

## THE EFFECT OF GIVING ONION (*Allium Sativum*) EXTRACT TO TREAT THE SEED OF TILAPIA (*Oreochromis Niloticus*) BACTERIAL INFECTED *Aeromonas hydrophila*

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### ABSTRACT

MAS disease (*Motile Aeromonas Septicemia*) is a disease caused by *A. hydrophila* bacteria that attack tilapia and other freshwater fish species. The use of antibiotics and chemicals in the continuous prevention and treatment of this disease can damage the aquatic environment and can cause bacterial resistance to antibiotics, so antibiotic use becomes ineffective. The study used a Complete Randomized Design (RAL) with an experimental method consisting of 5 treatments and 3 repeats. Treatment in this study is (P1) without soaking garlic extract and without infected bacteria *Aeromonas hydrophila*, (P2) without soaking garlic extract and infected bacteria *Aeromonas hydrophila*, (P3) soaking garlic extract with a concentration of 1% and disinfection of bacteria *Aeromonas hydrophila*, (P4) immersion of garlic extract with a concentration of 1.5% and infected bacteria *Aeromonas hydrophila*, (P5) soaked garlic extract with a concentration of 2% and infected with *Aeromonas hydrophila* bacteria. The results showed that the SR value of tilapia fish in each treatment had values ranging from 0.0% to 66.7%. Relative percent survival (RPS) tilapia seeds in each treatment have values ranging from 0.0% to 66.7%. The conclusion of this study is that giving garlic extract with different concentrations has a real different influence on survival rates and relative percent survival (RPS).

### INTRODUCTION

MAS (*Motile Aeromonas Septicemia*) is a disease caused by *A. hydrophila* bacteria that attacks tilapia and other freshwater fish species (Giyati, 2000). This MAS disease has clinical symptoms, namely reduced fish appetite, bleeding in the gills, there are wounds on the fish's body, scales are detached, tail fin is damaged, the stomach is enlarged due to fluid, and swelling of the lymph tissue, liver and kidneys can occur. cause a mortality rate of 80% (Tantu *et al.*, 2013). *A. hydrophila* bacteria is one of the bacteria that attacks fish from seed to adult stages. The seed stage is a death-prone phase, because at this stage the fish's immune system is not fully developed (Hanif *et al.*, 2004).

The use of antibiotics and chemicals in an effort to prevent and treat *A. hydrophila* bacterial infection can continuously damage the aquatic environment and poison

fish and result in bacterial resistance to antibiotics, so that the use of antibiotics becomes ineffective (Irawan *et al.*, 2003). . An alternative that can be used to replace the use of antibiotics is to use natural ingredients, one of which is garlic (*Allium sativum*) where garlic is known to contain *Allicin* which is an active substance that is able to kill bacteria and has anti-inflammatory power. The use of garlic as an herbal ingredient in aquaculture activities can cure fish infected with bacteria into one of the considerations for its use as an antibacterial agent as in previous studies, namely the use of garlic extract to treat Siamese catfish (*Pangasius hypopthalmus*) seeds infected with *Aeromonas hydrophila* bacteria (Muslim, 2009) and administration of garlic extract with different concentrations on the survival of tilapia infected with *Streptococcus iniae* (Zahraturrahmah, 2018).

This study aims to determine the effect of garlic extract (*Allium sativum*) on bacterial disease in tilapia fry (*Oreochromis niloticus*) caused by *A. hydrophila* bacteria. The benefit of this research is to provide scientific information about the effect of garlic extract in reducing infection in tilapia fry (*Oreochromis niloticus*) caused by *A. hydrophila* bacteria to increase their survival.

## METHODOLOGY

### Materials and tools

The materials used in this study were tilapia, garlic extract, *Aeromonas hydrophila* isolate, aquades, artificial feed, and fresh water. While the tools used in this study were jars, scales, aerators, seser, petri dishes, microscopes, small buckets, cameras, tissue, DO meters, thermometers, pH meters, test tubes, Erlenmeyer, oasis needle, autoclave, spatula, magnetic stirrer, hotplate, refrigerator, blender, laminar, and incubator.

### Research methods

The test animals used were tilapia seeds, tilapia seeds used were seeds measuring 4-5 cm. Maintenance was carried out for 14 days in a 15-liter jar with a volume of 10 liters of water. This study used RAL (Completely Randomized Design) with experimental methods with 5 treatments and 3 replications, namely treatment (P1) using no soaking garlic extract and without being infected with *Aeromonas hydrophila* bacteria; treatment (P2) without soaking garlic extract and infected with *Aeromonas hydrophila* bacteria. , treatment v (P3) used garlic extract immersion with a concentration of 1% and was infected with *Aeromonas hydrophila* bacteria , treatment (P4) used immersion garlic extract with a concentration of 1.5% and was infected with *Aeromonas hydrophila* bacteria , treatment (P5) used garlic extract immersion with a concentration of 2% and infected with *Aeromonas hydrophila* bacteria.

