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Effect of Probiotics on Survival Rate and Growth Performance of *Clarias gariepinus*

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ABSTRACT

Influence of probiotics (mixture of *Lactobacillus* sp. and *Bacillus subtilis*) on survival rate, growth performance and feed conversion ratio of African catfish *Clarias gariepinus* fingerling was examined. Two treatments, namely with (trial) and without probiotics (control) supplementations were performed. In both the treatments, commercial feed containing 31-33% crude protein was applied twice a day for 85 days. For the trial, the probiotics mixture was supplemented in both the feeds (10^5 cells/g feed) and tank medium (10^7 cells/mL). The fish were cultured into 3 m^3 tank. Each trial and control was done in duplicate. The results exhibited that the survival of African catfish was remarkably higher in probiotics treatment than the control ($P<0.05$). The growth performance [specific growth rate (SGR) and daily gain (DG)] and feed conversion ratio (FCR) were significantly ($P<0.05$) better in fish treated with probiotics compared with those in the control. To sum up, the supplementation of probiotics in feed and culture medium improved the survival rate, growth performance and feed conversion ratio of *C. gariepinus*.

INTRODUCTION

The *Clarias gariepinus* (African catfish) is one of the popular and essential commodities of aquaculture in Indonesia. Directorate General of Aquaculture (2015) reported that an increase in catfish production by 369% from 14,475 tons in 2009 to 679,379 tons in 2013. However, this production has not met the target set of 450% by the Ministry of Maritime Affairs and Fisheries. In order to increase the production of catfish culture, intensive aquaculture activities have been applied by the most farmers (Afrilasari et al. 2016). However, intensification of aquaculture caused some problems such as water quality degradation and enhancement of the possibility of the disease attack (Piedrahita 2003, De Schryver et al. 2008). Moreover, Fakhri & Sunarmi (2017) reported that the common problem in catfish culture is low feed digestibility that causes high feed conversion ratio.

Supplementation of probiotics in feed and rearing medium is one of the ways to overcome the obstacles faced in intensive catfish culture. The probiotics are live bacteria, yeast and fungi, which once supplied in sufficient quantities, give a beneficial effect on the host health (FAO/WHO 2002). Probiotics have been commonly applied in aquaculture in order to prevent disease outbreaks, increase immune responses, providing nutrients and enzymatic contributions, and keeping good water quality (Qi et al. 2009). In addition, a few researchers reported that the probiotics application help minimize the cost of aquaculture production through enhancing the biomass and efficiency of feed utilization (El-Haroun 2007, Al-Dohail et al. 2009, Dennis & Uchenna 2016).

In aquaculture, probiotics supplementation not only improves the gastrointestinal health, but also improves the water quality (Gomez-Gil et al. 2000). Omenwa et al. (2015) demonstrated that the supplementation of *Lactobacillus plantarum* and *Pseudomonas flourescens* in tank water results in the highest survival rate of *C. gariepinus* juvenile by 96.22%. The probiotics addition in intensive aquaculture to stimulate intestinal health is increasing (Ige 2013). Among probiotics bacteria, *Lactobacillus* and *Bacillus* strains have been widely used in aquaculture because of their capability to produce extracellular enzymes and high antagonism activity (Banerjee & Ray 2017). Therefore, the use of probiotics (mixture of *Bacillus subtilis* and *Lactobacillus* sp.) through, both feed and culture medium, is a relatively new approach to improve productivity in African catfish culture. This study was proposed to analyse the effect of probiotics mixture (*B. subtilis* and *Lactobacillus* sp.) supplementation in feed and rearing medium on the survival rate, daily gain, specific growth rate and feed conversion ratio of the fish *C. gariepinus*.

MATERIALS AND METHODS

Probiotics Preparation

The probiotics culture was prepared by inoculating

Table 1: The composition of medium for probiotics culture.

No.	Materials	Compositions
1.	Probiotic starter (L)	1
2.	Coconut water (L)	3
3.	Palm sugar (kg)	3
4.	Fresh milk (L)	3
5.	Bean sprouts extract (L)	3
6.	Water (L)	3

Table 2: Growth performances and feed conversion ratio of *C. gariepinus* with (trial) or without (control) supplemented with probiotics.

