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DURATION OF INOCULATION KAPPA-KARAGEENAN FOR EFFECTIVE ENDURANCE OF DUMBO CATFISH (*Clarias* sp.) AGAINST INFECTION *A. hydrophila* BACTERIAL

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Abstract

High mortality in developing catfish MAS (Motile Aeromonad Septicaemia) is the main issue in cultivation activities. It is caused by the low durability of the fish body and the poor environmental factors that triggered the disease. The disease is caused by the bacteria namely *A. hydrophila*. An alternative that can be done to reduce the high mortality on fish farming is *Clarias gariepinus*. It is used against the attacks of disease control with the use of an *immunostimulant*. The use of macro algae *immunostimulant* has been widely performed as *Kappaphycus alvarezii* type that is known to contain *k-carrageenan* that is believed it can increase the robustness of the fish. The optimal provision doses should be paid attention as *immunostimulant* is used. The duration of inoculation protection to achieve the optimal *immunostimulant* is also essential in giving the *immunostimulant*. The purpose of this research is to examine the duration of the inoculation of κ -carrageenan which is effective against the survival and the growth of dumbo catfish (*Clarias* sp.). This research consists of six treatments and three replicates. They are K (+): feeding without *k-carrageenan*, testing challenge with the bacteria *A. hydrophila* K (-): feeding without *k-carrageenan*, PBS-injected. PB1: Giving *k-carrageenan* every day for five weeks. PB7: Giving *k-carrageenan* for seven days at week 1. PB14: Giving *k-carrageenan* for 14 days on Sunday I and II. PB21: Giving *k-carrageenan* for 21 days on Sunday I, III and V. The results showed the issues of *k-carrageenan* with a grant of 14 days duration during the period of five-week maintenance of being able to provide 71% survival of absolute growth so to the catfish increased to 28g.

Keyword: *kappa-carrageenan*, *Clarias hydrophyla*, *Aeromonas* sp.

INTRODUCTION

Dumbo Catfish (*Clarias* sp.) is a freshwater fish that is widely and intensively cultivated in almost all regions of Indonesia. This is because the *Clarias gariepinus* fish is one of the leading commodities. It is also very popular, and has good market prospects. One way to meet the needs of both local and international markets is the intensive cultivation with high

stocking solid and intensive feeding and the use of the water repeatedly. This practice is potentially causing stresses to the fish, and it will affect fish's health conditions. High mortality in developing MAS (Motile Aeromonad Septicaemia) catfish is the main issue in the cultivation activities. It is caused by the low durability of the fish body, and the poor environmental factors that triggered the disease caused by the bacteria *A. hydrophila*.

An alternative can be done to reduce the high mortality rate in *Clarias gariepinus* fish farming. It is used against the attacks of disease control with the use of *immunostimulant*. The use of *immunostimulant* has attracted attention and was done as a more environmentally friendly approach to disease control fish (Raa 1996; Sakai 1999; Peddie et al. 2002). Materials can be *immunostimulant* extracted from seaweed. *Kappa-carrageenan* is a type of *carrageenan* which is extracted from *C. Ocellatus Kappaphycus Alvarezii*, *Gigartina* and *Radula* (Renn 1997).

Provision of optimal doses should be paid attention to *immunostimulant* used (Anderson, 1992). In addition, the duration of inoculation protection to achieve the optimum *immunostimulant* is also essential in giving *immunostimulant* (Couso et al. 2003). Suyati (2010) is stating that the inoculation of *kappa-carrageenan* through repeated injection until four times can increase the survival of the *Clarias gariepinus* fish. It has the highest IE post-Christmas 5.77% ± 93.33 infection bacteria *A. hydrophila*. Inoculation *immunostimulant* is sustainably needed to provide better immune ability. So that this research was conducted with the aim of testing the effect of inoculation of κ -*carrageenan* orally to enhance resilience of dumbo catfish (*Clarias* sp.) against *Aeromonas hydrophila* bacteria.

RESEARCH METHODS

The Research Preparation

The fish to be tested is *Clarias gariepinus* weighing 15-20 grams. It comes from catfish farmers in Parung, Bogor. Prior to the use in the experiment, catfish reared in a maintenance that comes with aerator. During the fish rearing, it was fed twice a day with a dose of 3%/bb/day. The water used in the experiment is filtered and deposited. Subsequently, the water is accommodated in fiber and is aerated. The treatment container used is with the dimension of 60x30x30 cm sized aquarium equipped with aeration. Before it is used, the aquarium is cleaned and sterilized with Chlorine 30 ppm for 24 hours, then it is rinsed with fresh water, and it is dried. Then the aquarium is filled with water and cleaned and aerated. The fish was adapted in an aquarium for 1 week before treatment.

