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Effect of Probiotic Dosage on Growth and Survival Rate of Tapah, stripped *Wallago leeri* Reared in Laboratory Conditions

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Abstract

The study was conducted to determine the effects of probiotics on growth ad survival rate of Tapah (*Wallago leeri*) in laboratory condition for a period of 60 days under five treatments with four replications. Treatments were P0 (0 ml/m^3 probiotics), P1 (3 ml/m^3 probiotics), P2 (5ml/m^3 probiotics), P3 (7 ml/m^3 probiotic) and P4 (9 ml/m^3 probiotics). The juveniles with initial weight of 22.5 g, which caught from Kampar River were reared for 60 days in 60 L aquaria at 10 ind. per aquarium. Each aquarium was equipped with medium aeration. Fish treated in P2 demonstrated significantly ($P \leq 0.05$) higher average weight gained, specific growth rate, percentage weight gain and survival rate than control, lower and higher dosage. Plasma cell Protein concentration was ranged from 3,83 to 5.43 %.

Keywords: Probiotics, tapah, growth, survival,

INTRODUCTION

Stripped wallago catfish, *Wallago leeri* local name ikan tapah, is one of the biggest freshwater catfish found across the rivers, reservoirs and in connected watersheds of the Thailand through the Malaysia Peninsula, to the inlands of Borneo and Sumatera in Indonesia (Talwar and Jhingran, 1991, Kottelat and Widjanarti, 2005). It is a bony fish, belonging to the family Siluridae, grows to about 150cm, weighing more than 85 kg (Kottelat et al. 1993; Roberts, 1989). Adults inhabit large streams and rivers (Ng, 1992), Fry occur at the mouth of small streams connected to larger rivers, where the bottom is muddy and with overhanging vegetation (Kottelat and Widjanarti, 2005; Ko, 2008).

Due to overfishing of wild populations and high demand of this species in domestic market, sharp decrease of the wild population in the last decade has been occurred. Therefore, aquaculture has been a solution to prevent the further population decreasing of this species. Aquaculture has become an economic activity of great importance around the world. Aquaculture's contribution to world food production, raw materials for industrial and pharmaceutical use, and aquatic organisms for stocking or ornamental trade has increased dramatically.

With the increasing intensification and commercialization of aquaculture production, disease is a major problem in the fish farming industry (Bondad-Reantaso et al. 2005). According to Browdy(1998), one of the most significant technologies that evolved in response to disease control problems is the use of probiotics. In 1991, Porubcan [1991a, 1991b] documented the use of Bacillus spp, to test its ability to increase productivity of Penaeus monodon farming and to improve water quality by decreasing the concentrations of ammonia and nitrite. Probiotics that currently used in aquaculture industry include a wide range of taxa-from Lactobacillus, Bifidobacterium, Pediococcus. Lake Streptococcus and Carnobacteriumspp. Bacillus, Flavobacterium, cytophage, Pseudomonas, Alteromonas, Aeromonas, Enterococcus, Nitrosomonas, Nitrobacter and Vibrio spp., and Yeast Saccharomyces, Debaryomyc (Irianto and Austin 2002; Sahu et al., 2008;Aquarista, *et.al.*,2012;Hemaiswarya et al., 2013). Probiotic administrations have been widely applied via water routine or feed additives (Moriaty 1998 ;Skjermo and Vodstein 1999) with either single or a combination of probiotics or even a mixture with prebiotics or other immunostimulants (Hai and Fotedar 2009).Several experiments recommended probiotics bacteria for increasing fish production and improving the health of fish by controlling pathogen bacteria [Gatesoupe FJ (1999) - Silvi S, Nardi M, Sulpizio R, Orpianesi C, Caggiano M, et al. (2008)].

Iribarren et al (2012) stated that the use of probiotic bacteria is one of the internal solutions to reduce environmental damage due to the accumulation of wastes in aquaculture media, produce optimal growth and feed efficiency, and reduce production costs. Thus application of probiotic is urgently required for environmental friendly aquaculture. This research was conducted to study the potential doses of probiotic to increase production of *Wallago leeri* culture through booster system.

MATERIAL AND METHOD

Place of study and period. . The experiment was conducted in the Aquaculture Technology Laboratory, Aquaculture Department, Faculty of Fisheries and Marine, University of Riau for a period of 60 days from 15 July to 15 September 2019.

