

Effect of Additional Aloe Vera (*Aloe vera*) on Artificial Feeds to Blood Cell Profile and Growth of Milkfish Seed (*Chanos chanos*)

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Abstract

Problems in cultivating milkfish are disease outbreaks and the high cost of feed. Therefore, a breakthrough is needed to overcome this problem by adding medicinal plants, namely Aloe vera, to the feed. This study aims at determining the effect of adding Aloe vera to artificial feed on the blood cell profile and growth of milkfish seed. The research method used was a complete randomized design with four (4) treatments and three (3) replications. The treatments on the test feed used were: A, (giving Aloe vera as much as 0 g/kg feed), B (giving Aloe vera as much as 5 g/kg feed), C (giving Aloe vera as much as 10 g/kg feed), and D (giving Aloe vera as much as 15 g/kg feed). The results showed that blood glucose values were still within the normal range. The lowest value in treatment was A, which was 42 mg/dL, and the highest in treatment was D, which was 66 mg/dL. The lowest Hb value was found in treatment A (5.1 g/dL), and the highest was 7.2 g/dL, found in treatment D. The highest average growth rate was obtained in treatment D with increasing biomass of 3.81 g, while the lowest growth was obtained in treatment A which resulted in average growth of 2.06 g. Water quality during the study was suitable for milkfish rearing media, namely temperature of 27 - 31°C, pH of 7.2 - 7.8, and salinity of 25 - 27 ppt.

INTRODUCTION

Milkfish (*Chanos chanos*) is one of the most widely cultivated fish commodities in Indonesia. Milkfish has a substantial economic value because of high consumer demand from year to year. Milkfish is a commodity that always experiences an increase in production every year. In 2014, milkfish production in Indonesia increased by 10.4% per year, namely 621,393 tons compared to the previous year in 2010, which was only 421,757 tons (Chilmawati *et al.*, 2018).

Constraints in milkfish cultivation are disease outbreaks and the high cost of

feed. The presence of disease in fish farming causes extensive losses. The fish's blood cell image can determine the health condition of the fish. Hastuti and Subandiyo (2015) argue that blood chemistry descriptions can be used to determine fish health. The fish blood cells' image includes blood glucose and Hb (hemoglobin). The cost of feed in fish farming can reach 75% of the total expense required (Yunaidi *et al.*, 2019). The dependence on imported feed ingredients leads to high prices and causes high production costs.

The condition of fish health and quality of feed strongly influence fish growth. The provision of quality feed is expected not only to increase fish growth but also to support the body's resistance to disease. Fish growth and fish survival rate are essential to support the increasing market demand. In addition, to reduce production costs (especially in feeds), a breakthrough is needed. Adding medicinal plants, such as Aloe vera (*Aloe vera*), is an alternative expected to increase fish immunity while reducing feed costs and supporting the survival rate of milkfish.

Aloe vera contains compounds that can provide benefits for pharmacological activity (Sitepu *et al.*, 2021). Kasiri *et al.* (2011) state that adding herbal plants or their extracts to feed can increase growth and protect fish from disease attacks during cultivation. Some contents of Aloe vera, such as amino acids, minerals, and vitamins, can help increase the growth of fish. Therefore, it is necessary to conduct a study on adding Aloe vera to the blood cells' image, growth, and also survival rate of milkfish seeds.

METHODOLOGY

Place and Time

This study was conducted from May 24th to July 22nd, 2021, and was located at the Brackish and Sea Water Laboratory, Faculty of Fisheries, Pekalongan University.

Research Materials

In this study, various equipment was used including refractometers (RHS 10 ATC, China), dissolved oxygen meter (Lutron PDO-519, Taiwan), digital thermometer (MC-245, Japan), pH meter (Waterproof EC-PCS Testr20, USA), digital weight scale (FSR-B 1200 gr X 0,01 gr, FUJITSU, Japan), disposable syringe 3 ml (OneMed, Indonesia), Hb and blood glucose meter (Digital EasyTouch GCHb ET-321, Taiwan), and 12 plastic jars with a volume of 10 L.

The materials used were milkfish seeds (3 cm length) obtained from local

fish farming in Pekalongan city and carried out for 35 days, commercial pellets, and *Aloe vera* flour.

Research Design

The study used a complete randomized design (CRD) with four treatments and three replications. The treatment doses used were A (0 g), B (5 g), C (10 g), and D (15 g). The dose used in this study was determined following Yunus (2021) with the best results at a dosage of 5 g/kg of feed on the effect of adding Aloe vera on the hematology of tilapia.

Work Procedure

Container and Fish Preparation

The media container used for research was 10 L jars, and each jar was filled with 5 L of brackish water and five milkfish seeds. The jars were sterilized by washing them first, then rinsing them clean before being used. The fish sample used was 3 cm milkfish seeds.

