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OPTIMIZATION OF PROBIOTIC EM4 (*Effective Microorganism-4*) GIVING WITH DIFFERENT CONCENTRATIONS ON THE GROWTH AND SURVIVAL OF BENEFITS OF MAS FISH (*Cyprinus carpio*)

Optimasi Pemberian Probiotik Em4 (*Effective Microorganism-4*) dengan Konsentrasi Yang Berbeda Terhadap Pertumbuhan Dan Kelulushidupan Benih Ikan Mas (*Cyprinus carpio*)

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ABSTRACT

Dodol, which belongs to the category of semi-moist foods (foods with intermediate moisture), EM4 probiotic is an external supplement that is often used in the world of fisheries which contains bacterialactic acid which plays a positive role in helping the digestive process of fish. This research was carried out in August-September 2022. The method used in this research is an experimental method with a completely randomized design consisting of 3 treatments and 3 replicates. In this research, the addition of EM4 used 3 different treatments, namely P1 (8 ml/kg), P2 (13 ml/kg), P3 (18 ml/kg). Each treatment had 3 repetitions. The feed is added with probiotics, then put into a sprayer to be sprayed onto commercial feed. The sprayed feed is aired for ± 15 minutes. Test fish growth measurements during maintenance were carried out every seven days. The results of the study showed that the addition of EM4 to feed had an effect on the growth performance of goldfish (Cyprinus carpio), but it has no absolute influence on his survival. The best dose was found with the addition of Em4 of 18 ml/kg (P3) which gave an absolute weight growth of 1.96g, absolute length of 2.90 cm, specific growth rate of 0.06%, feed conversion of 0.89 and survival of 100%.

Keywords: Probiotics, EM4, Growth, Survival Rate, Optimization, Goldfish (Cyprinus caripo)

ABSTRAK

Probiotik EM4 merupakan suplemen eksternal yang sering digunakan dalam dunia perikanan yang mengandung asam laktat bakteri yang berperan positif dalam membantu proses pencernaan ikan. Penelitian ini dilakukan pada bulan Agustus-September 2022. Metode yang digunakan dalam penelitian ini adalah metode eksperimen dengan Rancangan Acak Lengkap (RAL) yang terdiri dari 3 perlakuan dan 3 kali ulangan. Pada penelitian ini penambahan EM4 menggunakan 3 perlakuan yang berbeda yaitu P1 (8 ml/kg), P2 (13 ml/kg), P3 (18 ml/kg). Setiap perlakuan memiliki 3 kali pengulangan. Pakan ditambahkan dengan probiotik, kemudian dimasukkan ke dalam sprayer untuk disemprotkan ke pakan komersial. Pakan yang telah disemprotkan diangin-anginkan selama ± 15 menit. Pengukuran pertumbuhan ikan uji selama pemeliharaan dilakukan setiap tujuh hari sekali. Hasil penelitian menunjukkan bahwa penambahan EM4

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pada pakan berpengaruh terhadap performa pertumbuhan ikan mas (*Cyprinus carpio*), namun tidak berpengaruh mutlak terhadap kelulushidupannya. Dosis terbaik didapatkan pada penambahan EM4 sebanyak 18 ml/kg (P3) yang memberikan pertumbuhan berat mutlak 1,96 g, panjang mutlak 2,90 cm, laju pertumbuhan spesifik 0,06%, konversi pakan 0,89 dan kelangsungan hidup 100%.

Kata Kunci: Probiotik, EM4, Pertumbuhan, Kelulushidupan, Optimasi, Ikan Mas (Cyprinus carpio)

INTRODUCTION

One method of providing fish as a source of animal protein is aquaculture. High productivity is the main goal of aquaculture operations. Some important elements in aquaculture are feeding, water quality management, maintenance methods, and the availability of high-quality and high-quantity seeds.

In fish farming efforts, the main obstacle faced is that the assimilation of food nutrients is not optimal, causing food not to be used optimally for fish growth. One type of fish that has very promising prospects for cultivation is carp (Cyprinus carpio). Market demand for this fish is quite high, especially in North Sumatra where carp is often served in traditional events such as weddings and births (Rumondang et al., 2015). The main challenges in carp cultivation are the low survival rate and the growth process which tends to be long (Karlina, 2020).

