

The Effect of Use of Commercial Probiotics in Feed on Water Quality and Growth Performance of Bawal Fish (*Colossoma Macropomum*) Bioflok System

M. Ihsan Kamil^{1*}, Maya Istyadji¹, Yasmine Khairunnisa¹

¹Department of Natural Science Education, Universitas Lambung Mangkurat, Banjarmasin, Indonesia

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*Corresponding author: M. Ihsan Kamil

Department of Natural Science Education, Universitas Lambung Mangkurat, Banjarmasin, Indonesia

Abstract

Bioflok consists of special substances called probiotics, where probiotics contain beneficial microorganisms in cultivation activities. Probiotics can be given through feed and water (media). Probiotics can break down metabolic waste and stimulate an immune response so that fish health increases and affects growth. Probiotics have an effect on the growth of fish, but there is no information for freshwater pomfret. This study aims to determine the effect of probiotic types on bioflok volume, water quality, and fish growth in pomfret culture (*Colossoma macropomum*) bioflok system. The method used was giving different types of probiotics, treatment A control, B EM4, C Probiotic-7, D Aquaenzym. The results of the re-search showed that the lowest pH of the system was 7.85, namely the pH of the probiotic-7 type. The mean floc growth measurement results for treatment A was 12.93, treatment B was 17.54, treatment C was 17.59, treatment D was 15.98. Aquaenzym probiotic has a fish survival rate of 100% compared to other probiotics. The type of probiotic that has the best results in increasing the growth performance of freshwater pomfret (*Colossoma macropomum*) is probiotic-7.

Keywords: Probiotics, bioflok, pomfret, water quality, flock volume, growth.

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INTRODUCTION

Freshwater pomfret aquaculture commodity has prospects for rapid development. Today, the freshwater pomfret (*Colossoma macropomum*) has a very high economic value in food commodities. The development of various products from freshwater pomfret is one of the triggers for the high demand for this fish. So that cultivation prospects also need to be developed to meet this need.

The bioflok system is a system that refers to the use of microorganisms in decomposing complex compounds in fish (water) living media (Hadijah *et al.*, 2022). According to Hanafie *et al.*, (2021); and Julpano *et al.*, (2021) bioflok technology is able to utilize the metabolic results of fish that contain nitrogen which is converted into protein. Bioflok has special features compared to conventional cultivation, namely, maximizing limited land, environmentally friendly, saving water and feed use, odorless and waste water can be used for plant fertilizer due to the use of probiotics

and probiotics with microorganisms such as bacteria (Dara *et al.*, 2022).

Bioflok consists of special substances called probiotics, where probiotics contain beneficial microorganisms in cultivation activities. Probiotics can be given through feed and water (media). According to Talib and Noh, (2019) that the use of probiotics through water as bioremediation can improve water quality and decompose organic matter. Meanwhile, giving probiotics through feed can help increase feed conversion.

Basically, probiotics are easy to find commercially or processed by local people, but their use in South Kalimantan, especially Banjarmasin, is still very limited. Currently there is still a lack of research on the innovation of making and using various types of probiotics that can increase the growth rate and survival of fish, so it is necessary to test various types of commercial probiotics that are appropriate for the

growth performance of freshwater pomfret and as an alternative to probiotics in the biofloc system.

RESEARCH METHODS

The population in this study was the sum of all pomfret fish used in this cultivation research. The number of fish consisted of 10 fish in each repetition so that there were 120 freshwater pomfret studied. The sample used in the study was taken from part of the population in each formulation.

In this study, a real pilot study was conducted with the aim of seeing an effect of treatment on variables. Research is done by doing a treatment of the

object to be studied. The technique in the study used random probiotics in 4 different treatments with 3 repetitions.

$(t-1)(r-1) \geq 15$

t = Treatment / number of treatments

r = replications / number of repetitions

Using the above equation, it is obtained that the repetition of each treatment is 3 times. The trial design in the study can be seen as follows:

In this treatment group, different types of probiotics were given with the formulations in the table below:

Table 1: Research Design

Probiotic Type			
Control Non-probiotic	Treatments 1 EM4	Treatments 1 Probiotic-7	Treatments 1 Aquaenzym
Repetitions 1	Repetitions 1	Repetitions 1	Repetitions 1
Repetitions 2	Repetitions 2	Repetitions 2	Repetitions 2
Repetitions 3	Repetitions 3	Repetitions 3	Repetitions 3

The sample size in the growth rate test is all fish used as test subjects described in the table below.