Test Feed Preparation

Mixing Aloe vera flour in the test feed was carried out by dissolving it into 100 ml of distilled water for each treatment. Then it was put into a spray bottle. Aloe vera flour that has been dissolved was gradually sprayed on the test feed and stirred until they were homogeneous. Making Aloe vera flour was following this method: (1) cleaning Aloe vera by washing it thoroughly with water, (2) then slicing Aloe vera thinly, (3) putting the Aloe vera slices in the oven, and (4) then drying them in the sun for 1-3 days (Sari *et al.*, 2012).

Maintenance

Seran *et al.* (2020) state that the feeding volume should be 5% of fish biomass per day. The feeding time was divided into three times per day, namely at 06.00, 12.00, and 16.00 WIB. Water quality control was carried out once a week by siphoning at the bottom of the jar and changing the water by 50%.

Parameters

Blood Cell Profile

The blood cell profile in this study is described as glucose level and Hb level. Fish blood was obtained by cutting the caudal part of the fish. Then, it was tested using a Digital EasyTouch GCHb Model ET-321 (Taiwan) device. Fish blood was attached to different glucose and Hb strips and then inserted into the EasyTouch device. The results of glucose and Hb measurements will be displayed on the screen after waiting a few seconds.

Biomass Growth

Calculation of biomass growth using the formula by Effendie (1997), namely:

$$W = W_t - W_o$$

Where:

W = absolute weight growth (g)

W_t = final average weight (g)

W_o = initial average weight (g)

Survival Rate

The formula used to calculate the survival rate calculated used Effendie's (1997) formula:

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

Where:

SR = survival rate (%)

N_t = final number of fish

N_o = initial number of fish

Water Quality

Observations of water quality measured during the study were temperature, salinity, and pH. Measuring

temperature was using a thermometer, measuring salinity was using a refractometer, and measuring pH was using a pH meter.

Data Analysis

The data obtained from the observations are the blood picture in the form of glucose and Hb values, the growth of milkfish seeds, and their survival. Growth data were analyzed by statistical tests to answer the hypotheses. Previously, the growth data were for normality and homogeneity. The normality test was carried out using the Lilliefors test (Nasoetion and Barizi, 1983) to find out if the data of growth was normally distributed. The homogeneity test used the Bartlett test (Sudjana, 1996). This analysis was to determine whether the data were homogeneous or not. If the data obtained were normal and homogeneous, then they tested with analysis of variance (ANOVA) to determine the difference in each treatment on growth. The level of glucose and Hb of fish blood were analyzed descriptively.

RESULTS AND DISCUSSION

Blood Cell Profile

Based on the data in Table 1, it can be seen that blood glucose is still in the normal range for all treatments, 42-66 mg/dL. The Hb level of milkfish is also still in the normal category, 5.1 – 7.2 (g/dL). Data on fish glucose and Hb levels during this study are presented in Table 1.

Table 1. Data of glucose and Hb level.

Number	Treatment							
	A		B		C		D	
	Glucose (mg/dL)	Hb (g/dL)	Glucose (mg/dL)	Hb (g/dL)	Glucose (mg/dL)	Hb (g/dL)	Glucose (mg/dL)	Hb (g/dL)
1	43	5.2	54	6.2	60	6.5	69	7.1
2	41	4.8	56	5.6	57	6.2	65	7.4
3	42	5.3	55	5.9	57	6.1	64	7.1
Total	126	15.3	165	17.7	174	18.9	198	21.6
Average	42	5.1	55	5.9	58	6.3	66	7.2

Blood glucose levels showed an increase from treatment A to D. It indicates that adding Aloe vera affects

blood glucose levels, even though the results are still in the normal range. Normal fish blood glucose levels contain

40-90 mg/dL (Nasichah *et al.*, 2016). Glucose is a source of energy that will be used in the internal processes of the fish body. The increase in glucose along with the increase in the Aloe vera dosage shows that it stimulates an increase in the body's metabolic processes to deal with stress, poor water quality, or disease infection. According to Affandi *et al.* (2012), energy reserves in the form of glycogen in the muscles and liver, and other organs are converted into glucose to strengthen the body against stress.

Hemoglobin or Hb level shows an increase following the increase in dosage. Although there was an increase, the Hb value was still within the normal range. It refers to the hemoglobin level of the carp, which ranges from 4.9-9.65 g/dL (Kusrini *et al.*, 2019). Hb has an essential role in the circulatory system of the fish body, as a carrier of oxygen. The increase in Hb levels in this study was caused by the presence of flavonoids in Aloe vera helping

to improve the blood condition of fish. This is in line with Wahjuningrum *et al.* (2008) stating that flavonoid activity will improve the performance of blood-producing organs, so that blood production increases. A low Hb value will cause disturbances in oxygen transport. Furthermore, Yanuhar *et al.* (2021) argue that the decrease in Hb indicates an abnormality in fish health. It will have an impact on the low oxygen content in the blood.