### Data analysis

Data that has been collected during the research process was analyzed using ANOVA (analysis of variance) at a 95% confidence level through the SPSS program and Duncan's further test was carried out to determine the best treatment.

## RESULT

### Survival Rate (SR)

The results of observing the survival rate of tilapia fry treated with garlic extract during rearing can be seen in Figure 1

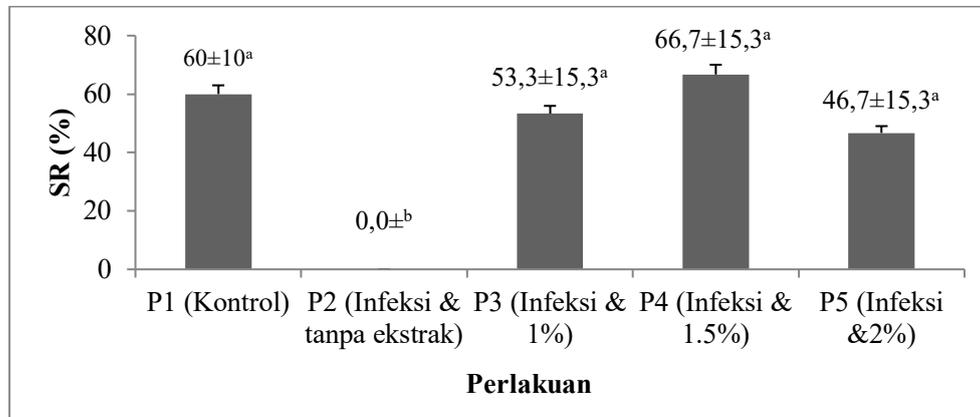


Figure 1. Survival rate of tilapia fry

### Relative Percent Survival (RPS)

The results of observations of RPS of tilapia seeds that were given treatment in the form of garlic extract during maintenance can be seen in Figure 2.

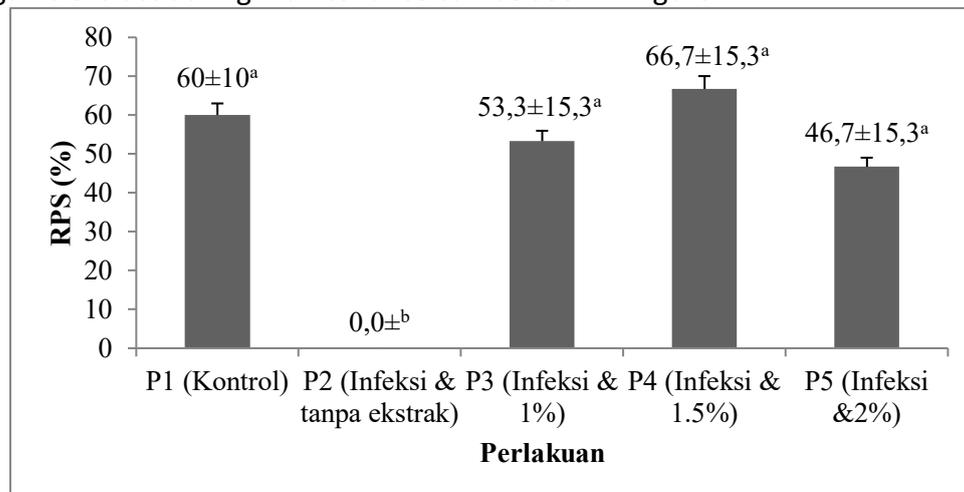


Figure 2. Relative Percent Survival (RPS) of tilapia seeds

### Clinical Symptoms During Infection



Figure 3. Scales and caudal fin detached

## Water Quality

The water quality parameters observed were temperature, pH, and DO. The water quality from the beginning of infection, treatment and maintenance was no problem for the fish, and the water quality was normal. Water quality parameters can be seen in table 5 below:

Treatment	First Week			Second Week		
	DO (ppm)	pH	Temperature (°C)	DO (ppm)	pH	Temperature (°C)
P1	5,1-5,3	7,2-7,3	28,1-28,2	5,1-5,2	7,2-7,4	28,3-28,4
P2	4,2-5,2	7,1-7,2	28,1-28,2	5,1-5,5	7,2-7,5	28,3-28,6
P3	5,0-5,1	7,2-7,3	28,2-28,4	5,0-5,2	7,2-7,3	28,2-28,4
P4	5,1-5,3	7,2-7,3	28,1-28,2	5,0-5,2	7,2-7,3	28,3-28,4
P5	4,4-5,0	7,0-7,5	28,1-28,3	4,3-4,7	7,0-7,4	28,1-28,3
Literature	3-6 ppm	5-9	25-30°C	3-6 ppm	5-9	25-30°C
	Gufuran (2010)	Khairuman (2013)	Rukmana (2008)	Gufuran (2010)	Khairuman (2013)	Rukmana (2008)