Artificial feed is used as a source of nutrition to increase the growth of carp (Cyprinus carpio) in cultivation. One of the main challenges is low survival and slow growth rates, which are partly due to the low digestibility of protein in the feed. Probiotics are one method that can improve feed digestion, helping to overcome these obstacles in fish growth. In cultivation practices, feeding is needed not only to support fish metabolism and daily activities, but also for their growth (Septian et al., 2013).

Products containing live and non-pathogenic bacteria are known as probiotics. Probiotics have an impact on the digestive system, therefore giving probiotics to fish will help the absorption and digestion of food. Probiotic microorganisms help improve feed nutrition. (Effective Microorganisms-4) Probiotics called EM4 are often used in the fisheries sector. Organisms are fed probiotics to improve their ability to grow, maintain health, and improve feed efficiency. By stimulating the activity of digestive enzymes, probiotics help break down proteins into simpler molecules, increase nutrient absorption, and act as a source of growth, with the aim of improving fish digestion.

Based on the explanation above, this is the basis of research on the provision of EM-4 probiotics with the title "Optimization of Probiotic EM-4 (Effective Microorganisms-4) Probiotic Administration with Different Concentrations on the Growth and Survival of Goldfish (Cyprinus carpio) Seeds". The purpose of this study was to determine the effect of EM-4 Probiotic administration on the growth of goldfish (Cyprinus carpio) seeds and to obtain the most effective optimization of EM-4 Probiotic administration for goldfish (Cyprinus carpio) seeds.

METHODS

This study was conducted from August to September 2022 at the Chemistry Laboratory of the Matauli Fisheries and Marine College. The tools used in this study were: Jars (25 Liters), Aerators, Digital scales, Ruler, DO meter, pH Meter/Litmus paper, Thermometer, Tangguk, Stationery, on the other hand, the materials used in the study were carp seeds (Cyprinus carpio), pellet feed (PF1000), EM4 probiotics, water (Media)

This study used an experimental method with a Completely Randomized Design (CRD), which included three treatments and three replications. The treatment levels used in this study

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were P0 Without probiotics (Control), P1 Addition of Probiotics to feed 8 ml/kg, P2 Addition of Probiotics to feed 13 ml/kg, P3 Addition of Probiotics to feed 18 ml/kg.

The hypotheses proposed in this study are:

Ho : Provision of EM4 probiotics with different concentrations does not affect the growth and survival of carp (Cyprinus carpio) seeds.

H1 : Provision of EM4 probiotics with different concentrations affects the growth and survival of carp (Cyprinus carpio) seeds.

Data Analysis

Analysis of variance was performed using the SPSS 20 for Windows computer program. To evaluate the differences between treatments, Duncan's advanced test was used with a 95% confidence level. The data obtained from this study will be analyzed statistically and presented in the form of a Variance Analysis Table and histogram or graph. To determine the effect of administering EM4 probiotics, an F statistical test was performed with a 95% confidence level. If the Fcount>Ftable value, then the H0 hypothesis will be rejected and H1 will be accepted. Conversely, if the Fcount <Ftable value, then the H0 hypothesis will be accepted and H1 will be rejected. The results of this analysis will be concluded and accompanied by suggestions.

Parameters measured

Absolute Weight Growth

Absolute weight growth is defined as the increase in the weight of fish maintained until the end of maintenance calculated using the formula (Pramata et al., 2020).

$$Wm = Wt-Wo$$

Description:
Wm : Absolute weight growth (gr)
Wt : Average weight of fish at the end of maintenance (gr)
Wo : Average weight of fish at the beginning of maintenance (gr)

Absolute Length Growth

The increase in fish length from the beginning of maintenance to the end can be calculated using the formula of Pratama et al., (2020), namely:

$$Pm = Lt-Lo$$

Description:
Pm : Absolute length growth (cm)
Lt : Average length at the end of the study (cm)
Lo : Average length at the beginning of the study (cm)

Specific Growth Rate (SGR)