Biomass Growth

The results of observations of the average growth of milkfish are presented in Table 2, which shows that the growth of milkfish biomass with the highest value is with an average value of 3.81 g obtained in treatment D. Meanwhile, the milkfish biomass growth obtaining the lowest value is with an average value of 2.06 g obtained in treatment A.

Table 2. Data of growth.

Number	Treatment				Total
	A	B	C	D	
1	2.03	2.44	3.39	3.78	
2	2.19	2.73	3.04	3.65	
3	1.96	2.41	3.15	3.99	
Total	6.18	7.58	9.58	11.42	34.74
Average	2.06	2.53	3.19	3.81	

The results of the normality test obtained from the average absolute biomass growth during the 30-day study showed that the data were normally distributed. Then the homogeneity test was conducted, and produced homogeneous data; therefore, the data

could be analyzed for variance (ANOVA). Based on the analysis of variance, F count $44.5064 > F$ table 1%, which is 7.59. It means that each treatment showed a very significant difference in the effect on the growth of milkfish.

Table 3. Data of the analysis of variance.

Source of Variance	df	JK	KT	F Count	F Table	
					5%	1%
Treatments	3	5.2473	1.7491	44.5064**	4.07	7.59
Error	8	0.3144	0.0393			
Total	11	5.5617				

There are internal and external factors that can increase fish growth. Internal factors are usually aspects that

are difficult to control in the form of heredity (genetic), age, and gender. Meanwhile, external factors are feed and

water quality (Habibullah *et al.*, 2015). Feed is an important thing in the process of size growth, both length and weight of fish. External factors including feed are very influential on the growth and survival of fish. Fish growth is affected by the nutrients contained in the feed. Therefore, before giving feed to fish, the adjustment of the nutritional content in the feed must be appropriate. Manik and Arleston (2021) explained that the nutrients that fish need, such as vitamins, minerals, fats, carbohydrates, and proteins, must be present in the feed and follow the needs of the fish.

Nutrition is one of the factors that affect the growth of fish. An adequate number of nutrients in the feed does not only serve to provide energy for metabolic activities in the fish's body but is also used to meet the needs of milkfish to grow (Yolanda *et al.*, 2013). Fulfilling nutritional needs is a factor that determines the growth of fish. Milkfish requires a minimum of 15% – 30% protein (Hadijah *et al.*, 2017). Aloe vera is a natural ingredient in which there are useful nutrients for increasing fish growth. Sitepu *et al.* (2021) stated that Aloe vera contained vitamins, calcium, phosphorus, minerals, iron, potassium, copper, magnesium, sodium, zinc food, as well as enzymes and amino acids. It provides 20 amino acids out of seven amino acids lysine. One of the functions of amino acids is a growth activator. Tantri *et al.* (2019) state that amino acids function to accelerate, so that they can reduce the production period in aquatic animal cultivation activities. The vitamins in Aloe vera can also help increase the fish's appetite and the growth process. This is in line with Amalia *et al.* (2013) who argued that vitamins act as catalysts in the metabolic process for the growth process.

From the results of observations carried out for 30 days, it seems that the growth of milkfish seeds showing the highest growth occurred in treatment D, which was the largest 3.81 g at 81 g because the percentage of adding 15 g/kg Aloe vera flour to the feed was very

influential on growth. Treatment C was added with 3.19 g, with Aloe vera flour added to the feed at around 10 g/kg feed. In treatment B, it is 2.53 g with the addition of Aloe vera flour 5 g/kg feed. Lastly, treatment A is the lowest, 2.06 g, because there was no additional Aloe vera flour here. It indicates that the different percentage of adding Aloe vera flour in artificial feed affects the growth of milkfish seeds. This is because Aloe vera has a complete nutritional content.

The highest dosage of Aloe vera flour was in treatment D, which was 15 g/kg of feed; therefore, it obtained biomass development with an average value of 3.81 g. This is because the dosage of adding Aloe vera flour to the feed can accelerate the increase in the growth of milkfish. Aloe vera contains amino acids, vitamins, and minerals that play an essential role in increasing the growth of milkfish. According to Pratama *et al.* (2019), fish need a balanced composition of essential and non-essential amino acids to support adequate growth. Prasetio *et al.* (2018) stated that the amino acid in Aloe vera is an ingredient that functions as an energy source and as an ingredient to help repair and grow organisms.