The provision of a suspension of the bacteria *A. hydrophila*

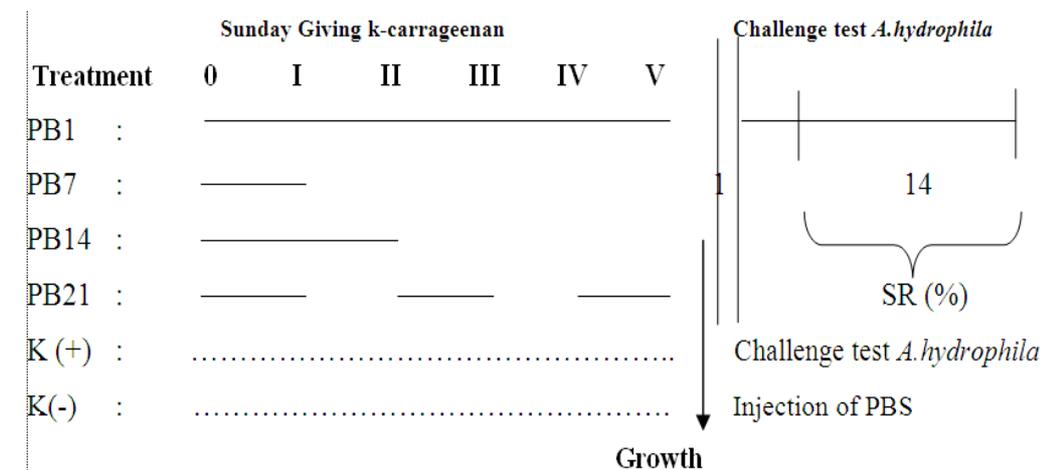
The bacteria *A. hydrophila* Fish Health Laboratory is obtained from Bogor Agricultural University. Before it is used to test the enhanced bacteria, it is challenged the virulence by injecting a healthy fish back on which is further isolated back from clinical symptoms indicating the fish is infected with *A. hydrophila*.

The duration of the inoculation of k-Carrageenan for effective Resistance against bacterial infections Catfish *Aeromonas hydrophila*. This research consists of six treatments and three replicates. The dose used is 10 g/kg feed. The treatment used is the period of inoculation *k-carrageenan* in the feeding as follows:

- K (+) : Feeding without k-carrageenan, tested challenge with the bacteria *A. hydrophila*
- K (-) : Feeding without *k-carrageenan*, PBS-injected
- PB1 : Giving *k-carrageenan* every day for five weeks
- PB7 : Giving *k-carrageenan* for seven days in week I
- PB14 : Giving *k-carrageenan* for 14 days in week I and II:
- PB21 : Giving *k-carrageenan* for 21 days in week I, III and V

Maintenance was carried out for five weeks after that, a challenge test was conducted with the bacteria *A. hydrophila* 10⁸ CFU/fish except the negative control group (K-) is injected only with PBS. The observed parameters were the fish survival rate and its growth. Fish survival is observed from day 1 to day 14 after a challenge test.

Research Scheme as follows



- : Sunday giving k-carrageenan
- : Sunday feeding without carrageenan
- 1-14 : Day observation

Data Analysis

The design of this experiment is a laboratory experimental model, using a complete randomized design (RAL). It consists of 6 treatments: three times and repeats. The experiment is to find out the differences of each treatment provision of *k-karageenan* through the feeding; then, it is analyzed by using ANOVA with its diversity. If there are differences among the treatments; subsequently, it is continued to further tests using SPSS program, Duncan ver. 17.

