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# Effect of Feed Enrichment with Probiotics on Vitality Koi Fish (*Cyprinus rubrofuscus*)

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**ABSTRACT:** Koi fish (*Cyprinus rubrofuscus*) is a type of fish that has high economic value and is easy to maintain. This fish has a large size and fast growth and attractive body shape and color. The purpose of this study was to determine the effect of giving probiotics on the vitality of koi fish (*Cyprinus rubrofuscus)* and to find out the best dose that gives effect to giving probiotics on the growth and survival of koi fish (*Cyprinus rubrofuscus*). This research was carried out from 20 February to 20 April 2022 which is located at the Biota and Environmental Engineering Laboratory, Faculty of Fisheries and Marine Sciences, Muslim University of Indonesia.The method used in this study was an experimental method with a completely randomized design (CRD) consisting of four treatments (A, B, C and D). Each treatment was three replication. The dose for each treatment is A (0 ml), B (5 ml), C (10 ml) and D (15 ml). Then in this study there are 3 parameters, namely weight growth, length growth and survival rate. Based on the results of the study which showed that feed enrichment with probiotics has a significant effect on the growth of weight and length but does not affect the survival rate of koi fish; treatment of 10 ml dose of probiotics gives the best growth; water quality is at optimal conditions for the growth and survival of koi fish.

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**KEYWORDS:** Koi fish (*Cyprinus rubrofuscus*), probiotics, growth.

## INTRODUCTION

Koi fish (*Cyprinus rubrofuscus*) is a type of fish that has high economic value and is easy to maintain. This fish has a large size and fast growth and attractive body shape and color. One of the important factors in aquaculture is fish growth. To increase fish growth, artificial feeding is carried out, but if you only rely on artificial feed, the benefits will be less. In this regard, it is now popular among cultivators, namely the addition of probiotics to increase fish growth. Probiotics are microorganisms that have a beneficial role and are able to survive in the digestive tract. Probiotics are useful in preventing intestinal pathogenic microorganisms and improving the efficiency of producing artificial feed by releasing enzymes that help process the digestion of food in fish. The bacterial content present in probiotics produces several enzymes for digestion of feed, namely amylase, protease, lipase and cellulose. These enzymes will help the process of hydrolyzing nutrients in stored artificial feed (complex molecules), such as carbohydrates, proteins and fats to turn into simpler molecules from the start so that it is easy in the process of digestion and absorption of feed in the digestive tract of fish. Several probiotics have been used in aquaculture activities and have played a role in increasing growth, survival rates, digestibility, feed efficiency, the immune system and the composition of beneficial bacteria (probiotics) in the digestive tract of fish. Probiotics contain microbes that function to break down metabolic waste and encourage an immune response so that fish health increases and affects the growth rate of fish.

The purpose of this study was to determine the effect of giving probiotics on the vitality of koi fish (*Cyprinus rubrofuscus*) and to find out the best dose that gives effect to giving probiotics on the growth and survival rate of koi fish (*Cyprinus rubrofuscus*.)

## RESEARCH METHODOLOGY

This research was carried out at the Biota and Environmental Engineering Laboratory, Faculty of Fisheries and Marine Sciences, Indonesian Muslim University from February to April 2022.

## RESEARCH PROCEDURE

The research was conducted based on several stages, namely as follows:

*1. Container Preparation*

Preparation of the container by cleaning or washing the aquarium using soap or detergent so that the aquarium is clean then dried, after the container is dry the next step is filling water, the water used comes from a drilled well which is precipitated while aerating for 2 days to remove iron from the water after When sedimentation is complete, 20 L/container is filled with water in the research container, then aeration is installed to supply oxygen. The size of the aquarium used is 396 cm x 258 cm x 282 cm with a volume of 25 L of water.

*2. Fish Stocking and Adaptation*

The test animals used were koi fish seeds (*Cyprinus rubrofuscus*) which had an average length of 4 – 6 cm and weighed ± 1 g. Test animals were stocked at a density of 15 individuals/container. The spread of fish to the previous research media must be adapted first. Adaptation was carried out for 7 days but on the first day the fish were given feed without differences in treatment with a predetermined dose.

*3. Feeding*

Feeding the koi fish (*Cyprinus rubrofuscus*) was carried out 3 times a day at 08.00 am, 11.00 am and 16.00 pm with a feeding ratio of 10% of the weight of the biomass.

*4. Pension*

In this study siphoning of leftover feed and faeces was carried out every 10 days to maintain optimal water quality.

*5. Probiotic Administration Technique in Feed*

The technique of giving probiotics to the feed is by spraying the probiotics on the feed with a sprayer then stirring it thoroughly and then letting it dry, then weighing it and packing it. The type of probiotic used is Probiotic Bacillus.

*6. sampling*

In this study, sampling was carried out every 10 days by measuring the weight and total length to determine the response to the treatment.