Table 2: Research Sample Size

Treatment	Sample size	Number of samples
T 1	overall sample results	120 fish
T2	overall sample results	120 fish
T3	overall sample results	120 fish
T4	overall sample results	120 fish

Observation of biofloc volume is carried out every 3 days by measuring the biofloc sedimentation volume formed using the INHOFF cone measuring instrument and then matching it with the standard from Sucipto, 2020, which is 60-80 ml/liter. Observation of water quality is carried out once a week, in the morning and evening. The water quality parameters observed were water temperature, dissolved oxygen (DO) content, degree of acidity (pH). Observations of water temperature and pH used a pH meter while DO observations used a DO meter.

The survival rate is the percentage of fish survival during the rearing period which can be calculated using the formula Zonneveld *et al.*, (1991).

$$SR = \frac{Nt}{No} \times 100\%$$

SR = Fish survival rate (%)

No = Number of fish living at the beginning (tails)

Nt = Number of live fish at the end (tails)

Feed conversion ratio (FCR) can be calculated using the formula Effendi (1997):

$$FCR = \frac{Pa}{Bt - Bo + Bm}$$

FCR = Feed Conversion Ratio

Pa = Total weight of feed given (g)

Bt = Final weight of fish (g)

Bo = Fish initial weight (g)

Bm = Dead fish weight (g)

Absolute length growth is used to calculate the increase in fish length during fish rearing using the formula Zonneveld *et al.*, (1991) as follows:

$Lm = TL1 - TL0$

TL1 = total length at the end of maintenance (cm)

TL0 = total length at the start of maintenance (cm)

Lm = absolute length growth (cm)

Calculation of fish body weight growth based on Effendi's formula (1979) is as follows:

$$SGR = \frac{\ln Wt - \ln Wo}{t} \times 100\%$$

SGR = Laju pertumbuhan harian (%)

Wt = Average individual weight at the end of the maintenance period (g)

Wo = Average individual weight at the beginning of the maintenance period (g)

t = Maintenance time (days)

Data analysis techniques carry out prerequisite tests, hypothesis tests and follow-up tests. The data

normality test was carried out using statistical analysis using SPSS 24. The data obtained included data on the number of fish deaths in 12 observations, tested for normality with the aim of seeing the distribution of the data. The normality test uses the Kolmogorov-Smirnov normality test with a significance level of 0.05. Decision making is based on the significance value of the Kolmogorov-Smirnov test if > 0.05 , the data distribution is declared normal. The next assumption test is a homogeneity test, which is used to see whether the variance of the data obtained is homogeneous or not. This study uses the homogeneity test with the Levene Test. The test uses the SPSS statistical software with a significance level of 0.05.

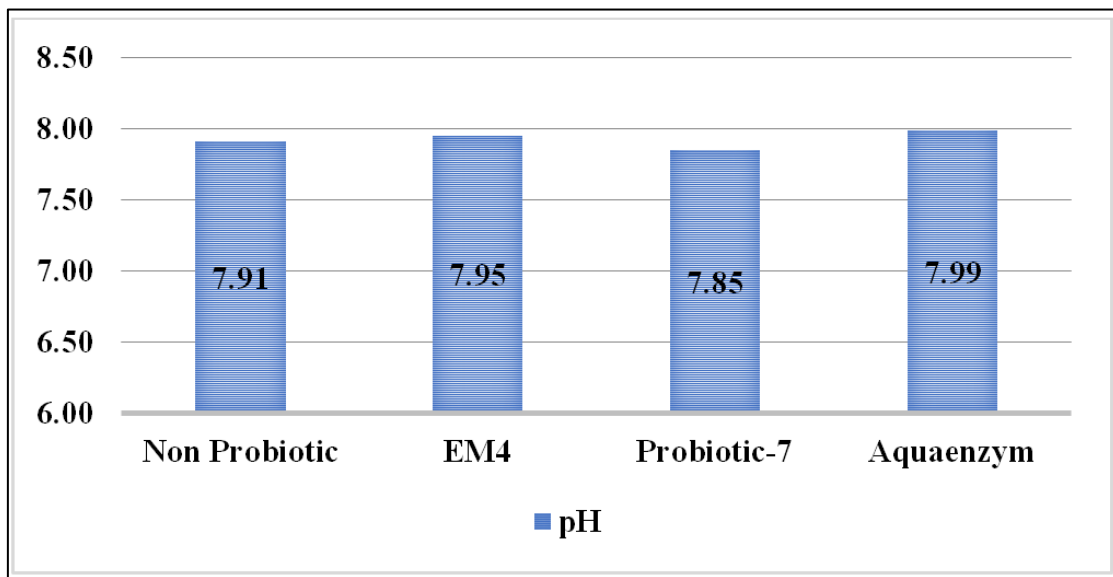
For normal and homogeneous data, the next test is an Anova test to see whether there is a significant difference from the data group we are testing. Anova test is calculated using SPSS statistics with the assumption that the data is normal and homogeneous. According to Alhanannasir and Sanjaya, (2018) to be able to find out the differences between samples in a research data can be known by using a follow-up test that compares the average value between

data groups with a multiple comparison test. The way that can be taken is by using the Tukey test, in making decisions Duncan's test uses two hypotheses to draw conclusions.

