



Effect of Feeding Fermented Catfish Pellets with Probiotics on Population Growth of Water Fleas (*Daphnia Magna*)

Cantika Glodia¹, Sucahyo²

^{1,2}Faculty of Biology, Universitas Kristen Satya Wacana, Salatiga, Indonesia

Email: cantikaglodiasammin@gmail.com, sucahyo2009@gmail.com

Abstract:

D. magna as feed has advantages: easy to digest, the size is in accordance with the larval mouth opening, has 42.65% protein in the form of nutrition, contains a number of digestive enzymes, does not degrade water quality and can be cultivated in bulk. In this study, commercial catfish pellets fermented with probiotics were used as feed for *D. magna*. The probiotic that is often used in aquaculture is EM4 (Effective Microorganism 4). EM4 is able to play a role in increasing protein levels and with the right dose, EM4 becomes the main determining factor in growth productivity. The purpose of this study was to determine the good duration of EM4 fermentation in population growth of *D. magna*. This study is an experimental study (CRD), using *D. magna* as a test animal, with 5 different treatments. The results of the density of *D. magna* which were given different treatments gave different results. The highest density was found in treatment 3 and the treatment that had the lowest density was in treatment 5. The results of the Nonparametric test (Friedman-Test) showed significant results ($P=0.00<0.05$). Significantly different treatments were in treatment 3 and 5, while treatment 2 and treatment 4 had no significant difference. The results of the study the highest specific growth rate obtained was in treatment 4 of 0.223%/day, then followed by treatment 3 of 0.211%/day, while other treatments were relatively lower. Thus, feeding in the form of catfish pellets fermented with EM4 on the population growth of *Daphnia magna* gave a significant effect on treatment 3 and treatment 5 with the highest specific growth rate in treatment 4.

Keywords:

EM4; *D. magna*; fermentation; catfish pellets; probiotics

I. Introduction

The world of aquaculture includes various kinds or forms of production from brackish waters, marine and fresh waters. Various kinds of aquatic organisms can be cultivated in various containers such as ponds, floating net baskets, and so on. With a closed or open system. Currently, many aquaculture activities have been developed because the need for protein food from waters is quite large. From the research results obtained show that protein derived from fish and other aquatic organisms is healthier for consumption than animal protein. It is undeniable that in the future, dependence on aquaculture products will increase due to the exploitation of the aquatic world in the fishing sector which has been under so much pressure that production in nature has been degraded on a large scale.

To obtain maximum production results from the aquaculture industry, various types of obstacles must be overcome, one of which is the provision of healthy seeds or larvae continuously (Pangkey, H. 2009). The health of the seeds or larvae is very dependent on the feed and the medium of life. Feeding that is not optimal has a major influence on the survival

and growth rate of larvae. There are two types of feed, namely natural feed and artificial feed, both of which play a role in helping the growth and nutritional fulfillment of fish larvae. Natural food is feed that is already available in nature or living organisms, both plants and animals that can be consumed by fish larvae. While artificial feed is feed that is processed from several kinds of mixtures of materials and then formed in such a way as desired (Prastya et al. 2016). The availability of natural food is often a problem. Often live food is seasonal so that at certain times it is difficult to obtain. Natural feed commonly used in hatcheries is the type of micro-organisms that live in the waters in the form of phytoplankton and zooplankton. The selection of natural feed in hatcheries is because the feed moves actively so that it attracts fish to eat it, the size is suitable for the larval mouth opening, does not affect water quality, is easily digested by the larvae because it contains digestive enzymes, and has nutritional content that is in accordance with the nutrients needed by the fish.

Daphnia magna included in the type of small crustaceans and commonly referred to as water fleas that can live in fresh water. *D. magna* is one type of natural feed that has the potential to be bred for freshwater fish hatchery activities and is a non-selective filter feeder (Darmawan. 2014). Young *D. magna* with a size of 1 mm can filter small particles up to 20-30 microns, while mature *D. magna* with a size of 2-3 mm can filter particles with a size of 60-140 microns (Mokoginta. 2003). The advantages of giving *D. magna* as natural feed for fish hatcheries include easy digestion, size according to the larva's mouth opening, 42.65% protein (Sihotang et al. 2012), contains a number of digestive enzymes: proteinase, peptidase, amylase, lipase, cellulase which functions as an exo-enzyme in the digestion of fish larvae (Pangkey. 2009), and the content of essential amino acids is quite high, feeding does not reduce water quality, and can be cultivated in bulk (Rakhman et al. 2012). Not only fish seeds for consumption of *D. magna* can also be used as ornamental fish feed. *D. magna* that lives in nature consumes feed in the form of bacteria, phytoplankton, ciliates, detritus, and so on (Zahidah et al. 2012).