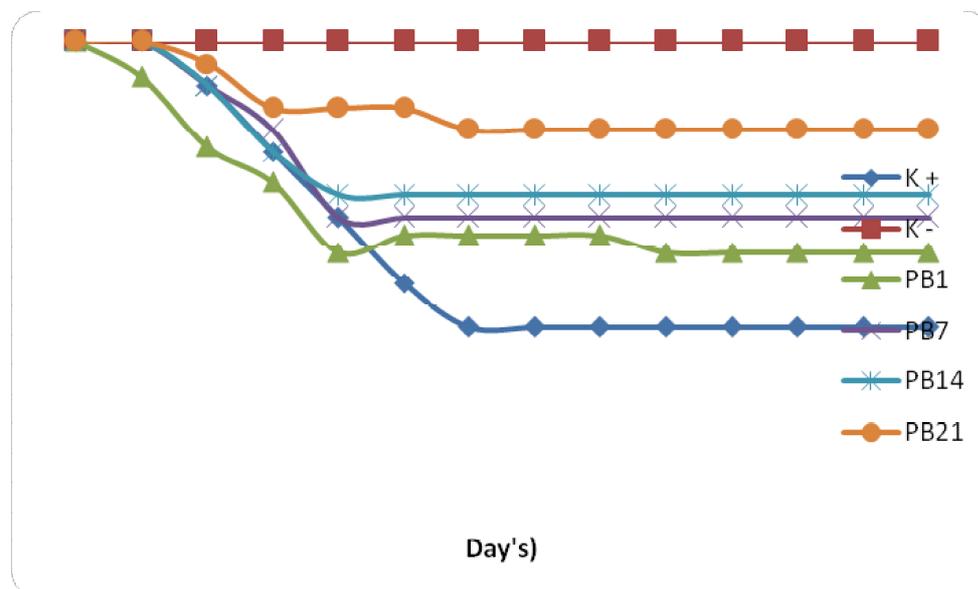
RESULTS AND DISCUSSION

The survival of catfish

Observation of catfish's survival rate with the duration of inoculation *k-carrageenan* for 37 days after testing the bacteria *A. hydrophila* challenge can be presented in Figure 1.

The stricken fish by *A. hydrophila* bacteria usually shows symptoms such as: the fish's body becomes darker, the ability to swim is declining, the fish's eyes are damaged and rather prominent, scales to lift, the whole fins are damaged, the gills are red-white, the fish is gasping for air on the surface, making it difficult to breathe, the fish's skin becomes invisible, and the subsequent bleeding occurs followed by cuts of stomach ulcers, ulcers-mackerel (dropsy) and when the surgery is conducted then it will be heart bleeding, kidneys and spleen (Kordi Ghufran mosque and 2004).

The results showed that the catfish's survival rate based upon the maintenance treatment is by giving *k-carrageenan* with different durations. It suggests the treatment of PB1 67.00%, PB7 364.99%, and it amounts to \$ 364.99 PB14% and 83.67% of PB21. And, it has positive control of 50.00% and negative control of 100%. The test statistics results shows that the treatment of PB1, PB7, PB14, and PB21 influence and different real ($P < 0.05$) of the positive control. Negative controls give the survival rate higher of 100%. This is because the treatment is not injected with bacteria *A. hydrophila*. However, it is injected with PBS (*Phosphate Buffer Saline*). The death of catfish is caused by the presence of inflammation, ulcers and hemorrhage. According to et al. (2000) that *A. hydrophila* produced a product that is toxin causing the blood to have haemolysis on in the catfish that leads to its death.



Picture 1: The fish survival on post-bacterial infection *A. hydrophila*

The Catfish Growth

The growth is increasing in length, weight and volume by units of time. Based on the results of the absolute weight measurement is added on each treatment, and it is presented in table 1 below.

Table 1. The increase in absolute fish weight in frequency treatment of *K-karagenan*

Treatment	The Early (g)	Weight (g)	The Absolute (g)
K	18	30	12
PB1	17	33	16
PB7	17	37	21
PB14	18	46	28
PB21	17	34	18

Based on the results of the study, it shows that the treatment by giving *k-carrageenan* makes good impacts against growth when it is compared with controls. According to Vilela-silva et al. (2008), polysaccharide sulfate is known to function as growth factors, coagulation factors and selecting binding partners. It also functions in the fertilization. When a non-protein in feeding energy is able to be put in the good use, then the protein feeding will be used for the growth optimally (Pappenheim and Hardy 2002). The use of *k-carrageenan* as an adhesive on the larval *Channa Striatus* feeds is providing the growth and feeding

efficiency (Nakagawa et al. 2007). Absolute highest weights are added in provision treatment of *k-carrageenan* fourteen days (PB14) is resulting to 28 g, subsequently successful PB7 treatment is amounting to 21g, PB21 treatment is amounting to 18 g, PB1 treatment is resulting to 16 g, and the control (K) is amounting to 12 g.

CONCLUSIONS AND SUGGESTIONS

Conclusion

Inoculation of kappa-karagen with the duration inoculation twice in a month is able to increase the catfish's growth, and provides the survival to 71%.

Suggestions

Making advanced research about the champion, and needing to test the digestibility of kappa-carrageenan on *Clarias gariepinus* fish.

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