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Growth Performance of Mahseer Fingerlings (*Tor soro*) Fed Potassium Diformate Added Diets

Lingga Ananda Riyani [a*], Ayi Yustiati [a], Emma Rochima [a] and Walim Lili [a]

[a] *Fisheries Department, Faculty of Fisheries and Marine Science, Padjadjaran University, Jl. Raya Bandung-Sumedang, Hegarmanah Jatinangor, Sumedang 45363, West Java, Indonesia.*

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Mahseer aquaculture activities are still relatively low because they are constrained by slow growth. Potassium diformate can promote growth by improving digestion. This research aimed to determine the optimum dose of potassium diformate addition to artificial feed for the best growth rate in mahseer fingerlings. The research was conducted from July – August 2022, in hatchery of the Faculty of Fisheries and Marine Sciences, Padjadjaran University, West Java, Indonesia. The method used in this research was experimental, using a completely randomized design (CRD) consisting of four treatments and five replications. The test fish used were 200 mahseer fingerlings measuring 4.5 – 5.0 cm. The treatment used was the addition of potassium formate to artificial feed at a dose of 0% (A), 0.1% (B), 0.3% (C), and 0.5% (D). The length of maintenance is 40 days, with the amount of feeding 3% of the fish's body weight. Parameters observed included specific growth rate (SGR), feed conversion ratio (FCR), survival rate (SR), and water quality (dissolved oxygen, temperature, pH). The research results showed that the addition of 0.3% potassium diformate gave

*Corresponding author: Email: lingga18001@mail.unpad.ac.id;

the best results with the highest SGR value of 0.75%, FCR of 3.80, EP of 26.87%, SR of 100%. It can be concluded that the addition of potassium difomate has a good effect on the growth of mahseer fingerlings if given at optimal doses.

Keywords: Mahseer (Tor soro); organic acid; growth.

1. INTRODUCTION

Mahseer (*Tor soro*) is a potential local fish that can be a candidate for aquaculture. Mahseer has cultural values among certain communities [1]. Mahseer a consumption fish, this fish has gained popularity because it has soft and thick meat and is tasty [2], in terms of nutritional content, mahseer has a high albumin protein content of 13.00 ± 0.04 g 100 m/L [3]. This reason makes the mahseer one of the local fish with a high selling price. Domestically, the price of mahseer ranges from IDR 800,000.00 – IDR 1,000,000/kg.

Mahseer aquaculture activities are still relatively low, because mahseer have slow growth [4]. The slow growth rate causes high production costs that must be incurred, especially costs to meet feed needs. Feed is the largest component of production costs in an aquaculture business. One of the factors that can affect the growth of fish is feed. Quality feed will support fast growth and high feed efficiency. Economically, high feed efficiency can reduce production costs [5].

Growth is a change in shape resulting from an increase in length, weight and volume in a certain individually [6]. The growth rate of fish is strongly influenced by the quantity and quality of feed and environmental conditions [7]. In an optimal environment, the feed given is very influential on fish growth.

Improving feed quality can be done by using feed additives. Feed additives are ingredient that does not include food substances added in relatively small amounts with the hope that it can help the process of absorbing nutrients in the digestive tract. Previously, in aquaculture, *antibiotic growth promoter* (AGP) was used to accelerate growth, but the use of AGP has a negative impact because it causes the transfer of resistance against pathogenic bacteria to animals and humans who eat it [8]. Prohibition of the use of AGP in Indonesia has been regulated in Law no. 41 of 2014 concerning animal husbandry and health. Indonesia officially only started banning AGP in feed in January 2018. This ban was also strengthened through regulation of the minister of agriculture No. 22 of 2017 concerning registration and distribution of feed.

Potassium diformate is a double salt formic acid molecule that can lower pH and reduce the growth of pathogenic bacteria in the digestive tract. With a low pH, the absorption of nutrients will be more optimal, thus accelerating [9]. Potassium diformate is an alternative to AGP. Potassium diformated by the European Union became the first substance promised as a non-antibiotic growth promoter. Potassium diformate can reduce the population of pathogenic bacteria in the intestine and has been shown to be effective in increasing growth performance and nutrient absorption in various aquatic species , while ensuring a safer product for consumers. The use of potassium diformate has been proven to be able to increase the growth of cultivated commodity species such as carp, nilem, tilapia, catfish, freshwater pomfret, catfish and vannamei shrimp.