In population *D. magna* is dominated by female *D. magna* which reproduce asexually. At optimum conditions, female *D. magna* can produce up to 100 eggs and can lay eggs again every 3 days. In her lifetime, female *D. magna* can reproduce 25 times, but what is often encountered is only 6 times. *D. magna* females can start reproducing after 4 days of age. In bad conditions, male *D. magna* can reproduce, so sexual reproduction occurs. The eggs produced are dormant eggs (resting eggs). Factors that cause this situation are lack of feed, low oxygen content, high population density, and low temperature (Pangkey. 2009).

Population growth of *D. magna* in a culture cannot be separated from the role of feed and the quality of its life media. The feed provided includes inorganic, organic (animal manure), phytoplankton (*Chlorella* sp.), and bacteria (Zakiyah et al. 2019). Until now, the cultivation technique of *D. magna*. A lot has been done but there are still shortcomings such as the cultivation of *D. magna* using bran feed, the drawback is that the stability of water quality is disturbed such as an increase in ammonia concentration which causes high water pH (Mubarak et al. 2009), containing 6.9% phytic acid in which this acid can bind minerals such as calcium, magnesium, zinc, and copper so that it has the potential to interfere with mineral absorption, besides that it can bind to protein, thereby reducing protein digestibility (Sihotang et al. 2012).

In a study conducted on population growth of *D. magna*, the feed used was commercial catfish pellets fermented with probiotics. The use of fish pellets in this study is because catfish pellets are easy to obtain, affordable prices, and contain high enough protein that *D. magna* can use in its growth. According to Ningsih et al. (2020) protein has the ability to provide amino acids for growth, defense, and repair of body tissues.

Fermentation in this study serves to remodel complex compounds such as carbohydrates, fats, and proteins into simple ones such as glucose, amino acids, fatty acids, and glycerol so that nutrient absorption becomes easier (Anugraheni. 2016). Probiotics themselves function as controllers of the development and population of harmful microbes so as to produce an optimal environment for beneficial microbes (Hariani et al. 2017). Not only that, probiotics are able to act as producers of antibiotics, immunostimulants, and can increase feed conversion ratios (Khotimah et al. 2016). The probiotic that is often used in aquaculture is EM4 (Effective Microorganism 4).

According to Rachmawati EM4 is able to play a role in increasing protein levels in feed. With the right dose, EM4 becomes the main determining factor in growth productivity (Abdillah. 2020). From various tests that have been carried out with the addition of EM4 as a probiotic in fisheries, it is very helpful in improving water quality in ponds by degrading organic waste in the form of leftover fish feed and depositing it and enriching microflora in water so that it can be used as a source of feed (Elpawati et al. 2015).

1.1 Destination

The purpose of this study was to determine the effect of giving catfish pellets fermented with EM4 on the population growth of *Daphnia magna*.

1.2 Formulation of the Problem

- How long is the fermentation time of Fish Pellet with EM4 which is good for *D. magna* feed?
- Does the addition of probiotics (EM4) have an effect on population growth of *D. magna*?

1.3 Scope of Problem

- Type of feed used (Catfish pellets)
- Probiotics used (EM4 Fisheries)
- Type of water used (C building water)
- Observation time (2 weeks)

II. Research Methods

2.1 Place and Time of Research

This research is an experimental study and uses RAL (Completely Randomized Design). Research on the Effect of Feeding Fermented Catfish Pellets with Probiotics on the Growth of the Water Flea (*Daphnia Magna*) population was conducted at the Aquaculture Laboratory, Faculty of Biology, Satya Wacana Christian University, Salatiga. The time of the research was from September to October 2021.

