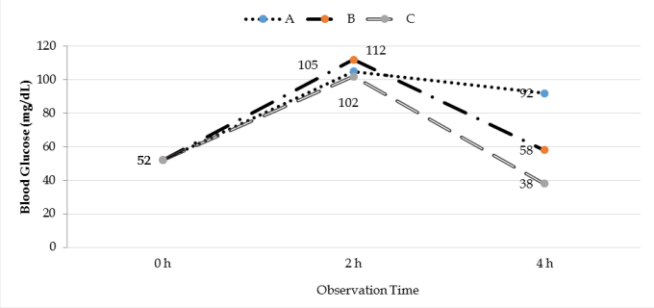

Figure 3. Glucose levels of snakehead fish (*Channa Striata*)

Blood glucose levels were measured three times, namely at 0 o'clock (before the fish eat). Then 2 hours (after the fish eat until they are full) and 4 hours (after the fish eat until they are full). This is done to determine changes in fish blood glucose levels every hour after eating.

Glucose levels for the 0 hour treatment (before the fish ate) were considered the same, namely 52 mg/dL. This is because the measurement of blood glucose (0 hours) is carried out when the digestive tract is empty (48 hours of fasting) so that there is no longer a supply of glucose from the feed. Blood glucose enters the body through foods that contain carbohydrates. Glucose is the most important monosaccharide that can be absorbed by the body to produce energy (Blanco, 2017).

In the second hour all treatments experienced an increase in glucose levels compared to the hour before the fish ate. The highest glucose level was found in treatment B (112 mg/dl), followed by treatment A (105 mg/dl) and the lowest in treatment C (102 mg/dl). This is supported by Blanco (2017) that glucose levels vary throughout the day which will increase after eating and return to normal within 2 hours.

At the fourth hour, all treatments decreased where treatment A (92 mg/dL). Then followed by treatment B (58 mg/dl) and finally in treatment C (38 mg/dL). Blood glucose decreased in the 4th hour after the fish ate because glucose had been used as an energy source. And what is left is a reserve for the body of the fish. This is in line with the statement of Guo et al. (2012), namely the decrease in glucose levels due to the process of glycolysis of blood cells. The process of glycolysis that occurs in cells begins with the formation of glucose molecules and ends with the formation of pyruvic acid. Where the formation of pyruvic acid in the process of glycolysis requires 2 ATP molecules which are used to transfer phosphate groups to glucose so that glucose has a higher energy store. The energy is used for the next reaction, namely the energy release reaction. The availability of glucose in the body indicates that the opportunity to use protein as an energy source can be minimized.

Conclusion

The best daily growth was obtained by using feed containing animal protein sources from earthworm flour of 1.85 g. The albumin level of snakehead fish at the end of rearing was highest in the treatment using feed made from earthworm flour (5.106%). Followed by golden snail flour (4.802%) and nilem fish meal (4.492%). Snakehead fish blood glucose levels 2 hours after consuming the feed increased in all treatments with the highest value in the treatment of golden snail flour and earthworms (112 mg/dL). Followed by nilem fish meal (105 mg/dL). However blood glucose levels 4 hours after consuming the feed decreased in all treatments with the highest reduction occurring in the treatment using earthworm flour (38 mg/dL). Followed by golden snail flour (58 mg/dL) and nilem fish meal (92 mg/dL).

Acknowledgments
The authors wish to acknowledge Universitas Tadulako for their assistance in facilitating this research project by granting access to their laboratory facilities, which were crucial in the collection and analysis of the data presented in this paper.

Author Contributions
NS and MS collects data and provide research facilities, UR processes and analyzes data, RAI Compiles and prepares articles.

Funding
The researchers involved in this endeavor have undertaken the responsibility of funding the project themselves. Without the support of any external sources, the researchers have committed their own resources, time, and effort to carry out this research.

Conflicts of Interest
The authors declare no conflict of interest

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