2. MATERIALS AND METHODS

Materials used, including mahseer fingerlings, were 200 measurings 4.5 – 5.0 cm, totaling 250 fish; 200 fish were used for research and the rest as stock. Other ingredients are commercial feed PF1000, potassium diformate as a feed additive, and distilled water as a binder to bind potassium diformate salt to feed so that potassium diformate can be mixed evenly. This research was conducted in a completely randomized design (CRD) with four treatments and five replications. The dietary addition of potassium diformate was tested by four levels, namely 0, 0.1, 0.3, and 0.5%.

2.1 Procedure

2.1.1 Preparation of rearing containers

Research facilities were prepared, such as 20 aquariums with a size of 40 x 30 x 30 cm, fiber tanks, hoses, and aeration stones. The research facility was washed with soap, then rinsed with running water, and soaked with 30 ppm chlorine solution for 24 hours with strong aeration. Then rinsed again using running water and dried. Each aquarium was filled with 20 L of water and given aeration as a container for raising fish during the study.

2.1.2 Test fish preparation

Fish are transported in the morning when the temperature is low. The transport plastic bag containing fish fingerlings is soaked in water in the fiber tub for 20 minutes to equalize the temperature of the water in the plastic with the water temperature in the fiber tub. The plastic bag is opened, the water in the fiber tub is mixed into the water in the plastic bag to slowly equalize the water quality until the fish comes out by itself. In the test fish, the acclimatization process was carried out twice, namely. 1) The fish were reared for 3 days in fiber tanks to ensure the test fish were healthy, by giving food 3 times a day regularly. 2) As many as 10 fish that have been weighed are put into each aquarium by feeding according to the treatment with a maintenance time of 3 days, so that the fish can adjust to the feed that has been mixed with potassium diformate.

2.1.3 Feed preparation

Commercial feed was weighed as much as 100 grams for each treatment and the remaining feed was given to stock fish. Weighing potassium diformate 10 days). potassium diformate is mixed in the feed evenly. Mixed feed. potassium diformate is sprayed with distilled water as much as 10% of the total feed, which serves to glue potassium diformate to the feed. The feed that has been mixed with potassium diformate is air-dried. The feed mixture is weighed at 3% of the fish biomass for each aquarium, then packaged using plastic clips for 3 meals. The nutritional content of the feed used can be seen in Table 1.

2.1.4 Research implementation

Mahseer that have gone through the grading and weighing process are stocked as many as 10 fish per aquarium (stocking density of 1 fish per 2 L), after going through the acclimatization process the mahseer are reared for 40 days. The frequency of feeding is three times a day at 08.00, 12.00 and 16.00 WIB. The amount of feed given is 3% of the total fish biomass in each aquarium.

Water quality maintenance is carried out by siphoning every 3 days in the morning. Sip water as much as 30% of the total volume of water in the maintenance container [8]. Siphoning is done to remove dirt at the bottom of the aquarium, thereby minimizing water pollution in each aquarium.

2.2 Observation

2.2.1 Daily growth rate

Observation of the daily growth rate (SGR) was carried out once every 10 days during the research. To determine daily growth, weighing was carried out when the fish were put into the aquarium (day 0), and then carried out on the 10th, 20th, 30th and 40th days. Weighing is done on the fish in each aquarium by preparing a digital scale and a weighing container, namely a petri dish filled with water. The weighing container is placed on the scale that is already on, then calibrated by pressing the tare button, so that the weighing container does not count the biomass. As many as 10 fish were taken from each aquarium and then put into a petri dish filled with water and the biomass was weighed. Weighing is carried out in the morning to reduce stress on the fish fry so as not to affect the observations that take place during rearing. The daily growth rate is calculated using the formula (Effendie 1997):

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

Explanation :