2.2 Test Animal Preparation

In this study, adult *D. magna* obtained from the Aquaculture Laboratory of the Faculty of Biology, Satya Wacana Christian University was used. Taking *D. magna*. This is done by

using a fine net (scopnet) and to make the individual catches uniform, they can be filtered again using ornamental fish nets with a diameter of 1.5 mm.

2.3 Making Feed used for Treatment

The feed used was commercial catfish pellets fermented with probiotics (EM4). Catfish pellets were crushed/mashed with a mortar and pestle, then weighed 5 grams using a digital scale and then mixed with 5 ml of EM4 then added water up to 1000 ml. The mixed feed is then put into a bottle with a volume of 1.5 L and stored in a dark place. Fermentation was carried out on feed, namely 0 days, 2 days, 4 days, and 8 days before being given to *D. magna*. For the control of feeding in the form of fish pellets which were crushed/mashed with mortar and pestle, then weighed as much as 5 grams and then mixed with 1000 ml of water. Feeding was carried out every two days as much as 100 ml/replicate. Before feeding is done, the feed is shaken first so that the particles are evenly mixed.

2.4 Maintenance Container

Daphnia will be reared in uniform media (Aquarium), 20 units with a volume of 5 L. There were 5 treatments in this study, namely feed without EM4 (Control), fermentation with EM4 0 days, 2 days, 4 days, and 8 days. each 4 replicates. The container to be used is cleaned first and then filled with 3 liters of water and aerated for 24 hours before stocking. *D. magna* adults stocked in culture containers with a density of 30 fish/L. Maintenance of *D. magna* is carried out for approximately 2 weeks.

2.5 Parameter

The parameters measured in this study were *D. magna* density, *D. magna* growth rate, DO, pH, and temperature.

- **Daphnia magna population calculation:**

The population calculation of *D. magna* was carried out by taking 100 ml of each replicate which had been homogenized evenly in the container and then placed in a petri dish with a dark base, making it easier to calculate *D. magna*. Calculation of the number of individuals is done by counting the four replications and then averaged. The result is converted in units of Individual/L.

Formula (Rahayu. 2009):

$$a = b \times p / q$$

Information:

a = Number of individuals of *D. magna* on culture media (ind/L)

b = Average number of *D. magna* from calculation repetition

p = Volume of culture media (Liters)

q = Volume of sample bottles (Liters)

- **Measurement of the growth rate of Daphnia magna:**

In measuring the growth rate of *D. magna*, population measurements will be carried out every two days by taking 100 ml of each replicate which has been homogenized evenly in the container and then placed in a petri dish with a dark base so as to facilitate the calculation of *D. magna*. The formula used is as follows (Ilham et al. 2019):

$$\mu = \frac{\ln.Nt - \ln.No}{t} \times 100\%$$

Information:

- μ = Specific growth rate (%/day)
- No = Initial population density *D. magna* (eng/L)
- Nt = Final population density *D. magna* (eng/L)
- t = Time from the beginning of the population to the end of the population (days)

- **Temperature Measurement**

Temperature measurement is carried out using a thermometer:

- The thermometer was dipped into the aquarium for each treatment then waited 5 minutes and the water temperature was recorded.

- **DO . Measurement**

DO measurement is done using a DO meter:

- DO meter is immersed in water, and observed and then recorded DO water.

- **pH measurement**

Measurement of pH is carried out using a pH meter by:

- The pH sensor rod was dipped into the aquarium, waited for 5 minutes and the pH of the water was recorded.

Observation of parameters in this study starting from DO, temperature, pH, growth rate and population calculation of *D. magna* was carried out every 3 days.

2.6 Data analysis

The data obtained were first tested for homogeneity of variance and normality. If the data meet the requirements, it will be continued with the Friedman-Test analysis (Analysis of Variance), and continued with the Tukey test. All calculations and analysis using SPSS. The physical and chemical parameters were analyzed descriptively.