Specific growth is the percentage difference between the final weight and the initial weight in the maintenance period. According to Zonneveld et al., (1991), the specific formula for calculating the specific growth rate is:

$$SGR=(Wt-Wo)/t\times100\%$$

Description:
SGR : Specific growth rate
Wt : Average weight of fish at the end of maintenance (gr)

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Wo : Average weight of fish at the beginning of maintenance (gr)
T : maintenance time (days)

Feed Conversion Ratio (FCR)

Feed conversion is the ratio between the total weight of feed that has been given and the fish biomass obtained within a certain time, during cultivation activities carried out with the fish biomass obtained and calculated during maintenance. Tasyah et al., (2020). The formula used to calculate the feed convention is:

FCR = (Total feed consumed (g)) / (Biomass (g))

Feed Efficiency

To calculate the feed utilization efficiency value, the Amalia et al. formula is used. (2019), as follows:

$$EPP=(Wt-Wo)/F\times100\%$$

Description:
EPP : Feed Utilization Efficiency (%)
Wo : Carp Biomass Weight at the Beginning of Maintenance (g)
Wt : Carp Biomass Weight at the End of Maintenance (g)
F : Amount of Carp Feed Given During Maintenance (g)

Survival Rate (SR)

Survival rate is the total survival rate of fish when they are released until the end of the maintenance period can be calculated using the formula (Pratama et al., 2020).

$$SR=Nt/No \times 100\%$$

Description:
SR : Survival Rate (%)
Nt : Number of carp seeds at the end of treatment (tail)
No : Number of carp seeds at the beginning of treatment (tail)

RESULT

Average Weight Growth of Carp

During 5 (five) weeks of maintenance, the study of body weight growth data of carp (Cyprinus carpio) showed an increase in all treatments from week 0 to week 5. Fish growth was relatively fast from week 0 to week 3 and increased further from week 4 to week 5. The largest average weight growth was recorded in P3 with the addition of EM4 probiotics of 18 ml/kg, namely 3.48 grams, followed by P2 with a dose of 8 ml/kg of 3.25 grams, P1 with a dose of 13 ml/kg of 2.62 grams, and the smallest value in P0 without the addition of EM4 probiotics, namely 2.35 grams.

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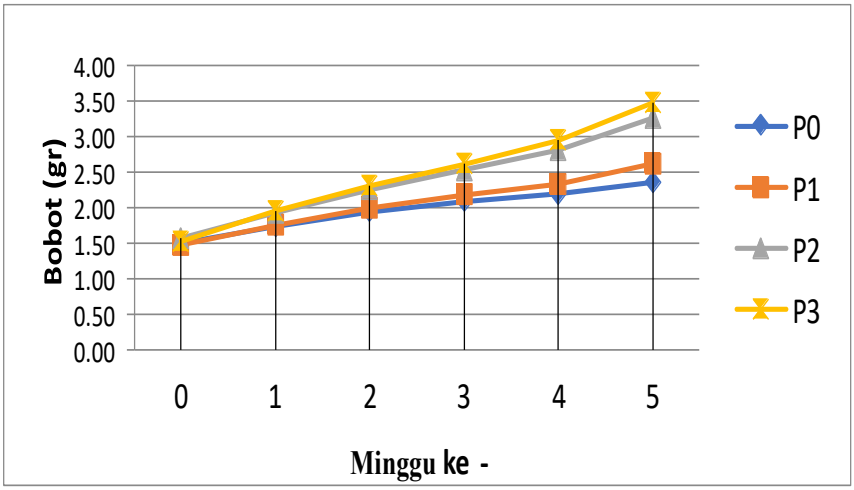

Figure 3. Graph of average weight growth of goldfish

Absolute Weight Growth of Goldfish

The results of observations of goldfish body weight during five weeks of maintenance showed that feed that added EM4 probiotics was different from feed that did not.