α : Specific growth rate
Wt : Average weight on the 40th day (g)
W_0 : Average weight on day 0 (g)
t : 40 days

2.2.2 Feed conversion ratio

To determine the feed conversion ratio is done by weighing the weight of the fish in each aquarium at intervals of 10 days during the rearing period. Dead fish were weighed and recorded. The feed eaten by fish counted and recorded. Feed conversion can be calculated using the formula (Effendie 1997):

$$FCR = \frac{F}{(Wt + D) - Wo}$$

Explanation:

FCR : Feed conversion ratio
Wt : Average weight on the 40th day (g)
W_0 : Average weight on day 0 (g)
D : Weight of fish that died during the study
F : Total feed given (g)

Table 1. Nutritional content PF 1000

Nutrition	%Daily Value*
Protein	Min 39 – 41%
Fat	Min 5%
Coarse fiber	Max 6%
Ash contents	Max 16%
Dry measure	Max 10%

2.2.3 Survival rate

Survival rate observations were carried out every day during the study. If at the research implementation stage there are dead fish, the fish's weight is measured and then recorded and discarded. Survival or survival rate (SR) is calculated using the formula (Effendie 1997):

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

Explanation:

SR : Degree of survival (%)
Nt : Number of live fish, 40th day (individual)
N_0 : Number of live fish, day 0 (individual)

2.2.4 Water quality

Observation of water quality is carried out once every 10 days for 40 days of maintenance time. Observations were made in the morning. Parameters of water quality observed included dissolved oxygen (DO), temperature, and pH.

2.3 Data Analysis

Data analysis was carried out to determine how influential the KDF was on the growth rate of the test fish. Data analysis was carried out with the F test. If there was a significant difference between the treatments, it was continued with Duncan's multiple tests at an error level of 5%. Water quality analysis was carried out in a comparative descriptive manner.

3. RESULTS AND DISCUSSION

3.1 Daily Growth Rate

The results of observations on growth showed that the addition of potassium diformate to the feed obtained significant results. The growth obtained at each sampling (10 days) showed an increase, especially on the 20th to 40th day, meaning that the addition of potassium diformate to the feed resulted in a fairly good response to growth (Fig. 1).

To find out the percentage of fish weight growth per day, it is done by calculating the daily growth rate. SGR calculations in the mahseer in this research were carried out for 40 days. The calculation results can be seen in the graph (Fig. 2).

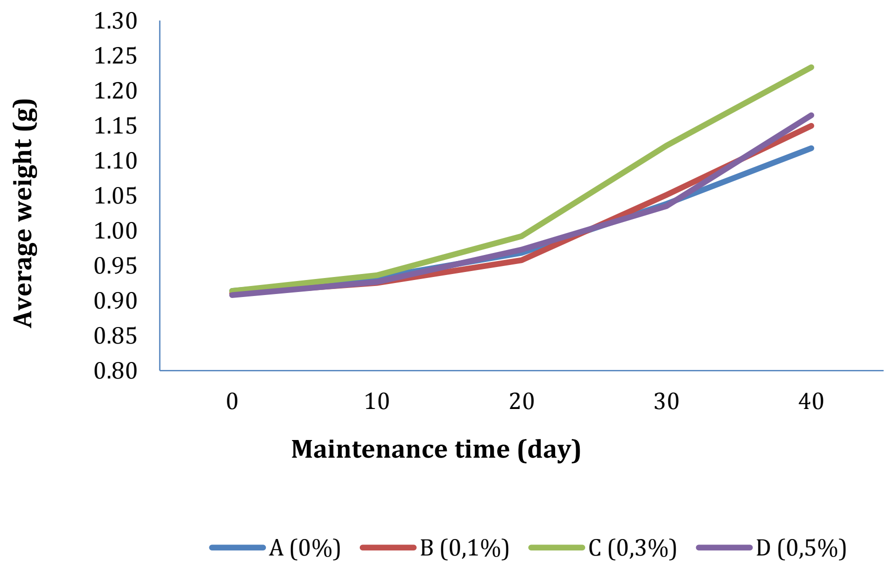

Fig. 1. Growth chart every 10 days during maintenance

The SGR results obtained in this research ranged from 0.50 - 0.75%, this value was quite good when referring to research conducted by Pasaribu [9] with results ranging from 0.49 - 0.92% in mahseer treated with growth hormone (rGH) in commercial feed for 60 days.