III. Discussion

From the research conducted, namely the Effect of Feeding Fermented Catfish Pellets with Probiotics (EM4) on the Growth of *Daphnia magna* the following results were obtained:

3.1 Density of *Daphnia magna* in Various Treatments

Based on the results of the study, feeding pellet fermented feed added with EM4 with different fermentation times gave different results for each treatment. The highest population growth of *D. magna* was in 2 days of fermentation (Treatment 3) while on the other hand the lowest population growth was in 8 days of fermentation (Treatment 5), which can be seen in Figure 1 below:

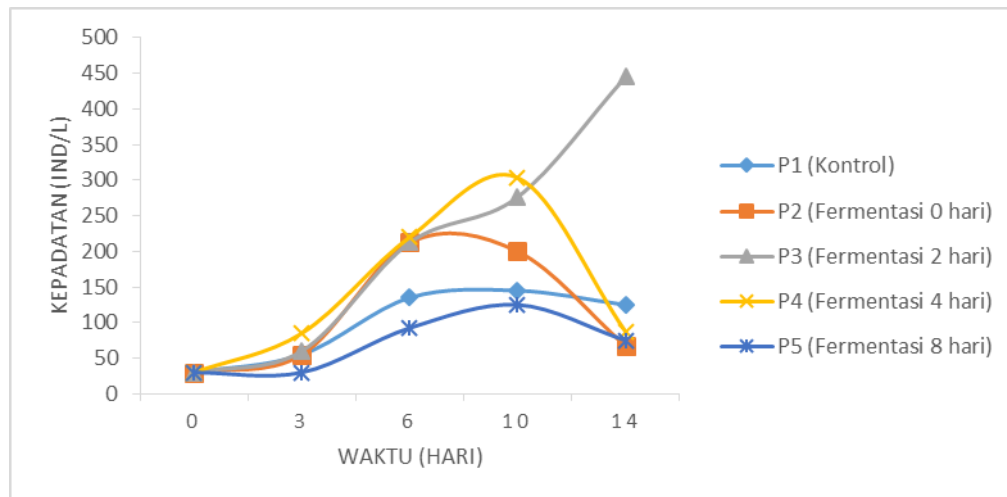


Figure 1. *D. magna*. Population Growth Pattern Chart

In this study, *D. magna* was fermented using EM4. EM4 contains *Lactobacillus* sp. (lactic acid-producing bacteria), photosynthetic bacteria, *Streptomyces* sp, cellulose-degrading fungi and yeast (*Saccharomyces cerevisiae*) (Muklisnah. 2018). This fermentation process is to increase the composition of nutrients such as carbohydrates, proteins, and reduce fat content. This is because the content in EM4 such as *Saccharomyces cerevisiae* (Hiprita et al. 2013) produces metabolite products in the form of amylase enzymes, proteolytic peptidases (Richardson. 2012). The resulting product is able to break down the components in the feed so as to produce better nutritional content, texture, and biological availability and can produce output in the form of high growth and development of *D. magna*.

The feed used was catfish pellets PF 781-1 which contained 27% protein, 4% fat, 5% crude fiber, 12% ash content, and 12% water. In Richardson et al (2012) The crude fiber content in the substrate decreased during the fermentation process. This decrease was caused by the activity of the cellulase enzyme produced by *Saccharomyces cerevisiae*. *Saccharomyces cerevisiae* can hydrolyze cellulose into glucose, thereby reducing the crude fiber content, and the decrease in crude fiber is indirectly related to the increase in carbohydrates.

The results of nonparametric statistical analysis (Friedman-Test) showed that feeding fermented pellets with EM4 added to the *D. magna* population gave significant results ($P=0.00<0.05$) (Table 1). The treatments were significantly different, namely in treatment 3 (2 days fermentation) and 5 (8 days fermentation), while in treatment 2 (fermentation 0 days) and treatment 4 (4 days fermentation) there was no significant difference. Significant growth occurred from day 6, and the peak of *D. magna* population was on day 10 (Figure 1). This rapid increase in peak population is due to the age or short life cycle of *D. magna*, *D. magna* can survive up to 12 days with optimal living media conditions and good feed availability (SriRahayuni et al. 2017).