Table 3. Absolute Weight Growth of Goldfish During the Study

Treatment	Absolute Weight Growth (gr)			
	P0	**P1**	**P2**	**P3**
1	0,82	1,24	1,52	2,12
2	0,75	1,03	1,67	2,21
3	0,95	1,17	1,84	1,55
Total	**2,52**	**3,45**	**5,03**	**5,88**
Rata-rata	**0,84±0,10^{a}**	**1,15±0,11^{a}**	**1,68±0,16^{b}**	**1,96±0,36^{b}**

Note: Different superscript letters in the same row indicate significant differences (P<0.05).

Table 3 shows the average results of absolute weight growth of carp. The highest value for P3 with a probiotic dose of 18 ml/kg, which is 1.96 grams, followed by P2 with a probiotic dose of 13 ml/kg, which is 1.68 grams, and P1 with a probiotic dose of 8 ml/kg, which is 1.15 grams, and the lowest value for P0, which is 0.84 grams.

Average Length Growth of Carp (*Cyprinus carpio*)

Figure 4 below shows the results of observations of carp growth during 35 days of maintenance.

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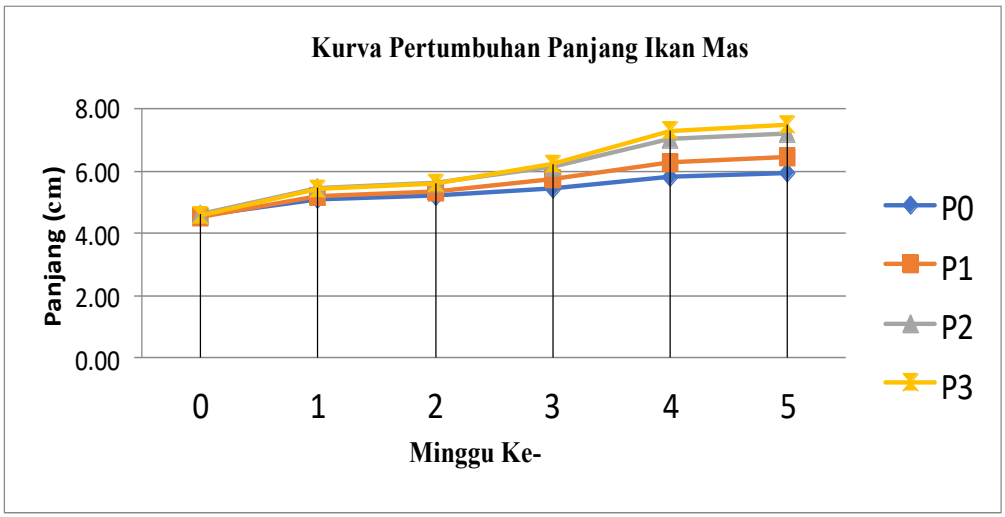

Figure 4. Average length growth graph of goldfish image

The curve illustrates that the average length growth of carp (Cyprinus carpio) increased in all treatments. The use of EM4 probiotics in feed resulted in a greater increase in the length of carp (Cyprinus carpio) compared to the control group P0. The highest length was achieved in P3, which was 7.49 cm with an EM4 dose of 18 ml/kg; followed by P2 which reached 7.18 cm with a dose of 13 ml/kg; followed by P1 which reached 6.43 cm with a dose of 8 ml/kg; and the lowest was P0, which reached 5.91 cm.

Absolute Length Growth of Carp (*Cyprinus carpio*)

Absolute length growth can be defined as the difference between the length of the fish at the beginning and at the end of the study, multiplied by 100%. The results showed that the variation in absolute length growth of carp during the maintenance period varied between 1.35 cm and 2.90 cm. The highest absolute growth was recorded in the P3 treatment group with a probiotic dose of 18 ml/kg, reaching 2.90 cm. Followed by P2 using a probiotic dose of 13 ml/kg which reached 2.56 cm, P1 with a probiotic dose of 8 ml/kg achieved an average length growth of 1.90 cm, and the lowest at P0 which was 1.35 cm.

