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THE EFFECT OF ADDING PETROFISH PROBIOTICS TO FEED AT DIFFERENT DOSES ON THE GROWTH AND SURVIVAL OF BARRAMUNDI (*Lates calcarifer*) JUVENILES

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ABSTRACT

This study aimed to determine the effect of adding Petrofish probiotics to feed at different doses on the growth and survival of barramundi (*Lates calcarifer*) juveniles. The research utilized an experimental method with 120 barramundi juveniles as test subjects, distributed into treatments A (Control), B 10mL/kg dosage), C (15mL/kg dosage), and D (20mL/kg dosage). The study was conducted for 30 days, using 12 buckets containing ten barramundi juveniles. The research employed a Completely Randomized Design (CRD) with Analysis of Variance (ANOVA), followed by Least Significant Difference (LSD) post hoc tests. The results indicated significant effects for each treatment. The highest absolute weight and length were observed in treatment D at 0.90 g and 0.73 cm, followed by treatment C at 0.77 g and 0.67 cm, treatment B at 0.69 g and 0.62 cm, and the lowest values were in treatment A at 9.51 g and 0.58 cm.

Keywords: Barramundi, Probiotik Petrofish, Survival Rate

1. INTRODUCTION

The barramundi (*Lates calcarifer*), better known as barramundi, is a type of marine fish commodity with a high economic value[1]. Cultivation of barramundi has become a commercial cultivation business that needs to be developed. This happens because it grows relatively quickly and has high adaptability to environmental changes[2].

One of the most critical factors in cultivation is feed. Feed generally consumes around 60-70% of the total production costs incurred in cultivation activities. This causes the importance of fish's feed to meet their energy needs to live and grow. Fish growth will increase if the feed provided can be digested well by the fish so that the energy obtained by the fish from the feed can be utilized optimally[3].

In the barramundi cultivation business, feed is always an obstacle to cultivating cultivation. This makes cultivation less profitable because only 25% of the feed provided is converted as production output, and the rest is wasted as waste[4]. Probiotics are live microorganisms in fish farming that can prevent disease, increase production, and reduce economic losses[5]. Probiotics have anti-microbial effects, which control pathogens in the digestive tract to help the food absorption process in fish digestion[6]. It is hoped that the provision of probiotics in this feed can help increase fish growth and improve fish survival so that the quality of barramundi fish rises in the form of an increase in weight per fish rearing time.

2. RESEARCH METHOD

Time and Place

This research was carried out in March - April 2023 for 30 days at the Takalar Brackish Water Aquaculture Fisheries Center (BPBAP), Mappakalompo

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Village, Galesong District, Takalar Regency, South Sulawesi Province.

Method

This research used a Completely Randomized Design (CRD) with four treatments and three replications. The treatment will be given by giving different doses of petrofish to barramundi seed feed. The requirements for this research are as follows: Control (A), dose 10 mL/kg feed (B), 15 mL/kg feed (C), and 20 mL/kg feed (D).

Procedure

Container Preparation

The containers used in this research were buckets with a volume of 50 L and 12 in total, equipped with hoses and aeration stones. The barramundi seeds used in this research were from BPBAP Takalar and numbered 120 fish.

Feed Preparation with the Addition of Probiotics

Preparation of probiotics in feed includes the process of mixing petrofish probiotics in feed, namely by taking probiotics according to the dose used, namely 10 mL/kg feed, 15 mL/kg feed, and 20 mL/kg feed, taking probiotics which will be put into a spray bottle using a tool. injection[5]. The bacteria in the probiotic will be activated by mixing each dose of petrofish with 100 mL of water and then shaking until homogeneous (mixed evenly). Then, spray the mixed probiotics evenly on the feed. After the pelleted feed is mixed with probiotics according to the dosage, the feed is air-dried for 24 hours.

Test Fish Maintenance

Fish were kept for 30 days. Feed barramundi juveniles three times a day, namely in the morning at 07.00, at 12.00, and afternoon at 17.00. The feed given to the fish is otohime pellets, amounting to 5% of the biomass weight. Sampling will occur weekly, including growth, survival, FCR, and EP. Then, for water quality, including pH, temperature, DO, and salinity.