Experimental fish and rearing aquarium. Tapah fingerlings were collected from the fisherman who caught the fish in the connected watershed of Kampar rivers. The fingerlings were transported to the Lab. Before experiment, the fingerlings were acclimatized to the aquarium water and feeding for one week. Twenty previously prepared glass aquarium measured 60x60x30 cm were used as rearing containers.

Each aquarium was filled in 60 L (0.006 m^3) of filtered ground water aerated with medium aeration and stocked 10 individual. The initial mean body weight and length of was approximately 22.5 g and 16.5 cm respectively.

Probiotc bacteria application and feeding protocol. Probiotic used in this experiment was booster multi-cell which contain *Nitrosomonas*sp, *Nitrobacter*sp, dan *Baccillus*sp bacteria and applied as water pro-biotic.. Application of probiotic was done every 7 days or 8 times and the doses used were as recommended by the fish booster center. During the experiment the fish was fed on trace fish two times a day ad-satiation.

Research procedure and data collection. The study used a completely randomized design (CRD) with 5 treatments namely treatment P_0 = control dose 0 ml/m^3; P_1 = probiotic dose 3ml/m^3 ; P_2 = probiotic dose 5 ml/m^3; P_3 = 7ml/m^3; and P_4 = 9ml/m^3. each with 4 replications. Before treated, the fish were first adapted for one week in 20 glass aquarium (60 x 30 x 30 cm) with a density of 10 ind. aquarium^{-1} . Each aquarium was supplied with an aerator and recirculation system using small submersible water pump. Uneaten food and fish feces were removed once a day by syphoning. Water replacement was also carried out every 2-3 days as much as 30% of the water volume. After acclimatization, the density of fish in each aquarium was stocked at 5 individuals. Fish were fed with trace fish ad-satiation with a with frequency of 2 times a day (08.00 am and 16.00 pm). Probiotic was applied every 7 day: 1^{th} day, 8^{th}day, 15^{th}day; 22^{n}day; 29^{th}day, 36^{th}day; 43^{rd}day, 50^{th}day.

The fingerlings were weighed at the beginning and at the end of the experiment to assess the growth performance in terms of specific growth rate SGR=%/day), percentage growth weight gain and survival rate.

Specific growth rate (SGR) is calculated using the following formula:

$$SGR = \frac{Ln\ (Mean\ Final\ Weight)\ - Ln\ (Mean\ Initial\ Weight)}{60} \times\ 100$$

Ln = natural log

Weight Gained (WG,g) = Mean final weight – Mean initial weight

Average Daily Growth (ADG, g) = (Mean Final Weight – Mean Initial Weight)/60

$$Percentage\ Weight\ Gain\ (\%)(PWG) = \frac{Weight\ Gained}{Initial\ Body\ Weight} \times\ 100$$

Evaluation of the survival of the fingerlings at 60 days rearing period

$$Survival\ Rate\ (\%) = \frac{Number\ of\ fish\ alive\ in\ 60\ days}{Number\ of\ fish\ Initially\ stocked} \times\ 100$$

Data analysis. Analysis of variance was applied to evaluate the effect of different probiotic dose on weight gain, SGR,, SR while the different effect between dose was analyzed by Duncan's Test using SPSS 24 for Windows. Comparisons were made at $p < 0.05$.

RESULTS AND DISCUSSION

The growth parameters of Tapah catfish *W. leeri* juveniles treated with different doses of water-probiotic are presented in Table 1. The application of probiotic in water significantly increased growth performances up to 0.5 ml/L and decreased at the higher dose (Table 1.)

Table 1: Growth performances and survival of Tapah, Wallago leeri juvenile treated with different doses of booster probiotics

Parameters	Probiotic dose (ml/m^3)				
	Control (P0)	3 (P1)	5 (P2)	7 (P3)	9 (P4)
Weight gain (g)	80.89	100.59	110.95	103.16	99.26
Average daily growth (g)	1.35	1.68	1.85	1.72	1.65
Spefcific growth rate (%/d)	1.67	1.91	2.02	1.93	1.89
Percentage weight gain (%)	359.5	447.1	493.1	458.5	441.2
Percentage Survival (%)	70	85	90	90	80
Protein of blood plasma %	3.83	4.08	5.43	5.23	4.39

The highest final weight was found in fish treated in P2 (5 ml/m^3) (133.45±2.41 g) compared to those in P0 (103.39±1.49 g), P1 (123.09±2.03 g), P3 (125.66±1.83 g), and P4 (121.76±1.55 g). Individual weight gain (g) of Tapah in different dose treatments was 80.89 g, , 100.59 g., 110.95 g, 103.16 g and 99.26 g in P0, P1, P2, P3 and P4, respectively. Probiotic dose of 5 ml/m^3 (100.95 g) was significantly higher than control, lower (3 ml/m^3) dose and higher doses (7 and 9 ml/m^3).