## DISCUSSION

In this study, the administration of soaking garlic extract on the survival rate of tilapia fry with a concentration of 1.5% (P4) gave the highest value of 66.7% followed by administration without soaking garlic extract and without bacterial infection (P1). ) with a value of 60%, garlic extract with a concentration of 1% and bacterial infection (P3) with a value of 53.3%, garlic extract with a concentration of 2% and bacterial infection (P5) with a value of 46.7% and a value of The lowest was found in the treatment without soaking garlic extract and bacterial infection (P2) with a value of 0.0%.

Based on analysis of variance, it showed that soaking garlic extract with different concentrations had a significantly different effect or significant ( $p < 0.05$ ) on the survival rate of tilapia, so Duncan's further test was carried out to determine the best treatment. Further test results showed that P4 was not significantly different from P1, P3 and P5, but significantly different from P2.

The high survival rate of tilapia fry at P4 (1.5%) is thought to be due to the 1.5% concentration of garlic extract containing the compound *Allicin* which functions as an antibacterial so that it can increase the survival rate of tilapia fry. This is in accordance with the statement of Wibowo (1989). *in* Muslim *et al.*, (2009), explained that garlic (*A. sativum*) has an active compound, namely *Allicin* where *Allicin* is an active substance that can kill bacteria and can cleanse the blood of toxins produced by bacteria. According to Handayani & Siswanto (2019), it was also explained that the use of garlic extract to treat *A. hydrophila* bacteria that attacks tilapia seeds (*Oreochromis niloticus*) has a 66.67%-90% survival rate of tilapia seeds. Garlic can be used to control pathogens, enhance immune responses, and have a positive effect on the survival rate of fish (Aniputri *et al.*, 2014). According to Fatimah (1992) *in* Murjani (2011) that the survival of fish is highly dependent on the adaptability of fish to food and the environment, fish health status, stocking density, and sufficient water quality to support growth.

The low survival rate of tilapia fry in P2 treatment was presumably because garlic extract was not given but still infected with bacteria which caused tilapia fry susceptible to death due to the absence of antibacterial. It is characterized by state fish suffering from ulcerative lesions on the fish's body, irrespective scales, tail fins and the fish dying geripis this mendadak. Hal accordance with pernyataan Angka (2001), stating that the fish are attacked by bacteria *A. hydrophila* show clinical symptoms such as damage to the caudal fin, pelvic fin, and pectoral fin, then sores (ulcers), flatulence and if the pelvic fins are not treated immediately,

bacteria will spread in the blood vessels and continue to the internal organs of the abdomen causing swelling (drops) and causing a yellow discharge.

While the P5 treatment had a poor survival value with a value of <50%. This is in accordance with the statement of Husen (1985) *in* Mulyani (2014) that the survival rate of 50% is good, 30-50% survival is moderate and less than 30% is not good. This is presumably because the high concentration of garlic extract given to P5 resulted in stress and death of fish. This was shown during the study on P5 tilapia seeds had abnormal movements and were always on the surface of the rearing media and had the most mortality compared to other treatments soaked using garlic extract. This is in accordance with Anderson's (1974) statement *in* Lukistyowati *et al* (2007) which states that if there are factors that disturb fish, be it parasites, chemicals or physical stimuli that trigger stress in fish. In addition, the presence of fish jumping up and down uncontrollably also indicates that the fish feel uncomfortable with their environment.

In this study, the value of the *Relative percent survival* (RPS) of tilapia fry with garlic extract immersion with a concentration of 1.5% (P4) gave the highest value of 66.7% followed by administration without soaking garlic extract and without soaking it. bacterial infection (P1) with a value of 60%, garlic extract with a concentration of 1% and bacterial infection (P3) with a value of 53.3%, garlic extract with a concentration of 2% and bacterial infection (P5) with a value of 46, 7% and the lowest value was found in the treatment without soaking garlic extract and bacterial infection (P2) with a value of 0.0%.