Observed Parameters

Absolute Weight Growth

This parameter can be calculated using the formula according to Mulqan et al.[7] as follows:

$$Wm=Wt-Wo$$

Information:
Wm = Absolute weight growth (g)
Wt = Final average weight (g)
Wo = Initial average weight (g)

Absolute Length Growth

This parameter can be calculated using the formula according to Mulqan et al.[7] as follows:

$$Lm=Lt-Lo$$

Information:
Lm = Absolute length growth (cm)
Lt = Final maintenance length (cm)
Lo = Initial length of maintenance (cm)

Survival Rate (SR)

This parameter is calculated using the following formula:

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

Information:
SR = Survival rate (%)
No = Number of fish at the start of the study (fish)
Nt = Number of fish at the end of the study (fish)

Feed Conversion Rate

The feed conversion ratio is calculated according to Fahrizal & Nasir[8], as follows:

$$FCR=\frac{F}{Wt-Wo}$$

Information:
FCR : Feed Conversion Ratio
F : Total amount of feed given (g)
Wt : Total weight of fish at the end of the study (g)
Wo : Total weight of fish at the start of the study (g)

Feed Efficiency

Feed efficiency (EP) is calculated based on the following formula:

$$EP=\frac{Wt-Wo}{F}\times 100\%$$

Information :

EP : Feed Efficiency (%)
Wt : Average weight of fish at the end of the study (g)
W0 : Average weight of fish at the start of the study (g)
F : Total amount of fish feed given (g)

Water quality

Water quality measurements are carried out once a week when sampling fish. Sampling is carried out in each research container. The standard parameters measured for the growth of barramundi seeds are temperature, pH, DO, and salinity.

Data Analysis

Data processing analysis for each parameter was tested using One Way Analysis of Variance (ANOVA) to see the effect of petrofish on feed at different doses. If there is a difference, continue with the Least Significant Difference (BNT) test.

3. RESULT AND DISCUSSION

Absolute Weight Growth

Results of research on the effect of adding petrofish probiotics to feed at different doses on barramundi seeds produce different average weights of fish seeds. The graph of absolute weight growth can be seen in Figure 1.

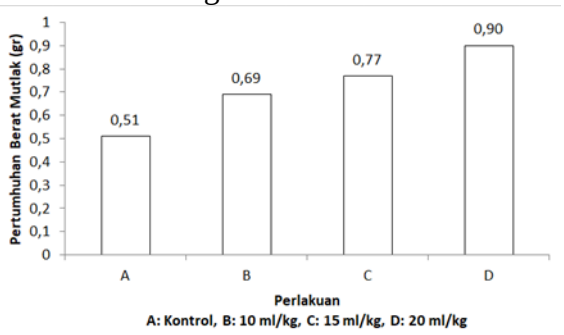

Figure 1. Absolute weight

Based on Figure 1, in treatment D with a dose of 20 mL/kg feed, the highest average absolute weight value was 0.90 g, and treatment A or control had the lowest value at 0.51 g. In the research of Gomes et al.[9], it was shown that the provision of probiotics in artificial feed influenced the weight growth of milkfish seeds, where the highest weight was in treatment D (15 mL/kg feed), namely 10.82 g, while the lowest weight in A (control), namely 7.73 g. Treatment D with a probiotic dose of 20 mL/kg feed showed the highest absolute growth in weight due to the activity of the probiotic bacteria *Lactobacillus* sp.

Bacteria *Lactobacillus* sp. will convert carbohydrates into lactic acid. Then, lactic acid can create a lower pH atmosphere. Lactic acid can produce a low pH in the substrate, causing an acidic atmosphere. *Lactobacillus* sp. can increase acidity in the substrate by 1.5–2.0%. In acidic conditions, *Lactobacillus* sp. can inhibit pathogenic bacteria and putrefactive bacteria.

The creation of acidic conditions in the intestine will increase the secretion of proteolytic enzymes in the digestive tract to break down proteins into amino acids, which the intestine will absorb. Food proteins are broken down into amino acids by proteolytic enzymes and peptidases in the digestive tract[6]. The low growth of white sea bass was found in treatment A compared to other treatments because the feed given was a feed that was not sprayed with probiotics, so there was no increase in digestive enzymes because the feed shown did not contain live microorganisms contained in probiotics so that the protein hydrolysis process was complex for into simple compounds does not occur in the digestive tract of fish.

Probiotic bacteria in the feed that enters the digestive tract can suppress pathogenic bacteria in the intestine, helping the food absorption process more quickly. Providing probiotics in feed affects the speed of feed fermentation, so it will help

the process of food absorption in fish digestion[10].