Weight gain in % of Tapah Wallago catfish in different treatment of probiotic dose was 359.5, 447.1, 493.1, 458.5 and 441.2 % in P0, P1, P2, P3, and P4 respectively. This value was slightly higher compared to that other catfish like Pabda fish (*Ompok pabda*) (Jinia et al. 2019), *Mystus cavasius* (Ali et al. 2018), which were ranged from 257.9 % to 380.94%.

The specific growth rate (SGR) was ranged from 1.67 to 2.02 %/day. Tapah juvenile treated in 5ml/m^3 was significantly the highest (3.02 %/d) compared to those in control (1.67%/d), lower dose (3 ml./m^3, 1.915/d) and higher doses (1.93 and 1.89%/d).

The survival rate of Tapah was ranged from 70 to 90%. The addition of probiotic in the rearing water could significantly improve the survival of tapah up to 20 %. There was no significant differences in survival rate of Tapah juvenile among the probiotic doses treated. The protein of blood plasma was significantly ($P \leq 0.05$) higher (5.43%) compared to control and other dose treatments.

Application of probiotic through water of tanks or ponds may also have an effect on fish health by improving several qualities of water, since they modify the bacteria composition of the water and sediments (Ashraf 2000; Venkateswera ,2007). When probiotics are applied in culture water they multiply and over grow the pathogenic organism present in the water. (Chandrakala, and Soundharanayaki, 2017) Beside this Venkateswara (2007) reported that probiotic bacteria are generally called bacteria which can improve the water quality of aquaculture and inhibit the pathogens in water thereby increasing production. This experiment proved that probiotic doses of 5 ml/m^3 treated in the water (water probiotic) gave the highest growth and survival. The growth ot Tapah juvenile decreased at probiotic doses higher than 5 ml/m^3 (7 and 9 ml/m^3) (Fig. 1 and 2). This probably was due to the overdose administraton. According to Sakai (1999) overdosage administrations of probiotics can induce immune- suppression of continuous responses of nonspecific immune systems, A probiotic dosage may bring positive and negative results to different receivers, whose responses to different probiotic levels have been observed (Panigrahi *et al*. 2004; Bagheri *et al*. 2008). A high dose did not result in a greater level of protection (Perez- Sanchez *et al*. 2013). Appropriate probiotic levels depend on the probiont species, fish species and their physiological status, rearing conditions and the specific goal of the applications (Merrifield *et al*. 2010b).

The water probioics contain multiple strains of bacteria like *Bacillus acidophilus*, B. subtilis B. lecheniformis, Nitrobacter sp, Aerobacter and Sacharomyces cerevisiae. Application of probiotic through water of tanks and ponds may also have an effect on fish health by improving several qualities of water, since they modify the bacteria composition of the water and sediments (Ashraf 2000; Venkateswera ,2007). When probiotics are applied in culture water they multiply and over grow the pathogenic organism present in the water. (Chandrakala, and Soundharanayaki, 2017) Beside this Venkateswara (2007) reported that probiotic bacteria are generally called bacteria which can improve the water quality of aquaculture and inhibit the pathogens in water thereby increasing production.

According to the published literature, probiotics as water additive could have several favorable effects on finfish aquaculture, although two main advantages have been especially emphasized: (i) the ability to control the quality of water by bioremediation, and (ii) the biocontrol of pathogens through antagonistic effects (Liu, and Han, 2004).