Table 4. Absolute Length Growth of Goldfish During the Study

Treatment	Absolute Length Growth (gr)			
	P0	**P1**	**P2**	**P3**
1	1,38	1,95	2,50	3,26
2	1,28	1,85	2,55	3,29
3	1,39	1,89	2,63	2,15
Total	**4.05**	**5.69**	**7.67**	**8.70**
Average	**1.35±0.06^{a}**	**1.90±0.05^{a}**	**2.56±0.06^{b}**	**2.90±0.65^{b}**

Note: Different superscript letters in the same row indicate significant differences (P<0.05).

Specific Growth Rate (SGR)

Daily growth rate (DGR) or Specific Growth Rate (SGR) refers to the change in weight, size, or volume of fish over a period of time.

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Table 5. Specific growth rate of carp during the study

Specific growth rate of carp			
Treatment	**Average Weight (gr)**		**Average Specific Growth Rate (%)**
	Begin	**Last**	
P0	1.51	2.35	0.02±0.01a
P1	1.47	2.62	0.03±0.02a
P2	1.57	3.25	0.05±0.02b
P3	1.52	3.48	0.06±0.05b

Note: Different superscript letters in the same row indicate significant differences (P<0.05).

The results of the table above show that the specific growth rate of carp seeds during the study ranged from 0.02 to 0.06%. Treatment variations showed significant differences in the specific growth rate values, with the highest value recorded in the P3 treatment (0.06%) and the lowest in the P0 treatment (0.02%).

FCR (*Feed Conversion Ratio*)

The feed conversion ratio (FCR) describes the efficiency of converting feed consumed by fish into fish body biomass.

Table 6. FCR (*Feed Conversion Ratio*)

Treatment	**FCR (*Feed Conversion Ratio*)**			
	P0	**P1**	**P2**	**P3**
1	1,83	1,64	1,54	1,45
2	1,89	1,81	1,54	1,45
3	1,76	1,64	1,54	1,58
Total	**5,48**	**5,09**	**4,62**	**4,48**
Average	**1.83±0,05b**	**1,70±0,08b**	**1,54±0,00a**	**1,49±0,06a**

Note: Different superscript letters in the same row indicate significant differences (P<0.05).

According to the table above, the P0 treatment has the highest feed conversion ratio of 1.83, followed by the P1 treatment with a value of 1.70. Furthermore, the P2 treatment has a feed conversion ratio of 1.54, and the lowest is the P3 treatment with a value of 1.49. The P3 treatment using a dose of 18 ml/kg showed the best feed conversion ratio in this study, with the lowest FCR when compared to other treatments.

Feed Efficiency

The feed efficiency value is calculated by comparing the increase in fish body weight and the amount of feed consumed by fish during 35 days of maintenance.

Table 7. Feed efficiency

According to the table above,	According to the table	According to the table above, the P0 treatment	According to the table

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	Feed Efficiency	Feed Efficiency	Feed Efficiency	P3
The feed efficiency value is calculated by comparing the increase in fish body weight and the amount of feed consumed	The feed efficiency value is calculated by comparing the increase in fish body weight and the amount of feed consumed	The feed efficiency value is calculated by comparing the increase in fish body weight and the amount of feed consumed	The feed efficiency value is calculated by comparing the increase in fish body weight and the amount of feed consumed	39.81

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by fish during 35 days of maintenance.	by fish during 35 days of maintenance.	by fish during 35 days of maintenance.	by fish during 35 days of maintenance.	
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Feed Efficiency	Feed Efficiency	Feed Efficiency	Feed Efficiency	**112.64**
The feed efficiency value	The feed efficiency	The feed efficiency	The feed efficiency	**37.55±3.92**

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is calculated by comparing the increase in fish body weight and the amount of feed consumed by fish during 35 days of maintenance.	value is calculated by comparing the increase in fish body weight and the amount of feed consumed by fish during 35 days of maintenance.	value is calculated by comparing the increase in fish body weight and the amount of feed consumed by fish during 35 days of maintenance.	value is calculated by comparing the increase in fish body weight and the amount of feed consumed by fish during 35 days of maintenance.

The results of the table above show that the highest average result was achieved in the P3 treatment, which was 37.55 ± 3.92, followed by the P2 treatment of 33.43 ± 0.01, then P1 of 24.15 ± 4.53, then P0 of 17.03 ± 3.52. The results of the study showed that the P3 treatment had the highest feed efficiency value. This also shows a significant effect on the P2 treatment, as well as a significant difference between the P1 and P0 treatments.