The highest SGR in this research was obtained in treatment C (0.3%) with a value of 0.75% per day. SGR results from treatments B (0.1%), C (0.3%), and D (0.5%) when compared to treatment A (0.5%) indicated that the addition of potassium diformate to feed with the range of doses used does not cause a negative impact on the growth of mahseer. At a dose of 0.5% it produces a smaller SGR compared to a dose of 0.3%, this proves that the higher potassium diformate dose does not always have a better effect on fish growth. The use of potassium diformate which is an organic acid salt with excessive doses will cause fish osmoregulation disorders, so that fish use more energy to get rid of excess salt in the body while the energy used for growth will decrease.

Usage potassium diformate which is an organic acid salt with an excessive dose will cause fish osmoregulation disorders, so that fish use more energy to get rid of excess salt in the body while the energy used for growth will decrease. The use of organic acids that exceed the optimum dose risks damaging the stomach and duodenal mucosa [10]. Potassium diformate, which is an organic acid, is capable of releasing low H+, so feeds associated with acids at high doses can inhibit the work of digestive tract enzymes, which results in decreased palatability resulting in reduced feed consumption [11]. Various kinds of chemical signals in the digestive tract will affect receptors that affect appetite (palatability) so that they affect feed consumption [12].

The best growth in this research is obtained in the addition potassium diformate as much as 0.3%. This result is supported by research conducted by Yustiati et al. [13], that addition potassium diformate as much as 0.3% gives the best results for the growth rate of nilem fish (*Osteochilus hasselti*) by 2.7%. Similar results were obtained from research conducted by Yustiatiet et al. [14], addition potassium diformate with a dose of 0.3% produced the highest growth rate for carp (*Cyprinus carpio*) of 3.21%. Addition potassium diformate of 0.3% gave the best results on the daily growth rate of gouramy (*Osphronemus goramy*) of 1.31% [15]. Addition potassium diformate with a dose of 0.3% is the highest treatment to increase the LPH of tilapia (*Oreochromis niloticus*) of 2.07% [16].

Potassium diformate as a growth promoter can increase feed digestibility by lowering the pH of the digestive tract, thereby stimulating pepsin activity more quickly. Under acidic environmental conditions, the digestive tract will absorb more nutrients, because pepsin enzyme activity can be optimum at a low pH, thereby optimizing pepsin activity in breaking down proteins into peptides [17]. According to Booth (2003), a decrease in pH contributes to an increase in the growth and utilization of minerals.

Based on research Elala et al. [18] addition potassium diformate with a dose of 0.3% in feed can reduce the pH of the tilapia stomach and intestine (*Oreochromis niloticus*), the initial pH of the stomach was 3.40 to 2.96 at the end of the study, while in the upper intestine it decreased from 6.88 at the start of the study to 6.43. Use potassium diformate 0.3% can reduce the intestinal pH of carp (*Cyprinus carpio* L.) from 6.97 to 6.26 [14]. The greater the increase in dose potassium diformate in the feed, it will also decrease the resulting intestinal pH value.

The low pH value of the digestive tract of fish can cause pathogenic bacteria in the digestive tract to die and commensal bacteria to develop very well. Mechanism potassium diformate reduce pathogenic bacteria in the digestive tract of aquatic animals, i.e. by penetrating through the cell wall and releasing protons in the cytoplasm, the bacteria consume large amounts of ATP to release protons in an effort to maintain intracellular pH balance. Resulting in damage to the bacterial DNA and subsequently resulting in death. The organic acids contained in potassium diformate can maintain the balance of the population of bacteria that are resistant to acidic conditions in the digestive tract such as lactic acid bacteria which function to maintain the health of the digestive tract, so that the digestive process in the fish's body can work properly and produce optimal growth. Pathogenic bacteria live and thrive at a pH of 6.9 – 7.8 while commensal bacteria develop at a pH of 5.4 – 6.8 [19].