Group/treatment	Trial	Control
Initial weight (g)	5.18±0.20	5.16±0.17
Final weight (g)	83.28±0.52	79.34±0.88
DG ($g.d^{-1}$)	0.92 ± 0.004	0.87±0.009
SGR (% d^{-1})	3.27 ± 0.039	3.22±0.028
FCR	0.92 ± 0.03	1.22 ± 0.02

probiotics starter containing *Bacillus subtilis* and *Lactobacillus* sp. into medium composed of organic carbon, micronutrients, and minerals for microorganisms growth (Table 1). The probiotics culture was incubated under the anaerobic condition for five days period to obtain optimal density of approximately 1×10^9 cells/mL.

Experimental Conditions

This study was conducted at intensive catfish culture in Buring Village, Kedungkandang District, Malang, Indonesia. Catfish culture was carried out for 85 days (from June 2017 to September 2017). The study consisted of two treatments, namely with (trial) and without probiotics (control) supplementations. The probiotics was supplemented in both feed and rearing medium. The study was conducted in duplicate using 3 m^3 in the volume of tanks with continuous aeration. Each tank was stocked with 800 catfish fingerlings of size 7±1 cm or 5±1 g. During the study period of 85 days, catfish were fed with commercial feed containing 31-33% crude protein. For probiotics supplementation, the commercial feed was fermented with probiotics ingredients at approximately 10^5 cells/g feed for three days in airtight plastic containers at room temperature (25-30°C) prior to the feeding. In addition to the fermented feed, probiotics (10^7 cells/mL) were also supplemented in culture water at 5 mL/m^3 along the culture periods. On the other hand, the fish of control tanks were fed without any probiotics supplementation. The fish were fed twice per day at 09:00 a.m. and 15:00 p.m. at satiation. Water quality parameters in *C. gariepinus* culture, including water temperature range (27.50°C-29.80°C), pH range (6.72-7.15) and dissolved oxygen range (4.24 mgL^{-1} - 4.94 mgL^{-1}). At the end of the study, several performances including survival rate, specific growth rate, daily gain and feed conversion ratio were calculated.

Survival Rate

Survival rate (SR) was calculated according to Lauzon et al. (2010) as follows:

$$SR\ (\%) = (\text{number of final fish} / \text{number of initial fish}) \times 100$$

Specific Growth Rate

Specific growth rate (SGR) was estimated as follows (Zhou et al. 2010):

$$SGR\ (\%\ d^{-1}) = \frac{\ln \text{final weight (g)} - \ln \text{initial weight (g)}}{\text{time (days)}} \times 100$$

Daily Gain

The daily gain ($g.d^{-1}$) (DG) was calculated based on Yanbo & Zirong (2006) as follows:

$$DG\ (g.d^{-1}) = \text{Final weight (g)} - \text{Initial weight (g)}/\text{time}$$

Feed Conversion Ratio

Feed conversion ratio (FCR) was determined based on Lauzon et al. (2010) as follows:

$$FCR = \text{offered feed (g)} / \text{total weight gain (g)}$$

Statistical Analysis

Data were analysed by *t*-test at $p<0.05$ significant level. Homogeneity and normality tests were performed prior to the *t*-test analysing. Data were analysed by SPSS 24.0 program.

RESULTS

Survival rates (SR) in trial and control are presented in Fig. 1. After 85 days, the survival rate of 93.89±0.4% in the treated tank was remarkably greater than in control with the survival rate of 83.33±1.6% ($P<0.05$). It indicates that probiotics (*Bacillussubtilis* and *Lactobacillus* sp.) supplementation considerably improved the survival of *C. gariepinus* culture.

Data of daily gain (DG), specific growth rate (SGR) and feed conversion ratio (FCR) were reviewed in Table 2. At the start of the study, there was no significant difference in initial weight between two treatments ($P>0.05$). Nevertheless, the mean final weight of the trial (83.28±0.52 g) was outstandingly better ($P<0.05$) than the control (79.34±0.88 g).