Based on analysis of variance, it showed that soaking garlic extract with different concentrations had a significantly different effect ( $p < 0.05$ ) on the *relative percentage survival* of tilapia seeds, so Duncan's further test was carried out to determine the best treatment. Further test results showed that P4 was not significantly different from P1, P3 and P5, but significantly different from P2. The high cure rate of tilapia fry at P4 with a concentration of 1.5% as much as 66.7% showed the highest yield. This is because the concentration of 1.5% is the best treatment and can cure tilapia fry infected with *A. hydrophila* bacteria. Giving garlic extract can increase the fish's body resistance to fish infected with *A. hydrophila* bacteria. This is in accordance with the statement of Syamsiah and Tajudin (2003) Giving garlic extract for the treatment of *A. hydrophila* bacteria in catfish can increase survival, this is presumably due to the influence of the active substances contained in garlic, namely *allicin* and *scordinin* where *allicin* could kill bacteria. In this case, the bacteria *A. hydrophila*. *Allicin* contained in garlic can also kill gram-positive and gram-negative bacteria and serves to increase the body's resistance to prevent the entry of germs.

The low success rate of recovering tilapia fry at P2 with a value of 0.0% was due to the treatment that the fish were infected with bacteria and without garlic extract so that all tilapia seeds died and at P5 with a value of 46.7% it was not good with a value of <50% on the success rate of fish recovery. This is in accordance with the statement of Ellis (1988) *in* Lukisyowati (2004) which states that the success rate to obtain an effective relative survival is when the RPS is 50%. by 52%.

At P5 with a concentration of 2% with a low recovery rate of tilapia fry due to the high concentration given to tilapia fry, causing tilapia fry to experience clinical symptoms of sluggish fish movement, swimming around, and always swimming to the surface of the water. This is because the administration of garlic extract with a concentration that is too high is less effective and can kill fish seeds. This is in accordance with the statement of Pelezar and Chan (1986) *in* Sari *et al*, (2014) who argue that the higher the antimicrobial concentration used, the faster it will kill bacteria, but the use of too high a concentration is less effective in

treatment because it can kill fish and bacteria. It is also less economical to use. The excessive soaking of the garlic extract caused the fish to be unable to survive, because the fish when soaked in a weak state were infected with *A. hydrophila* bacteria so that this condition was exacerbated by stress.

Clinical symptoms observed during infection were known that in general the symptoms that occurred in fish were fish began to swim around, decreased appetite, loss of swimming balance, some fish began to limp. Fish began to become infected with the characteristics of swimming in circles, swimming to the surface. , fish fins begin to fall off, scales come off, sores or ulcers and bleeding in the operculum. This is also in accordance with the results of research Figure (2001) which found that fish infected with *A. hydrophila* bacteria showed clinical symptoms such as damage to the tail fin, pelvic fins, and pectoral fins. , then sores (ulcers), flatulence and if the pelvic fins are not treated immediately, the bacteria will spread in the blood vessels and continue to the internal organs of the abdomen causing swelling (drops) and causing a yellow discharge. Furthermore, the fish died on the 4th day after infection.

Symptoms of healthy fish after being treated for 30 minutes and kept for 14 days can be seen that the observation of clinical symptoms in fish after being treated with garlic extract on the first day of fish has not shown any change, this is because the garlic extract has not worked in the fish's body so that the resulting mortality occurs on the first day because the bacteria in the fish's body are still working. On day 4 to day 14 the fish in P4 (concentration 1.5%), followed by P3 (concentration 1%) and P5 (concentration 2%) showed symptoms of fish that gradually disappeared, their body color was brilliant compared to fish in the treatment. P2 (Bacterial infection and without the addition of extract), the movement starts to be agile and the fish start to eat. This is stated by (Lukistiowati, 2004 *in* Muslim *et al.* 2009) that catfish given garlic extract can increase the body's resistance of the fish, so that the movement becomes agile, and the color is brilliant.

Water quality parameters during infection, treatment, and maintenance were still at optimal levels and could still be tolerated by tilapia fry. This can be seen in table 5. That is, the temperature parameters measured in the first week in treatment P1 ranged from 28.1-28.2 °C, in the second week 28.3-28.4 °C and did not differ much in treatment. P2, P3, P4 and P5. The DO measurement in the first week in all treatments ranged from 4.4-5.3 ppm while the second week measurement ranged from 4.3-5.2 ppm. And for pH measurements in all treatments in the first week it ranged from 7.1 to 7.3 and in the second week it ranged from 7.2 to 7.5. The infection of *A. hydrophila* bacteria in tilapia during the maintenance of water quality values in this study was still at a normal level so that it could support the survival rate of tilapia. This is in accordance with the literature obtained, namely temperatures of 25-30 °C (Rukmana, 2008), pH 5-9 (Khairuman, 2013), and DO 3-6 ppm (Gufran, 2010).

## CONCLUSION

Based on the results of the study, it can be concluded that the addition of garlic extract can increase the survival rate (SR) and relative percent survival (RPS) of tilapia fry that were challenged with *A. hydrophila* bacteria

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