Table 1. Tukey HSD Test Results* (Treatment)

Tukey HSDa, b

Treatment	N	Subset	
		A	B
Fermentation 8 days	20	70.5000	

Control	20	98.0000	98.0000
Fermentation 0 days	20	112.9500	112.9500
Fermentation 4 days	20	145.2500	145.2500
Fermentation 2 days	20		204.7000
Sig.		.489	.152

Table 2. Tukey HSD Test Results* (Time)
Tukey HSDa, b

time	N	Subset		
		A	B	C
Day 0	20	30.0000		
Day 3	20	57.0000	57.0000	
Day 14	20		159.9000	159.9000
Day 6	20		174.5000	174.5000
Day 10	20			210,0000
Sig.		.977	.091	.813

From the results of observations on the population density of *D. magna* reared with different culture media/treatments, it showed an increase in population growth as shown in Figure 1. The difference in population density was due to differences in the treatment of *D. magna*. The population density graph pattern forms a sigmoid curve which consists of adaptation, exponential, stationary, and death phases. In treatment 1, 2, 4, and 5 the graphic pattern formed a sigmoid curve, while in treatment 3 it still increased even until the end of the study.

The adaptation phase is the adjustment phase of *D. magna* to the culture media. This is because there is a change in the concentration of nutrients from the previous culture media to the new culture media. According to Izzah et al (2014) the lag phase shows the length of adaptation of *D. magna* to the culture media so that it affects the fast and slow growth of *D. magna*. In this study, the average adaptation phase occurred on day 0 to day 3. According to Rahayu (2010) the increase in *D. magna* population occurred after day 4, this was due to a parthenogenesis process of reproduction that produced *D. magna* individuals and took place in fertile media/cultivar conditions.

The exponential phase is a phase where population growth is accelerated, in this phase there is an increase in the number of *D. magna* individuals in utilizing media nutrients because the nutrients in the media are sufficient for their growth. In the exponential phase of the study starting on day 3 to day 6 for treatment 2, for treatments 4 and 5 until day 10, while in treatment 3 there were still additional individuals even until the end of the study. According to Erika (2022) the increase in population growth can occur because of the interaction of the influence of one factor changing when the level of another factor changes. The high population increase in treatment 3 followed by treatment 4 was thought to be because the 2-day fermentation and 4-day fermentation were able to break down pellets and EM4 into materials that could be used by *D. magna* in meeting nutritional needs, in addition to the media / place of maintenance there is no excessive change so that it is still conducive to the

survival of *D. magna*. In Syahrizal (2018) feeding with 2 days of fermentation increases the growth rate of catfish fry.

In treatments 2 and 5 the increase in population was almost the same as the increase in population in the control. It is suspected that in treatment 2, because the fermentation process is too short, EM4 has not been fully activated which results in no change in nutrition in the feed by microorganisms present in EM4, while in treatment 5, the fermentation process is quite long, namely 8 days, which affects the abundance of feed. and pH of *D. magna* cultured waters and resulted in the death of *D. magna*.

According to Darmawan (2014) in the exponential phase, *D. magna* experienced a decline in population due to the availability of feed contained in the cultivation container no longer sufficient for *D. magna* to grow optimally. The difference in exponential phase length between treatments was thought to be due to differences in nutrients in the media, abundance of feed, and the influence of water quality. Under adequate feeding conditions, *D. magna* can grow and molt until it becomes an adult individual and reproduces parthenogenesis, so that the number of individuals increases several times (Zahidah, 2012).

The stationary phase is characterized by reduced nutrients in the culture media, thus affecting the reproductive activity of *D. magna* and resulting in a reduced growth rate. In this study, the stationary phase for treatments 1, 2, 4, and 5 occurred on day 10, while in treatment 3 there was still an exponential phase or an increase in the number of individuals on day 10. The factor that influenced this phase was decreased feed availability. According to Darmawan (2014), the population growth rate of *D. magna* can decrease because the availability of feed in the cultivation media is not sufficient for the number of *D. magna*. The stationary phase is usually followed by a death phase, where in this phase the population of *D. magna* decreases. In this study, the death phase occurred on day 14 for treatments 1, 2, 4, and 5, while for treatment 3 still experienced an increase in the number of individuals. According to Izzah (2014) one of the factors that causes death in *D. magna* cultivation is the lack of nutrients in the culture media.