Survival Rate

The results of the study showed the survival rate of carp during the experiment. The survival of carp in all treatment conditions showed a consistent pattern above 90%.

Table 8. Survival of carp during the study

Treatment	N_0 (ekor)	N_t (ekor)	SR (%)
P_0	45	42	93,33±6.50
P_1	45	43	95,67±7.50
P_2	45	45	100±0.00
P_3	45	45	100±000

Keterangan: ± Standar deviasi (SD). P0 (kontrol), P1 (dosis EM4 8 ml/kg pakan), P2 (13 ml/kg), dan P3 (18 ml/kg).

The research findings indicate that the highest percentage of fish survival value was in treatments P2 (13 ml/kg EM4) and P3 (18 ml/kg EM4) which was 100%, then followed by treatment P1 (8 ml/kg EM4) with a survival rate of 95.67% and P0 (control) with a survival rate of 93.33%.

DISCUSSION

Effect of EM4 on Growth of Goldfish Weight

The addition of EM4 probiotics has a significant effect on the growth of goldfish weight. The results showed that a higher dose of EM4 probiotics contributed to a better increase in fish weight compared to the control group. The highest dose, which was 18 ml/kg (P3), gave the best results with an average weight of 3.48 grams, followed by a dose of 13 ml/kg (P2) with an average weight of 3.25 grams, and a dose of 8 ml/kg (P1) with an average weight of 2.62 grams. Meanwhile, the control group (P0) without the addition of EM4 only showed a weight gain of 2.35 grams.

Adding probiotics to the feed media allows for optimal goldfish growth, which means that the source of feed energy is believed to be used more for goldfish growth than for maintenance, the reason for this growth is because the energy in the feed is more than what is needed to survive (Rachmawati, 2017).

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This effect is also seen in absolute weight growth, where P3 has the highest value of 1.96 grams, followed by P2 with 1.68 grams, and P1 with 1.15 grams. The control group (P0) only achieved an absolute weight growth of 0.84 grams. Analysis of variance showed that variations in the dose of EM-4 probiotics had a significant effect on growth ($p < 0.05$). The addition of EM4 probiotics helps suppress pathogenic microorganisms in the digestive tract, which allows fish to absorb nutrients more efficiently. Thus, the use of EM4 at the right dose, especially at the highest dose (18 ml/kg), can significantly increase feed efficiency and weight growth of carp.

Growth in Length of Carp

The growth in length of carp (Cyprinus carpio) shows a positive effect of the addition of EM4 probiotics in the feed. In this study, the highest average length growth was recorded in the P3 treatment (dose 18 ml/kg) with a length of 7.49 cm. Furthermore, treatment P2 (dose 13 ml/kg) produced an average length of 7.18 cm, and treatment P1 (dose 8 ml/kg) achieved a length of 6.43 cm. The control group (P0) without the addition of probiotics had the lowest length growth, which was 5.91 cm.

The addition of EM4 probiotics plays a role in increasing nutrient absorption through the activity of digestive enzymes produced by probiotic microorganisms. With increased nutrient absorption, fish use more energy for growth, both in terms of length and weight. The study showed that the increase in fish length was in line with the increase in the dose of probiotics given.

In addition, the absolute length growth of fish for five weeks also showed significant variation. The highest absolute length growth was recorded in P3 with a length of 2.90 cm, followed by P2 with a length of 2.56 cm, and P1 with a length of 1.90 cm. The control group (P0) showed the lowest absolute length growth of 1.35 cm. Thus, the results of this study show that the addition of EM4 probiotics with a higher dose has a significant impact on increasing the length growth of carp.

Specific Growth Rate

The addition of EM4 probiotics to carp feed significantly affected the specific growth rate (SGR). In treatment P3 (EM4 dose 18 ml/kg), the highest specific growth rate was achieved with a value of 0.06%. Furthermore, P2 (EM4 dose 13 ml/kg) showed a growth rate of 0.05%, and P1 (EM4 dose 8 ml/kg) had a growth rate of 0.03%. In contrast, the control treatment (P0) which was not given probiotics only showed a specific growth rate of 0.02%.