RESULTS AND DISCUSSION

Based on research conducted on freshwater pomfret rearing for 40 days using the biofloc system. Obtained research data included the effect of probiotic types on water quality, flock growth and growth performance of freshwater pomfret including indicators of survival, feed conversion ratio, absolute weight growth, absolute length growth, daily growth rate. The treatment was given depending on the type of probiotic, namely treatment without probiotics, EM4 probiotics, Probiotic-7 and Aquaenzym.

The quality of the water in the study was measured from the degree of acidity (pH), water temperature, and the amount of dissolved oxygen. The pH level of biofloc based on the type of probiotic is presented in the graph below:



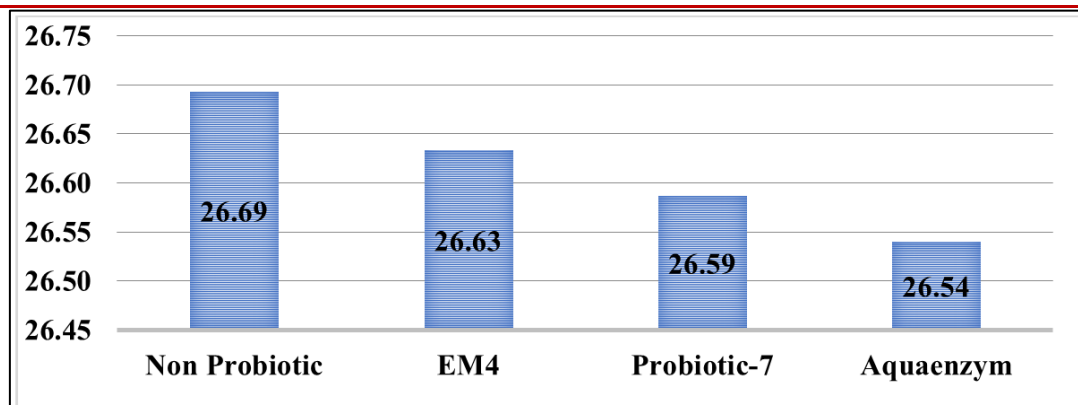
Picture 1: Daily water pH difference of the biofloc system

According to research by Smith *et al.*, (2018), the use of certain probiotics, such as *Lactobacillus acidophilus*, has been shown to have a significant effect on water pH. They reported that administering *L. acidophilus* in an aquaculture system resulted in a steady increase in the pH of the water, keeping the pH value within the optimal range for fish growth.

In the research results, the pH range of probiotics and ordinary water is from 7.85 to 7.99. The average pH value measurement in the biofloc system

did not have a pH difference of more than 1 point. Basically, the pH needs of freshwater pomfret range from 7-8.5. This confirms that the pH of the system formed is still within the pH range of pomfret freshwater fish. The lowest system pH is 7.85, which is the pH of the probiotic-7 type.

The results of temperature measurements on biofloc based on the type of probiotic are presented in the graph below:



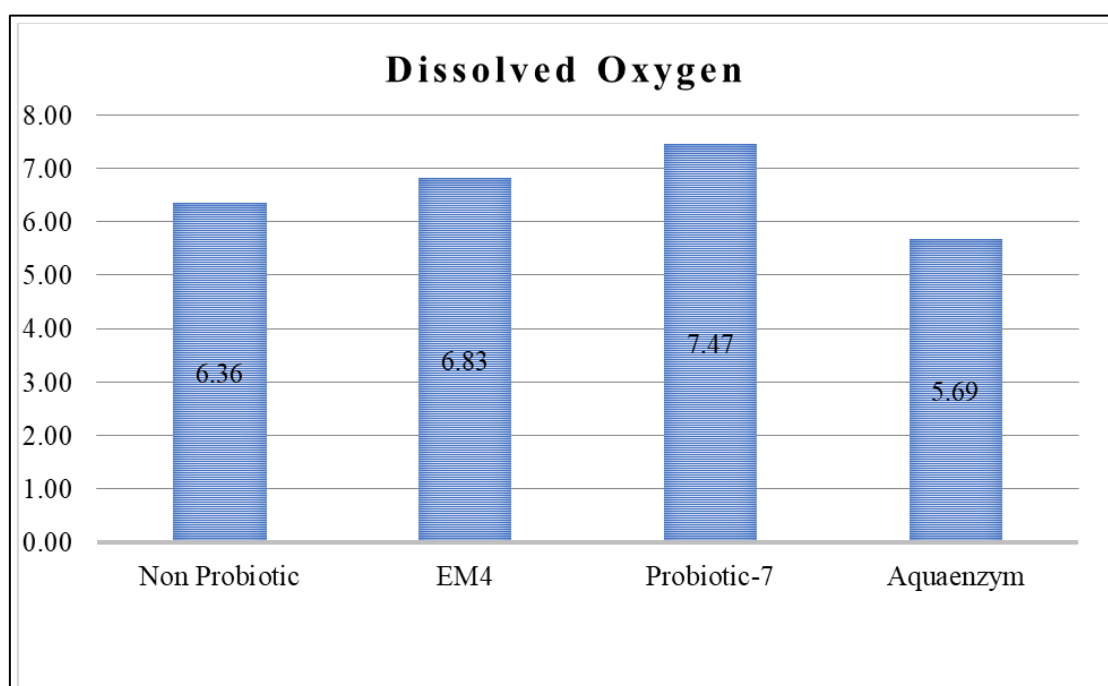
Picture 2: The difference in the mean daily temperature of the water

Water temperature in the results of research on different types of probiotics has a significant effect on temperature, where temperature is a major factor that affects the metabolic system of flocks. The system without the addition of probiotics has the highest temperature compared to the addition of probiotics. According to research conducted by Johnson *et al.*, (2019), administration of probiotics *Bacillus* sp. significantly affect the temperature of the water in the aquaculture system.

They found that the population of *Bacillus* sp. helps in regulating water temperature, reducing

excessive temperature fluctuations, and maintaining optimal temperature conditions for fish for good growth. Water temperature in the results of research on different types of probiotics has a significant effect on temperature, where temperature is a major factor that affects the metabolic system of flocs.

The results of measuring the amount of dissolved oxygen in the biofloc system based on the different types of probiotics are presented in the following graph:



Picture 3: The average difference in dissolved oxygen

Dissolved oxygen (DO) is an important indicator of water quality because it plays a role in the metabolism and health of aquatic organisms. According to research conducted by Nguyen *et al.*, (2020), the use of probiotics *Nitrosomonas* sp. significantly affect the concentration of dissolved oxygen in the aquarium.

They found that the presence of *Nitrosomonas* sp. increases DO concentration, because these microorganisms play a role in the nitrogen cycle and assist in the removal of harmful compounds such as ammonia. In the results of this study the system that had

the most dissolved oxygen was the floc system with the addition of probiotic type probiotic-7.

The measurement results of the growing flocs are calculated by the average value in the table below:

Table 3: Floc growth measurement results

REPETITION	Probiotic Type			
	Non-Probiotic	Em4	Pb-7	Aquaenzym
flock growth rate	12,93	17,54	17,59	15,98
DIFFERENCE VALUE	2,28 ± 3,45a	2,45 ± 3,98c	3,07 ± 3,95c	1,34 ± 3,87b

In this study, the probiotic that had a higher floc growth rate was probiotic-7. Probiotic product that has been specially developed for use in biofloc systems in aquaculture. This probiotic contains microorganisms *Lactoba-cillus acidophilus*, *Bifidobacterium bifidum*, *Lactobacillus plantarum*, *Streptococcus thermophilus*, *Saccharomyces cerevisiae*, *Bacillus subtilis*, *Pediococcus acidilactici*.

The study conducted by Wu *et al.*, (2017) evaluated the effect of two different types of probiotics on floc growth in a biofloc system. The floc cultivated with *Bacillus subtilis* has a higher density and larger size. This indicates that the type of probiotic *Bacillus subtilis* can increase the growth and density of floc more effectively in a biofloc system.

The measured freshwater pomfret growth performance is presented in the table below.

Table 4: Data Summary the results of research on the growth performance of Freshwater Pomfret

Probiotic type	Parameter				
	Survival Rate (%)	Feed Conversion Ratio (%)	Absolute Weight Growth (%)	Absolute Long Growth (%)	Daily Specific Growth Rate (%)
Non	100	3,12	20,2	5,2	0,8
Em4	98	10,89	22,6	5,7	0,8
Pb7	97	17,06	23,73	6,73	0,8
Aquaenzym	100	5,82	10,83	4,97	0,6

The data in the table above shows that probiotic-7 produces the lowest survival rates compared to other types of probiotics. In the results of this type of treatment without the addition of probiotics with the addition of aquaenzym probiotics, the fish survival rate was 100% compared to using EM4 and Probiotics7 probiotics.