3.2 Growth Rate of *D. magna*

The growth rate is the maintenance of the number of individuals at a certain time in a population (Muklisna et al. 2018). The resulting data on the population of *D. magna* obtained is then tabulated into a graph. The percentage of the specific growth rate can be more clearly in the following graph:

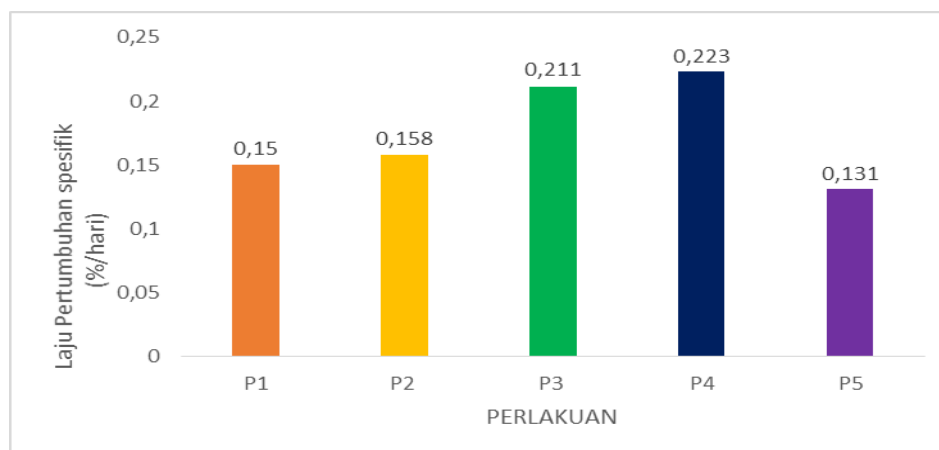


Figure 2. *Specific Growth Rate of D. Magna*

Based on the results of the study, the highest specific growth rate was in treatment 4 (4 days fermentation) of 0.223%/day, then followed by treatment 3 (2 days fermentation) of 0.211%/day, while other treatments were relatively lower, namely in treatment 2 (Fermentation 0 days) was 0.158, followed by Treatment 1 (Control) at 0.15, and Treatment 5 (Fermentation 8 days) at 0.131.

The high growth rate was suspected because the feed contained in the culture media could be utilized properly by *D. magna* so that the growth needs of *D. magna* could be fulfilled. The low growth rate occurs because the nutrient content contained in the culture media cannot meet the availability of feed in the culture media, resulting in food competition and resulting in fewer quantities (Endra et al. 2017). Another factor that led to a decrease in the number and death of *Daphnia m.* namely the possibility of inedible organic matter which eventually settles to the bottom of the culture container and becomes ammonia/toxic to *Daphnia m.* (Fahmi et al. 2021).

Factors that caused differences in growth rates between treatments were environmental factors and factors from *Daphnia m.* alone. Environmental conditions or living media of *D. magna* greatly affect the growth rate of *D. magna* as well as the availability of available feed. Other possible factors that influence the growth rate are factors originating from within the body of *Daphnia m.* such as stadia, age, size, activity, and physiological conditions in *D. magna* (Endra et al. 2017).

Figure 2. shows the difference in the growth rate of each treatment. In treatment 2 (Fermentation 0 days) it was not much different from treatment 1 (Control). It is suspected that the nutrients in the culture media are low. In Endra et al (2017) the short fermentation time results in limited opportunities for microorganisms to multiply so that the substrate components that can be broken down into cell mass will also be small but with a longer time it will provide opportunities for microorganisms to grow and develop. Factors that influence the process of increasing protein content in fermented products are the type of organic material used, the type of bacteria used as a fermenter and the length of time used in the fermentation process (Endra et al 2017).

In treatment 4 (4 days of fermentation) and 3 (fermentation of 2), it was seen that the growth rate was higher than the control. This is presumably with 4 days of fermentation and 2 days of fermentation capable of breaking down pellets and EM4 into materials that can be used by *D. magna* in meeting nutritional needs, in addition to the media/place of maintenance there is no excessive change so that it is still conducive to the survival of *D. magna*. In Syahrizal (2018) feeding with 4 days of fermentation provides the best survival for catfish fry.