Apart from that, the microorganisms contained in petrofish probiotics can be wholly absorbed into the feed, increasing the performance of microflora in fish digestion and food absorption. The food the fish absorbs will become energy and be used for metabolic processes and growth to increase the fish's weight[4].

Bacillus sp. bacteria can associate in the digestive tract to increase nutrient absorption and produce amino and fatty acids. *Bacillus* sp. acts as a probiotic bacteria, quickly breaking down feed and suppressing pathogenic bacteria in the digestive tract. Enzymes produced by *Bacillus* sp. are protease enzymes. Protease enzymes are biocatalysts for protein breakdown reactions. Besides that, *Bacillus* sp. will affect the aroma and taste of the feed provided so that the fish will be stimulated to consume the feed[11]. Using probiotics in feed can reduce crude fiber and feed fat, making it easier for fish to absorb feed and easily absorbed by the body so that growth rate increases[12].

The mechanism of action of probiotics is that lactic acid bacteria in the fish intestines can balance digestive tract microbes, which can increase digestibility, converting carbohydrates through an enzymatic series into lactic acid, which can lower pH, thus stimulating the production of endogenous enzymes to increase nutrient absorption and feed consumption[13].

Absolute Length Growth

Based on the research results regarding the effect of adding petrofish probiotics to feed at different doses on barramundi juveniles. Produce different average lengths of fish juveniles. Treatment D, with a dose of 20 mL/kg feed, had the highest absolute length of 0.73 cm, and A (control) had the lowest total length value of 0.58 cm (Figure 2).

Growth is influenced by heredity, sex, age, parasites, feed, and water conditions[6]. Based on data obtained from test and treatment parameters, fish growth in treatment D was higher than in treatments A, B, and C; this is because a dose of 20 ml/kg feed can increase the presence of the number of bacteria that enter the digestive tract and live in it. According to Arsyad et al.[14], these 12 bacteria can dominate the fish's digestive tract, and the presence of pathogenic bacteria will decrease so that the fish can utilize these good bacteria to grow and be healthy.

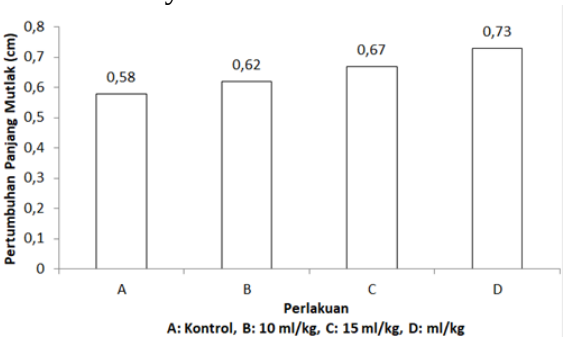

Figure 2. Absolute length growth

Gusnadi et al.[15] stated that the proportion of probiotic bacterial colonies in feed causes the activity of probiotic bacteria to work optimally in fish digestion so that the fish's digestive capacity becomes higher in absorbing food essences and producing good growth. In improving feed nutrition, the bacteria contained in probiotics have a mechanism for producing several enzymes for digesting feed, such as amylase, protease, lipase, and cellulose. These enzymes will help hydrolyze feed nutrients (complex molecules), such as breaking down carbohydrates, proteins, and fats into simpler molecules, facilitating digestion and absorption in the fish's digestive tract. The beneficial bacteria in probiotics added to feed can increase the bacteria in the fish's digestive tract.

The probiotics given can hydrolyze proteins into simpler compounds so that they are easily absorbed through blood vessel walls and used as deposits to increase growth. The increase in growth rate is also thought to be due to the contribution of digestive enzymes by probiotic bacteria, which improve the digestive process of cultivars[16].

Based on the results of calculations using analysis of variance (ANOVA), it shows that the administration of petrofish probiotics in feed genuinely influences the absolute length growth of barramundi fry. $F_{count} > F_{tab}$ at the 1% level,

Survival Rate

The research results show that adding petrofish probiotics to the feed of barramundi juveniles shows the survival of the fish seeds, as shown in Figure 3.