3.2 Feed Conversion Ratio

Feed conversion ratio (FCR) is a measure that states the ratio of the amount of feed needed to produce 1 kg of cultivated fish. The FCR of Mahseer in this research can be seen in the following graph.

Based on the graph (Fig. 3) the value of the feed conversion ratio obtained ranges from 3.80 – 5.71 meaning that by feeding as much as 3.80 – 5.71 kg will produce 1 kg of meat. FCR in this research is quite good when referring to research conducted by Radona et al. [20], fish fingerlings *Tor* sp. measuring 1.5 cm in length produced an FCR ranging from 4.58 to 7.46 which was fed with different protein content for 40 days of rearing. The size of the FCR value is influenced by the absorption of nutrients which are different for each species, size, and aquaculture system.

The results of the analysis of variance showed that the addition of KDF to Mahseer showed significant differences between the treatments on the FCR value. A low FCR value or close to 1 shows the best value. In this research, the lowest FCR value was obtained in treatment C (3%), namely 3.80. Based on Duncan's test, the addition of KDF in treatment C (0.3%) was significantly different from treatment A (0%).

The low FCR in treatment C shows the optimal ability of fish to digest and absorb feed with the addition of 0.3% KDF given during rearing, so as to optimally convert feed into meat. The low FCR value indicates that the feed given is good enough and in accordance with the needs of fish to support growth [21].

The ability of fish to use feed nutrients depends on various factors, one of which is the activity of digestive enzymes. The use of KDF can optimize pepsin enzyme activity by lowering the pH of the digestive tract. The optimal pepsin enzyme activity is a key factor for increasing protein digestibility and accelerating absorption, so that a low feed conversion value is obtained. Based on the FCR value obtained, the addition of KDF to feed at a dose of 0.3% is the optimal dose to produce the lowest FCR. KDF at a dose of 0.3% was able to reduce the FCR by 66.55% from the treatment without the addition of KDF, thereby reducing production costs.

3.3 Survival Rate

Survival rate become one of the factors that can determine the success of a fish farming business. In aquaculture, survival rate is used as a benchmark to see the tolerance of fish to their environment. The greater the percentage value of the survival rate obtained, the higher the fish's ability to survive.

Based on the results of observations of the survival rate of mahseer for 40 days of rearing, it shows various values (Fig. 4). This result is obtained from the total number of live fish at the end of rearing divided by the number of fish at the beginning of rearing expressed in percent units. Based on the analysis of variance that has been carried out, the provision of potassium diformate to feed has no significant effect between treatments on the survival rate of masheer.