In treatment 5 (8 days of fermentation) had the lowest growth rate compared to other treatments. It is suspected that treatment 5 had the longest fermentation time which caused a very drastic change in the nutritional content of the feed and decreased pH. The longer fermentation time also causes the pH level to decrease (Acid), with the lower pH causing the microorganisms in EM4 to not work optimally. The by-product of fermentation in the form of alcohol in this study is also expected to be quite high. In Lestari et al (2018) stated that the increase in alcohol content was in line with the longer fermentation caused by fermenting fungi (yeasts) which developed and metabolized to produce alcohol. One type of fermented fungus found in EM4 is *Saccharomyces cerevisiae*. According to Azizah et al (2012) *Saccharomyces cerevisiae* produces invertase and zymase enzymes to break down monosaccharide and disaccharide sugars into alcohol and CO₂. The invertase enzyme will

break down lactose into simple sugars, which are then converted into ethanol by the enzyme zymase.

3.3 Water Quality Maintenance Media *D. magna*

The water quality of the maintenance media is one of the important factors in the survival of *D. magna*. In this study the measurement of water quality includes measurements of DO (dissolved oxygen), temperature, and pH. Water quality data can be seen more clearly in the following table:

Table 3. Dissolved Oxygen, pH, and Temperature in the Maintenance Media of *D. magna*

Treatment	Parameter					
	Temperature (0C)		pH		DO (mg/L)	
	Beginning	End	Beginning	End	Beginning	End
P1 (Pellet)	25.2	27.6	7.0	6.3	6.2	4.0
P2 (0 Day Fermentation)	25.4	27.7	7.0	6.2	5.8	3.8
P3 (2 Days Fermentation)	25.3	28.5	6.9	6.0	6.1	2.7
P4 (4 Days Fermentation)	25.1	27.7	6.7	6.0	6.5	3.4
P5 (8 Days Fermentation)	25.2	27.5	6.6	5.0	6.4	3.5
Eligibility According to Library	24-280C Darmawan (2014)		6.5-9 Rahayu et al (2012)		>3 mg/L Mubarak et al (2009)	

In this study, the water quality range for *D. magna* cultivation was dissolved oxygen (DO) 6.5-2.7 mg/L, temperature 28.5-25.10C, and pH 7.0-5.9.

Oxygen plays an important role as an indicator of water quality, because oxygen plays a role in the oxidation and reduction of organic and inorganic materials. With the fulfillment of the need for oxygen can increase the rate of growth and prevent mortality in organisms. Oxygen has an important role in the survival of *D. magna*. According to Sri Rahayuni et al (2017) the optimal dissolved oxygen level for the growth of *D. magna* is >3 mg/L. In this study, dissolved oxygen levels in treatments 1, 2, 4, and 5 were considered optimal compared to dissolved oxygen levels in treatment 3. The decrease in oxygen levels was due to the highest density of *D. magna* in this treatment so that oxygen levels decreased/poor. In conditions of poor oxygen levels *D. magna* will synthesize hemoglobin, consequently *D. Magna* is bright red. In this study also did not use aeration so that the oxygen obtained by *D. magna* was only in the container. Utilization of oxygen by bacteria / microbes can also reduce dissolved oxygen.

Temperature is an abiotic factor that affects the increase and decrease in the activity of organisms such as reproduction, growth, and death. Outside the optimum range, *D. magna* tends to be unable to reproduce (dormant). A good temperature for the growth and reproduction of *D. magna* according to Darmawan (2014) is 24-280C. During the study, the observed temperature fluctuations ranged from 25-28.50C, where this temperature was included in the range of *D. magna*, which grew normally and had no significant effect.

pH affects the level of water fertility, this is because pH affects the life of microorganisms. The optimum pH for the growth of *D. magna* according to Rahayu et al

(2012) is in the range of 6.5-9. During the study the pH was in the range of 7.0-5.0. In treatments 1, 2, 3, and 4, the pH was optimum for the growth of *D. magna*, while in treatment 5 the pH was quite low (acidic), namely 5.0. This is because the feeding in the form of fermented pellets took longer than other treatments, namely 8 days. The longer the feed fermentation time, the more acidic the feed will be. This causes a decrease in the pH of the water.

IV. Conclusion

Based on the research that has been done, it can be concluded that the feeding of fermented catfish pellets with probiotics (EM4) with different fermentation times has a significant effect on the population density of *D. magna*. The length of fermentation that has a significant effect on population growth is 2 days fermentation and 8 days fermentation. Significant growth occurred from day 6, and the peak of *D. magna* population was on day 10. The highest specific growth rate was in treatment 4 (4 days Fermentation) of 0.223%/day.

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