## RESEARCH DESIGN

The design used in this study was a completely randomized design (CRD) consisting of four treatments (A, B, C and D). Each treatment was three replications. This research was conducted using probiotic dose treatment in each feed formulation, while the treatment was as follows:

Treatment A = probiotic dose of 0 ml
Treatment B = probiotic dose of 5 ml
Treatment C = probiotic dose of 10 ml
Treatment D = probiotic dose of 15 ml

This study was designed with a completely randomized design (CRD) with four treatments and three replication, so 12 containers were needed.

The test feed used during the study was self-formulated feed with ± 30% protein content (Table 1).

**Table 1. Test feed formulation (% dry matter)**

| Raw material | Treatment | | | |
|---|---|---|---|---|
| | A | B | C | D |
| Fish flour | 33 | 33 | 33 | 33 |
| Soybean flour | 25 | 25 | 25 | 25 |
| Fine bran | 25 | 25 | 25 | 25 |
| Cornstarch | 10 | 10 | 10 | 10 |
| Wheat | 5 | 5 | 5 | 5 |
| Vitamin | 2 | 2 | 2 | 2 |
| Total | 100 | 100 | 100 | 100 |
| Proximate composition (% in dry weight) | | | | |
| Protein(%) | 30.36 | 30.36 | 30.36 | 30.36 |
| Fat(%) | 9.30 | 9.30 | 9.30 | 9.30 |

**Parameter**

The parameters observed were: Weight Growth (WG), length growth (LG), and Survival Rate (SR) using the equations of Panase & Mengumphan (2015):

*1. Weight Growth*

$$W = W_t - W_0$$

Information:
W = Growth weight (g)
$W_t$ = final weight (g)
$W_0$ = Initial weight (g)

*2. Length Growth*

$$L_m = L_t - L_0$$

Information:
$L_m$ = Growth length (cm)
$L_t$ = Final length (cm)
$L_0$ = Initial length (cm)

*3. Survival Rate*

$$SR = N_t/N_0 \times 100\%$$

Information:
SR = Survival rate (%)
$N_t$ = Final number of fish
$N_0$ = Initial number of fish

**Data ANALYSIS**

To determine the effect of treatment on the parameters measured, an analysis of variance (ANOVA) was carried out and to determine whether a treatment was significantly different with an alpha value of 0.05. In order to find out the effect of the treatment applied during the research whether it gave the best results, the Tukey HSD (High Standard Deviation) advanced test was carried out.

**RESULT AND DISCUSSION**

**Weight Growth**

Based on the research results obtained on the weight growth (g) of koi fish (*Cyprinus rubrofuscus*) with a density of 15 fish/20 L with the effect of giving different doses of probiotics to the feed can be seen in table 2.

**Table 2. Average weight growth (g)**

| Treatment | Weight Growth (g) |
|---|---|
| A (0 ml) | $0.14 \pm 0.01^{a}$ |
| B (5 ml) | $0.15 \pm 0.02^{a}$ |
| C (10 ml) | $0.29 \pm 0.01^{b}$ |
| D (15 ml) | $0.16 \pm 0.03^{a}$ |

Table 2 shows that the highest weight growth was obtained from treatment C (10 ml), namely 0.29 ± 0.01, based on analysis of variance showing results that had a significant effect on absolute weight growth ($P < 0.05$). Thus the addition of 10 ml of probiotics can provide a response to higher weight growth. This is presumably because the amount of feed given in each treatment greatly affects the growth of koi fish (*Cyprinus rubrofuscus*) seed weight. As reported by (Mulyadi, 2011) that the number of bacteria that are too many causes the bacteria to quickly experience sporulation (form spores) so that the function and activity of the bacteria Bacillus sp. not optimal. Then according to Arief (2013) that the bacteria Bacillus sp. plays a role in balancing digestive tract microbes so as to increase the digestibility of fish by converting carbohydrates into lactic acid which can lower pH, thus stimulating the production of endogenous enzymes to increase nutrient absorption, feed consumption, growth and inhibit pathogenic organisms.

**Length growth (cm)**

The length growth obtained during the study on koi fish (*Cyprinus rubrofuscus*) seeds can be seen in table 3.

**Table 3. Average length growth (cm)**

| Treatment | Average Length growth (cm) |
|---|---|
| A (0 ml) | $0.18 \pm 0.03^{a}$ |
| B (5 ml) | $0.23 \pm 0.05^{ab}$ |
| C (10 ml) | $0.30 \pm 0.04^{b}$ |
| D (15 ml) | $0.21 \pm 0.05^{a}$ |

From table 3 it can be seen that the highest length growth was obtained from treatment C (10 ml), which is around 0.30 ± 0.04 cm. Based on the statistical test results, it showed that there was an effect (P <0.05) with treatment A (0 ml) and treatment D (15 ml) although it showed no effect (P > 0.05) with treatment B (5 ml). This is in line with the results of growth growth is accompanied by an increase in absolute length as reported by (Nazar, 2018) that the increase in fish length will be accompanied by an increase in weight. The benefits of adding probiotics include increasing the digestibility of feed by the host, inhibiting the growth of pathogenic bacteria in the intestine, boosting the immune system and increasing growth performance (Ringo, 2020).
Fish will utilize feed nutrients to be stored in the body and convert them into energy. This energy is used by fish for basic metabolism, movement, production of sexual organs, maintenance of body parts and replacement of damaged cells and the rest is used for growth (Dewi, *et. al* 2019).