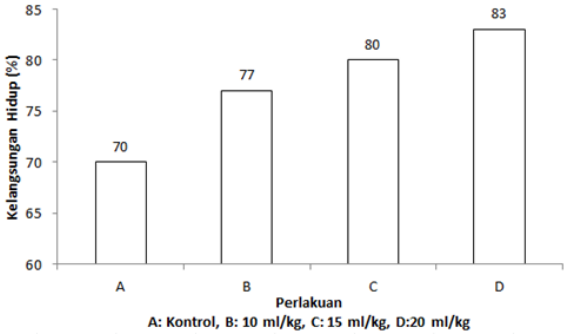

Figure 3. Survival rate

From Figure 3, it can be seen that the average survival rate for barramundi juveniles has differences. The highest average was in treatment D with a dose of 20 ml/kg feed with a survival of 83%, and control treatment A with a value of 70%, which was the lowest SR.

There was no difference in survival rate between the control and the treatment given the probiotic mixture in the feed because the fry in the rearing container could not tolerate the large number of bacteria entering the fish's digestive tract. There is a tendency for fish given synbiotic treatment to have a relatively higher survival rate compared to those treated with probiotics. This is related to the ability of the fish digestive tract to accommodate probiotics without being balanced by providing special nutrition for these bacteria[17].

Based on the results of calculations using analysis of variance (ANOVA), it shows that the administration of petrofish probiotics in feed does not affect the survival of barramundi juveniles. $F_{count} > F_{tab}$ at the 5% level means the difference between each. The treatments had no difference, so no further tests or BNT other tests were carried out on this survival parameter.

Feed Conversion Rate

Based on the research results regarding the effect of adding petrofish probiotics to feed at different doses on barramundi seeds, different average FCR values are produced, as shown in Figure 4.

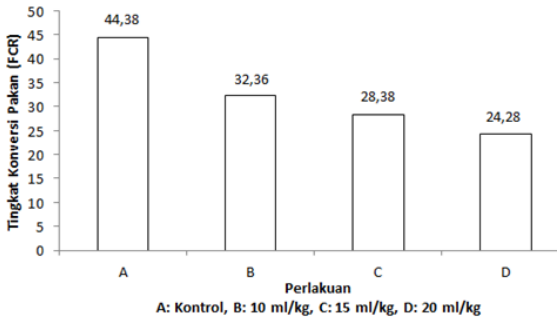

Figure 4. Feed conversion rate

Based on Figure 4, treatment D with a dose of 20 mL/kg feed has the lowest average FCR value of 24.28, and treatment A or control has the highest value of 44.38. Feeding barramundi juveniles with a mixture of probiotics provides differences for each treatment. The good effects for the host caused by probiotics are adjusting the number of bacteria in the digestive tract and producing lactic acid, which can increase the effectiveness of enzymes and help the digestion of food[15]. The lower the feed conversion value, the less is needed to produce 1 kg of fish meat. This means that the more efficiently the feed is converted into meat[16].

The best level of feed use efficiency will be achieved at the lowest feed conversion calculation value, where, in this treatment, the condition of feed quality is better than that of other treatments. Good feed quality conditions result in more energy being obtained by catfish and used for growth, so fish with less feed are expected to have an increased growth rate[19]. Factors that influence the level of feed efficiency are the type of nutrient source and the amount of each nutrient source

component in the feed. The amount and quality of feed given to fish affects fish growth. The higher the feed efficiency value, the better the fish's response to the feed, which is indicated by rapid fish growth[20].

Based on the results of calculations using analysis of variance (ANOVA), it shows that the administration of petrofish probiotics in feed genuinely influences the level of feed conversion in barramundi juveniles. $F_{count} > F_{tab}$ at the 1% level means the difference between each treatment is very significant, so a further test or BNT follow-up test is carried out on the FCR parameters.

Feed Efficiency

Based on the research results regarding the effect of adding petrofish probiotics to feed at different doses on barramundi juveniles. They produced different average EPP values, as shown in Figure 5. Treatment D with a dose of 20 ml/kg feed had a high average EPP value of 4.12, and treatment A or control had the lowest value of 2.26. Feeding barramundi seeds with a mixture of probiotics provides differences for each treatment.

Feed efficiency is the percentage of the amount of feed that can be absorbed by the fish's body so that the fish's weight increases. Feed with the addition of probiotics has better quality compared to feed without probiotics. Giving probiotics can increase the efficiency of feed utilization[15].