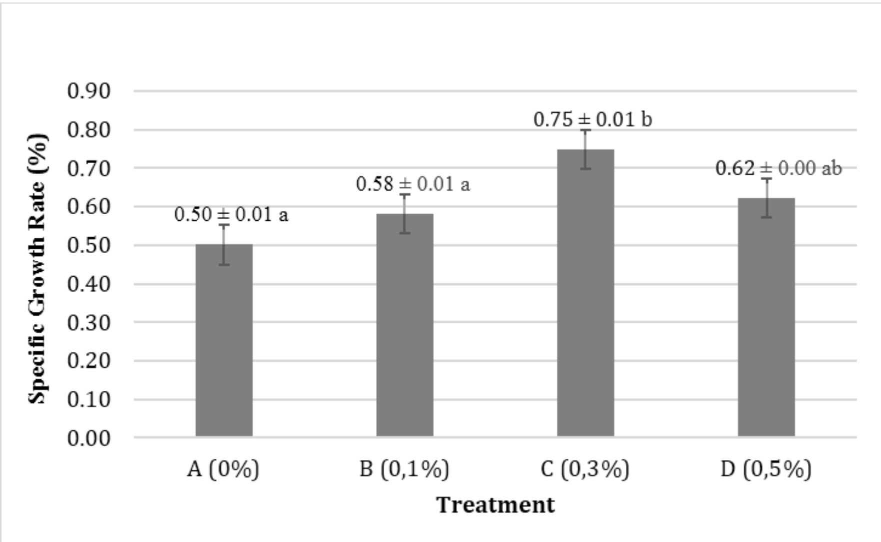

Fig. 2. Specific growth rate

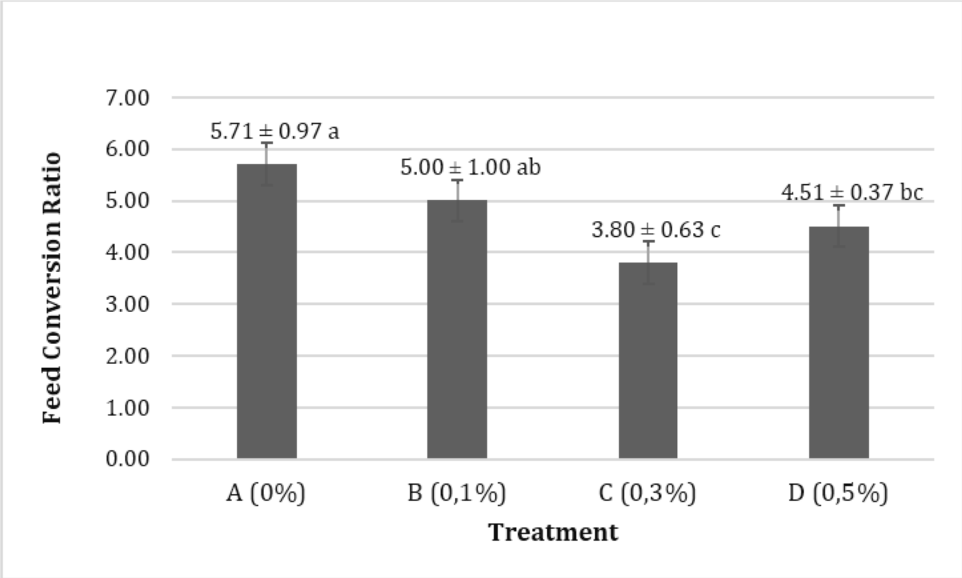

Fig. 3. Feed conversion rate chart

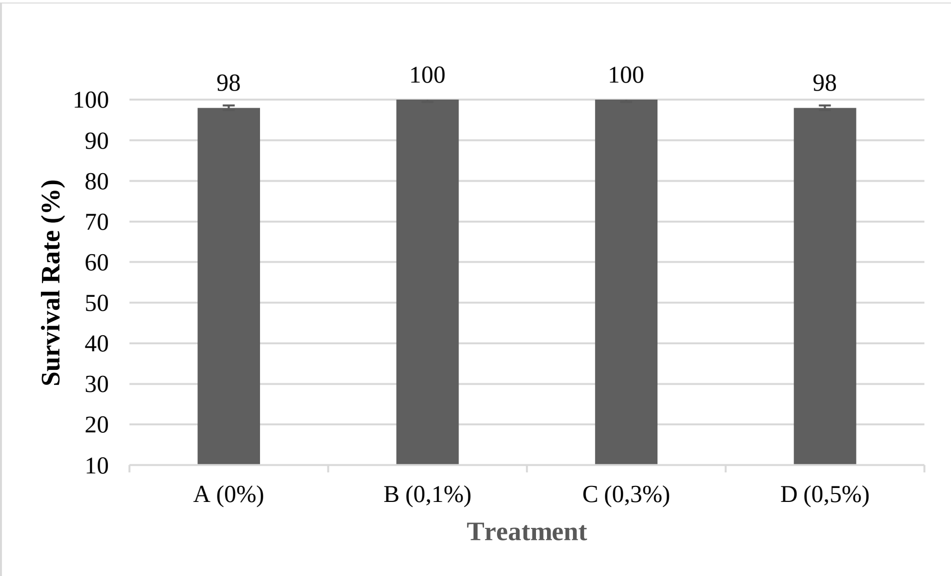

Fig. 4. Survival rate chart

Survival rate the highest was obtained in treatments B and C of 100% and the lowest in treatments A and D of 98%. If referring to SNI No. 01 – 6137 -1999 survival rate of mahseer from all treatments has a value of > 80% which is considered quite high, indicating that the mahseer gave a positive response to the feed added to potassium diformate. The results of this research can also be said to be good when compared to the results of Pasaribu research [22] with the results of survival rates in mahseer fingerlings ranging from 95.55 - 100%.