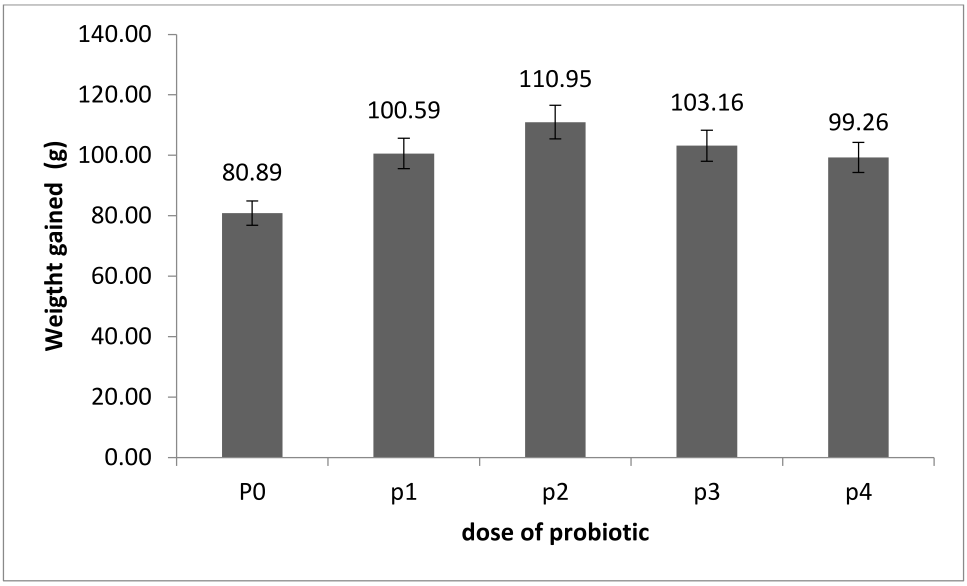

Fig. 1. Weight gained of Tapah (*Wallago leeri*) juvenile treated at different dosage of probiotic (P0= 0 ml; P1=3ml; P2=5ml; P3=7ml; P4= 9ml/m^3)

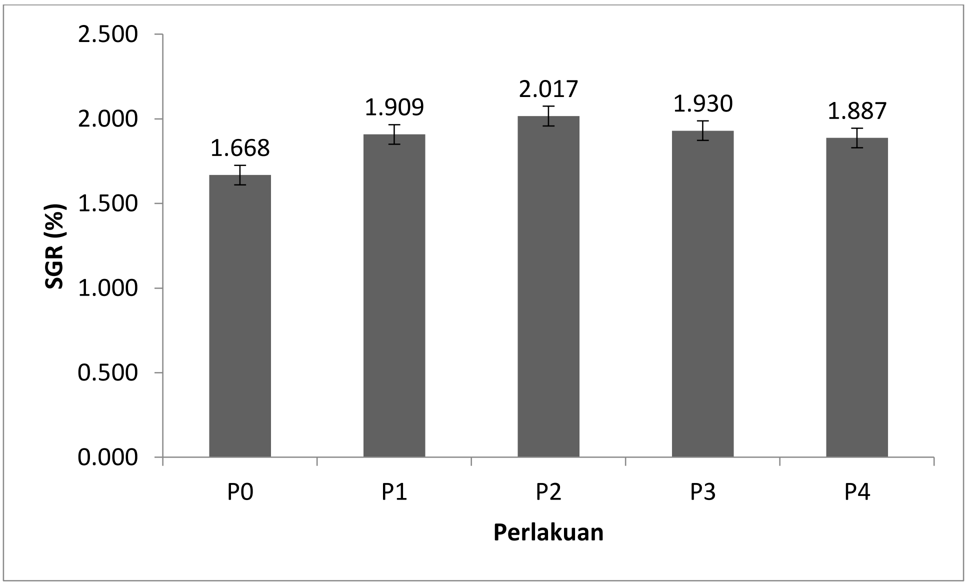

Fig. 2. Specific Growth Rate of tapah (*Wallago leeri*) treated at different dosage of probiotic (P0= 0 ml; P1=3ml; P2=5ml; P3=7ml; P4= 9ml/m^3)

CONCLUSION

Administration of probiotics in water gave a significant role in increasing the growth and survival of Tapah catfish (Wallago leeri). In term of weight gained, SGR (%/day), and survival rate (%), P2 treatment performed the best growth performance. But among probiotics treatment were not significantly different ($P>0.05$) in survival rate. .. On the basis of growth performance and survival rate 5 ml/m^3) probiotics (P2) is optimum for water treatment of Tapah *Wallagoleeri* An increase level of probiotics in water may not promote the growth and survival rete of Tapah *wallago leeri* in this study.

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