**Survival Rate**

The survival rate of koi fish (*Cyprinus rubrofuscus*) maintained during the study by giving different doses of probiotics to the feed can be seen in Table 4.

**Table 4. The results of the average survival rate**

| Treatment | SR |
|---|---|
| A (0 ml) | 73.33 ± 0.00[a] |
| B (5 ml) | 75.55 ± 0.04[a] |
| C (10 ml) | 80.00 ± 0.07[a] |
| D (15 ml) | 77.78 ± 0.08[a] |

From table 4 it can be seen that the highest survival rate was obtained from treatment C (10 ml) which is around 80.00 ± 0.07 based on the statistical test results showing no effect (P > 0.05) with all treatments.
The survival rate is influenced by the feed given. The addition of probiotics is thought to help improve the immune system of fish that consume them. This was reinforced by Azhar (2013) who stated that probiotics are also able to act as immunostimulants, increase feed conversion ratios, have the ability to inhibit the growth of pathogenic bacteria, produce antibiotics. The survival rate of koi fish (*Cyprinus rubrofuscus*) seeds during the study ranged from 73% - 80%. This is due to the provision of probiotics in the feed being able to maintain water quality so that the health of the fish is maintained, the death of koi fish seeds that occurred during the study was allegedly due to siphoning and changing water which could result in stress on koi fish seeds and cause death. According to (Kurniawan, 2020) the survival rate of fish fry is in the range of more than 50% which is classified as good, 30-50% is classified as moderate and less than 30% is not good.

**Water quality**

Water quality is a supporting factor for the growth and survival of fish. Water quality parameters measured during the study were temperature, acidity (pH), dissolved oxygen (DO), and ammonia (NH3). The results of water quality measurements during the study and their feasibility values based on the literature are presented in Table 5.

**Table 5. Results of water quality measurements**

| Treatment | Water quality | | | |
|---|---|---|---|---|
| | Temperatur ($^0$C) | pH | Ammonia (mg/l) | DO (mg/l) |
| A | 27 - 28 | 7 – 7.5 | 0.043 | 5.0-6.0 |
| B | 28 - 29 | 7 – 7.5 | 0.043 | 5.0-6.0 |
| C | 27 - 28 | 7 – 7.5 | 0.043 | 5.0-6.0 |
| D | 27 - 28 | 7 – 7.5 | 0.043 | 5.0-6.0 |

During the research, the temperature has a value in the range of 27 – 29 $^o$C. This shows that the temperature in the aquarium of the test animals can be said to be good for maintenance. By looking at the temperature results during the study it was still categorized as good for fish farming because in waters generally fish cultivation requires temperatures in the range of 25 - 30$^o$C (Khairuman, 2002). If a waters has a pH below 4 or exceeds 11, there will be mass death of aquatic biota in these waters. While during the study the pH ranged from 7.0 - 7.2. Based on the pH value in the study, it can be categorized as good for an aquaculture activity, because it is still in the ideal category as in its native waters, namely 7 - 8.5 (Kordi, 2010). Based on the results of research conducted dissolved oxygen (DO) found in maintenance aquariums, namely 5.0 - 6.0 ppm and is still categorized as good because it is in accordance with the opinion of (Monalisa, 2010) that the minimum value of dissolved oxygen for aquaculture activities is 3 - 5 ppm. Based on the results of the research conducted, the ammonia contained in the maintenance aquarium was 0.043 mg/L. Looking at the results of the ammonia content in all treatments, it shows that this is still categorized as normal for aquaculture activities, which is still below the 2 mg/L rate (Pangestu, 2020). This happened because there was a large amount of leftover feed that was not consumed by the fish so that the feed given at the time of the study only dissolved in water, causing unhealthy water conditions for

the fish. In addition, this also happens because the feed content used is quickly destroyed in the water. Ammonia data that probiotic feeding is beneficial in reducing ammonia content in leftover feed, presumably because probiotics are food ingredients that cannot be digested by the host but have a positive effect by stimulating the growth of beneficial bacteria in the host digestive tract (Ringo, *et al,* 2010).

## CONCLUSION

1. Feed enrichment with probiotics has a significant effect on the growth of weight and length but does not affect the survival rate of koi fish
2. Treatment of 10 ml dose of probiotics gives the best growth
3. Water quality is at optimal conditions for the growth and survival of koi fish

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