Research by Smith *et al.*, (2018) found that administration of probiotics containing *Lactobacillus*

acidophilus to freshwater pomfret resulted in a significant increase in survival. This shows that certain types of probiotics can increase the survival rate of fish.

The feed conversion ratio is a parameter used to measure the efficiency of using feed in livestock production, including freshwater pomfret. The table below presents the results of calculating the feed conversion ratio for fresh water pomfret rearing

Table 5: Feed Conversion Ratio Calculation Results

Repetition	Probiotic Type			
	Non-Probiotic	Em4	Pb-7	Aquaenzym
FCR	3,12	10,89	17,06	5,82
Difference Value	3,12 ± 0,12a	10,89 ± 1,76c	17,06 ± 4,65d	5,82 ± 3,76b

Different letters indicate a real difference

Research by Johnson *et al.*, (2019) reported that the administration of probiotics containing *Bacillus subtilis* to freshwater pomfret resulted in an increase in the feed conversion ratio. A lower feed conversion ratio indicates better feed efficiency, where fish can use feed more effectively for their growth. These results indicate that the use of certain probiotics can increase the efficiency of feed utilization by freshwater pomfret.

Research by Rodriguez *et al.*, (2020) showed that administration of probiotics containing

Lactobacillus plantarum to freshwater pomfret resulted in significant growth in absolute weight. Absolute weight growth is an important parameter that describes the increase in overall body weight of fish during the study period.

Weight growth in fish is absolutely the weight gain of the fish during the rearing period. The data in the table below shows the absolute weight gain of freshwater pomfret for 40 days of rearing period.

Table 6: The absolute weight gain value of fish

Repetition	Probiotic Type			
	Non Probiotik	Em4	Pb-7	Aquaenzym
1	21,90	13,90	24,50	10,30
2	20,30	26,30	25,40	11,10
3	18,40	27,60	21,30	11,10
Average	20,20	22,60	23,73	10,83
Difference Value	3,46 ± 0,71a	5,69 ± 0,34b	4,80 ± 4,48a	6,02 ± 0,92b

Different letters indicate a real difference

In this study the best absolute weight growth was given by the probiotic-7 type. This is because this type of probiotic contains 7 microorganisms that can affect the biofloc system and water quality, and maintain fish health.

The daily growth rate of fish refers to the rate of increase in body weight or length in one day. This is a measure used to monitor the speed at which fish grow over time. In this study the daily growth rate of fish was measured using the weight gain of the fish. The daily growth rate of pomfret reared for 40 days is shown in the table below:

Table 7: Fish Daily Growth Rate

Repetition	Probiotic Type			
	Non-Probiotic	Em4	Pb-7	Aquaenzym
Daily growth rate	0,08	0,08	0,08	0,06
Difference Value	0,08 ± 0,01a	0,08 ± 0,01a	0,08 ± 0,01a	1,02 ± 0,01b

Different letters indicate a real difference

The daily growth rate of fish refers to the rate of increase in body weight or length in one day. This is a measure used to monitor the speed at which fish grow over time. In this study the fastest growth rate was in treatments 2 and 3 and it was proven that the use of probiotics had a significant effect on the daily specific growth rate of pomfret.

Research by Li *et al.*, (2016) found that administration of probiotics containing *Saccharomyces cerevisiae* to freshwater pomfret resulted in a significant increase in daily growth rate. Daily growth rate is a measure of the speed at which fish grow per day, reflecting a faster growth rate.

Based on the recapitulation of research data on pomfret growth performance, Aquaenzym probiotics had a 100% fish survival rate compared to other probiotics. However, for other parameters such as feed conversion ratio, absolute weight and length growth, as well as daily specific growth rate, Probiotic-7 type probiotics are superior compared to other probiotics.

CONCLUSION

Different types of probiotics have an effect on water temperature and dissolved oxygen in the biofloc system but have no significant effect on the pH value of the floc system. Different types of probiotics have an influence on floc growth with the type of probiotic with the greatest floc growth being the probiotic-7 type. Different types of probiotics have an influence on the growth performance of freshwater pomfret including survival rate, feed conversion ratio, absolute weight growth, absolute length growth, and daily growth rate

with the type of probiotic that has the best results is probiotic-7.

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