Survival rate High mahseer in this research can be obtained because it uses good quality fingerlings. The percentage of survival rate is influenced by other factors such as water and feed quality. The quality of the water in this research has met the standard for cultivating divine fish which refers to SNI no. 01 – 6137 – 1999. This research was carried out in a controlled laboratory so that the survival rate of fish is higher compared to rearing in an open place. In addition, the feed given is in accordance with the needs of the mahseer fingerlings both in terms of size, amount, frequency, and nutritional content.

Several studies have proven that the addition of potassium diformate to feed is not only able to stimulate growth but also can increase the survival rate of fish. Potassium diformate is a feed additive that can kill pathogenic bacteria which can improve fish health resulting in a high survival rate. Survival rate nilem fish (*Osteochilus hasselti*) was highest after the bacterial challenge test was performed *Aeromonas hydrophila* a value of 81.67% was obtained with the addition of 0.3% potassium diformate [23].

Immunity of fish against disease will increase with the addition of the right dose of potassium diformate [24]. The addition of potassium diformate to feed with high doses that exceed the optimum dose becomes less effective for the survival rate of fish, because the use of potassium diformate at too high a dose can disrupt the osmoregulation system of fish. The use of potassium diformate must be considered so as not to exceed the optimum dose, so that the salt level is not higher than the salt level in the fish's blood because it will cause water to leave the fish's body and salt to enter the blood which results in dehydrated fish and eventually it will die [25].

3.4 Water Quality

Water quality has an important role in supporting the success of aquaculture because it has a big influence on the survival of fish. Good water quality for aquaculture is defined as water that supports the life and growth of fish, water as a aquaculture medium has several important parameters to control during aquaculture activities, namely temperature, dissolved oxygen (DO), and degree of acidity (pH). Water quality data for 40 days of fish rearing is presented in tabular form (Table 2).

Based on measurements of water quality in the rearing medium during the research, it was still in the range of water quality standards referring to SNI No. 01 – 6137 – 1999. Water temperature affects metabolic processes in fish. At high temperatures that can still be tolerated, fish can increase their metabolic rate so that fish are more active in searching for food, while at low temperatures, the activity of living organisms decreases. Fish growth will decrease if the temperaturis low below 13°C, while at temperatures below 5°C growth will decrease rapidly and stop eating [26].

Dissolved oxygen (DO) is an important parameter that determines fish life, in waters with low oxygen content fish will easily get stressed which can eventually lead to death. Based on the results of observations of DO in the medium for fish rearing ranged from 6.26 to 6.78 mg/L. Generally, the dissolved oxygen content that is good for fish farming ranges from 4 - 9 mg per L [27]. This shows that the fish's need for DO is met during rearing. Fulfillment of DO needs during maintenance is supported by the installation of aeration as a supply of oxygen to the rearing medium.

The degree of acidity (pH) of the water during preservation is still within the quality standard range. Ideal waters for fish survival have a pH ranging from 6.5 to 8.5 [27]. At pH between 4.0 - 6.5 and pH between 9 - 11 can result in slow fish growth. When the water becomes very acidic (pH < 4) or very alkaline (pH < 11) it will generally result in fish dying.

Table 2. Water quality range value

Parameter	Unit	Range value	Quality standard
Temperature	^{0}C	23,4 – 25,1	≤ 28
pH	-	6,50 – 7,95	6,5 – 8,5
DO	mg/L	5,76 – 6,84	≥ 5

4. CONCLUSION

Based on the study's results, adding potassium diformate to mahseer feed affected the specific growth rate and feed conversion ratio but did not affect the survival rate. The optimal addition of potassium diformate to the feed is 0.3% to produce the highest SGR of 0.748%, the lowest FCR is 3.80, and SR is 100%. So it can be concluded that adding 0.3% is the best treatment.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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