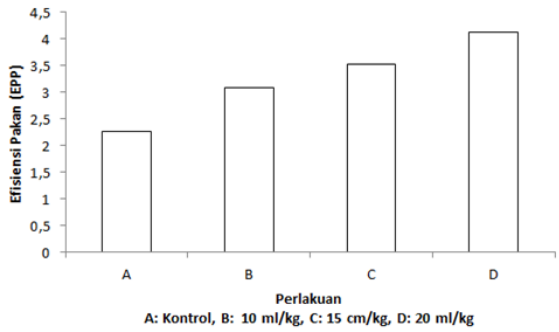

Figure 5. Feed efficiency

Factors that influence the success of probiotic products in increasing growth and feed efficiency in fish are the presence of probiotic bacteria in the fish's digestive tract. Probiotics enter the fish's intestines and then help the digestive process to increase digestibility. The digestibility of feed increases, and the feed will be used more efficiently by the fish because the feed nutrients will be easily absorbed by the body, which in turn will increase protein retention due to the absorption of feed nutrients[21].

Another factor that influences the level of feed efficiency is the type of nutrient source and the amount of each component of the nutrient source in the feed. The amount and quality of feed given to fish affects fish growth. The higher the feed efficiency value, the better the fish's response to the feed is intended[13]. Probiotics in feed can improve the digestive quality of the fish's body so that more feed is absorbed. Bacteria in the fish's digestive tract can secrete digestive enzymes such as protease and amylase. The amount of these secreted enzymes also increases according to the number of probiotic doses given, increasing the amount of feed digested.

Based on the results of calculations using analysis of variance (ANOVA), it shows that the administration of petrofish probiotics to feed has a genuine influence on feed efficiency for barramundi fry. $F_{count} > F_{tab}$ at the 1% level means the difference between each treatment is authentic, so a further test or BNT follow-up test is carried out on the EPP parameters.

Water quality

The water quality observed during the research was temperature, pH, and dissolved oxygen (DO). The water quality observed in the maintenance media can be seen in Table 1.

Adding petrofish probiotics to feed can help improve water quality, total organic matter, ammonia, and phosphate. *Nitrosomonas* bacteria can reduce ammonia levels and form simple compounds that can be utilized for growth by the organism. The autotrophic bacteria

found in probiotics can use uneaten food residue and fish feces at the bottom of the water and then oxidize the ammonia content.

Table 1. Water quality measurement results

Parameters Measured	A	B	C	D
Temperature	30.2 - 31.9	30.6 - 32.0	30.6- 32.1	30.0 - 31.7
pH	7.0 - 7.2	7.0 - 7.1	7.1 - 7.1	7.0 - 7.2
Salinity	30 - 32	31 - 31	30- 32	30 - 32
DO	4.99 - 5.30	5.00 - 5.10	4.97 - 5.14	5.02 - 5.16

Autotrophic bacteria prevent the accumulation of organic nitrogen at the bottom of the maintenance media, which can reduce water quality. The addition of probiotics to the rearing container also functions as a complementary feed source or contributes to the fish's digestive system and can suppress pathogenic bacteria because probiotic bacteria can produce anti-bacterial substances such as bacteriocins, lysozyme, proteases, siderophores, hydrogen peroxide or organic acids[23] so that fish growth can be achieved grow well. Water qualities conditions, including temperature, pH, salinity, and dissolved oxygen content, are within the normal range and follow Indonesian National Standards[23]. The optimal temperature for barramundi seeds ranges from 28 to 32^{o}C. In this range, the temperature parameters are still suitable for supporting the growth of barramundi seeds.

The pH value of water for the growth of white snapper seeds ranges between 7.5 and 8.5. The pH value obtained during the research is also quite good; this can be seen from the results obtained, ranging from 7.5 to 8. The salinity of water ranges from 30-32 ppt, and with the results of these measurements, it can be classified as good salinity during the research.

The DO value of water for the growth of barramundi is around a minimum of 4 ppm. The DO values obtained during the research ranged from 7.0 to 8.0 ppm. The DO value of water for the growth of white sea bass is around a minimum of 4 ppm. The DO values obtained during the research ranged from 7.0 to 8.0 ppm[23].

4. CONCLUSION

Adding petrofish probiotics to feed increases the absolute weight of barramundi seeds, and the best dose of probiotics for the growth of barramundi seeds is found in treatment D, which is 20 mL/kg of feed.

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