The vitamin content in Aloe vera also functions to optimize fish appetite and can be used for growth. This agrees with Yuniati and Almasyhuri (2012) who argue that vitamins can help in the process of metabolism and can accelerate the absorption of nutrients, which is useful for growth. The mineral content found in Aloe vera such as calcium and phosphorus play a role in increasing growth in fish. Other ingredients such as phosphorus and calcium are used to form and maintain bone tissue. This reinforces Pramitha (2018) stating that one of the functions of phosphorus is for bone formation. In addition, according to Lestari *et al.* (2016), calcium is needed for bone formation and to trigger growth.

The fat content in Aloe vera is also very useful for fish energy sources in carrying out metabolism in the body. According to Siregar and Makmur (2020),

fat is needed by the body for a direct and potentially efficient energy source. Iron (Fe) functions in forming minerals, hemoglobin, and enzymes (Diastari, 2019). Therefore, the presence of iron in feed helps the process of survival and growth in fish.

Aloe vera flour given in treatment C was 10 g/kg of feed with the result that the average growth value of total biomass weight was 3.19 g. It is then followed by treatment B by giving Aloe vera flour 5 g/kg of feed with the results of the average growth value, and the total weight of the biomass was 2.53 g. Treatment C and B obtained a smaller value than treatment D. The results of low biomass weight in treatments C and B were due to the lower doses allocated than in treatment D as the

substances present in Aloe vera flour did not have a maximum effect on the growth of milkfish seeds.

The Aloe vera flour added in treatment A was 0 g/kg of feed with the result that the average value of biomass growth was 2.06 g. In treatment A, the addition of biomass weight was the lowest compared to other treatments. It is supported by Prasetio *et al.* (2018) that, if the feed is consumed without the addition of Aloe vera powder in it, the growth of fish will be lower.

Survival Rate

The survival rate during the study is presented in Table 4.

Table 4. Data of survival rate.

Number	Treatment			
	A	B	C	D
1	5	5	5	5
2	5	5	5	5
3	5	5	5	5
Total	15	15	15	15
SR (%)	100	100	100	100

The survival rate is the percentage of organisms that live at the end of maintenance (Setiawati *et al.*, 2013). The Aloe vera flour given in the feed during maintenance from beginning to end did not affect the survival of milkfish. All treatments obtained 100% survival results. Several factors affect the survival of fish, namely internal and external factors, which caused the survival of milkfish fry to reach 100% in this study. Internal factors are factors of the individual fish, and external factors are influenced by the carrying capacity of the environment, water quality, and feed quality. The aspects of environmental quality and feed quality and quantity

support fish to live. This statement is supported by Prasetio *et al.* (2018), that the quality of the environment and the quality and quantity of feed support, will affect the high percentage of fish survival levels. According to Afriyanto *et al.* (2015), environmental aspects are crucial because the resistance of organisms will increase if the environment is good, while if the environment is not good, it will result in a decrease in fish resistance.

Water Quality

The value of water quality is presented in Table 5.

Table 5. Value of water quality.

Parameter	Observed Value	Standard Value	Reference
Temperature (°C)	27–30	27–30	Safitri <i>et al.</i> (2020)
pH	7,2–7,8	6,5–8,5	Safitri <i>et al.</i> (2020)
Salinity (ppt)	25 – 27	< 40	Safitri <i>et al.</i> (2020)

Water as a living medium for fish has an essential role in supporting the life of aquatic biota, especially fish. Therefore, water quality is a matter of concern in fish farming activities. Water quality that does not meet the requirements can cause fish to experience stress, which can eventually lead to death (Sholekhuddin *et al.*, 2019). Research activities in observing water quality parameters observed include temperature, pH, and salinity.

The results of observations in the study during 30 days of maintenance show that the temperature parameter values ranged from 27-30 °C. Safitri *et al.* (2020) stated that 27-30 °C is the optimal temperature for milkfish. Therefore, the yield data at temperatures ranging from 27-30 °C on the maintenance media are still safe and feasible for the survival of milkfish seeds.

The next parameter observed was pH ranging from 7.2 to 7.8. Safitri *et al.* (2020) said that milkfish live and grow in the pH range of 6.5–8.5. The pH data show that the range between 7.2-7.8 is still feasible for milkfish cultivation activities. Parameter data for salinity water quality ranged from 25–27 ppt for 30 days. Milkfish live and grow in a salinity range of <40 ppt. The data from the observations show the salinity range that is still feasible for milkfish cultivation activities.

CONCLUSION

The addition of Aloe vera flour with a difference in the percentage of doses in artificial feed has a very significant effect on the growth of milkfish seeds. The best growth level was in treatment D with a dose of 15 g/kg of feed and an average biomass value of 3.81 g. Adding Aloe vera with different dosages affects the milkfish's blood glucose and hemoglobin level. It is recommended to conduct further research on Aloe vera flour doses higher than 15 g/kg feed to find the optimal dose for milkfish growth.

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