EM4 probiotics help increase the efficiency of nutrient absorption from feed through the role of microorganisms that support the activity of digestive enzymes in the fish intestines. The increase in enzyme activity allows fish to use more nutrients from feed for growth, so that the specific growth rate increases along with the increase in the probiotic dose.

The significant difference in specific growth rate between the treatment with the addition of EM4 and the control shows that probiotics play an important role in accelerating fish growth. Fish fed with probiotics are able to utilize energy from feed more efficiently to support growth compared to fish without probiotics.

Feed Efficiency and Conversion Ratio

The results showed that the administration of EM4 probiotics had a significant impact on feed efficiency and feed conversion ratio (FCR) in carp. In treatment P3 (EM4 dose 18 ml/kg), the FCR produced was 1.49, which was the lowest value and indicated the highest feed efficiency. Treatment P2 (13 ml/kg) produced an FCR of 1.54, while P1 (8 ml/kg) produced an FCR of 1.70. The control group (P0) which was not given probiotics had the highest FCR of 1.83.

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The lower FCR in the treatment with a higher dose of EM4 indicates that the feed given is more efficiently converted into fish body biomass. The lower the FCR value, the more efficient the fish are in using feed for growth. EM4 probiotics help increase feed digestibility by supporting the activity of digestive enzymes that hydrolyze proteins into simpler compounds so that they are more easily absorbed.

In addition, the results of the study also showed that the P3 treatment had the highest feed efficiency value with an average of 37.55±3.92, followed by P2 with a feed efficiency of 33.43±0.01, P1 of 24.15±4.53, and P0 with 17.03±3.52. Higher feed efficiency indicates that fish in the treatment with a higher dose of EM4 are able to utilize feed more effectively for growth.

Survival Rate

The study showed that the administration of EM4 probiotics in carp (Cyprinus carpio) feed did not have a significant effect on the survival rate of fish, although in general the survival rate of fish remained high in all treatments. The highest survival rate, which was 100%, was recorded in treatments P2 (EM4 dose 13 ml/kg) and P3 (EM4 dose 18 ml/kg). Meanwhile, treatment P1 (EM4 dose 8 ml/kg) showed a survival rate of 95.67%, and the control group (P0) had a survival rate of 93.33%.

The high survival rate of fish in all treatments indicates that EM4 probiotics are able to maintain fish health by increasing immunity and reducing pathogen levels in the aquatic environment. Although there was no significant difference between the treatments given probiotics and the control, a higher dose of probiotics still provides additional protection to fish against various environmental factors that can threaten survival.

This is supported by research stating that probiotic bacteria in EM4 can help reduce ammonia levels and other harmful substances in the water, thus creating a better environment for fish survival. In addition, microorganisms in probiotics help strengthen the fish's immune system through the production of compounds such as polysaccharides, which play an important role in maintaining fish health. Overall, the administration of EM4 probiotics did not have an absolute impact on increasing fish survival, but was able to maintain a high survival rate throughout the study.

CONCLUSION

The findings of the study indicate that adding EM4 to feed affects the growth performance of carp (Cyprinus carpio), although it does not have an absolute impact on its survival. The optimal dose was found at the addition of EM4 of 18 ml/kg (P3) which resulted in an increase in overall weight of 1.96 g, an overall length of 2.90 cm, a specific growth rate of 0.06%, a feed conversion of 0.89, and a survival of 100%. Water conditions throughout the study also helped the growth and survival of carp, with temperatures between 28 - 30 ℃, pH 7, and dissolved oxygen (DO) content of 5.37 - 76.67 mg/L.

The use of EM4 Probiotics at an ideal dose of 18 ml/kg in carp seed feed showed significant results in increasing the growth of fish weight and length, specific growth rate, and feed conversion productivity. Although it does not affect survival absolutely, this study provides knowledge regarding the optimal administration of probiotics to increase feed utilization productivity, growth, and survival of carp (*Cyprinus carpio*).

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