Daily gain (DG) and specific growth rate (SGR) values in the trial were greatly higher compared with the control ($P<0.05$). The supplementation of probiotics in feed and rearing medium also resulted in the best feed conversion

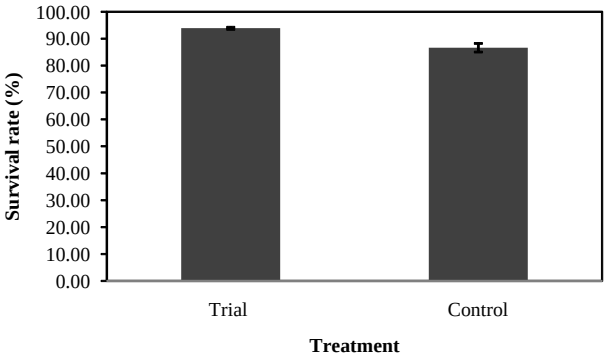

Fig. 1: The survival rate of African catfish after 85 days culture period with (trial) or without (control) supplemented with probiotics.

ratio (FCR) in comparison to the control ($P<0.05$). It exhibited that the probiotics significantly improved the growth performances and enhanced the feed utilization of *C. gariepinus* culture.

DISCUSSION

Supplementation of probiotics (mixture of *Bacillus subtilis* and *Lactobacillus* sp.) in feed and tank medium resulted in survival rate, growth performance and feed conversion ratio greater than that of the control without addition of probiotics (Fig. 1 and Table 1), indicating that the probiotics supplementation improved the economic efficiency of African catfish culture (*C. gariepinus*). This result agreed with Yanbo & Zirong (2006) in common carp and Putra et al. (2017) in African catfish. Al et al. (2008) stated that the supplementation of *L. acidophilus* and *B. subtilis* mixture improved the growth in *Tilapia nilotica*. El-Haroun (2007) also found that the application of probiotics (Biogen) of *Bacillus* in the fish diet increased the specific growth rate and reduced the feed conversion ratio of African catfish. The lower FCR values resulted in probiotics treatment indicated that probiotics supplementation enhanced feed utilization of African catfish. Moreover, Dennis & Uchenna (2016) studied the influence of probiotics on the survival rate of *C. gariepinus* larvae and found better survival in probiotics treatment (mixture of *L. acidophilus*, *B. subtilis* and *L. bugaricus*) compared with the control diet.

Probiotics are living microbial cell supplements which beneficially improve digestive process and health by optimizing the population and formation of microbial community inside the gastrointestinal in host (Bomba et al. 2002). The use of probiotics in aquaculture have main beneficial effects that are enhancement of animal growth (Ghosh et al. 2008, Merrifield et al. 2010), controlling of disease through improvement of immunity system (Nayak 2010, Qi et al. 2009) and exclusion of pathogen (Ibrahem 2015). Welker & Lim (2011) explained that probiotics could trigger the digestive enzymes production or via other modifications in the intestinal environment that can boost digestion and ultimately enhanced the fish growth performance. In addition, Bomba et al. (2002) suggested that improvement of digestibility and feed utilization in fish treated with probiotics, probably caused by the ability of probiotics to promote the beneficial microorganism population, microbial enzymatic activity and improve intestinal microbial balance in the gastrointestinal system. With regard to survival rate, the higher survival was found in probiotics treatment because probiotics have the ability to suppress the cell density of pathogenic bacteria through competition for nutrients and other resources (Queiroz & Boyd 1998). Forestier et al. (2001) have reported that Lactobacillus strain was able to attach in intestinal mucus and produce antimicrobial peptides. Similar characteristics were also found by Cherif et al. (2001) and Cladera-Olivera et al. (2004) for *Bacillus* strains.

CONCLUSIONS

In conclusion, the addition of probiotics mixture (*Bacillus subtilis* and *Lactobacillus* sp.) in feed and rearing medium positively increased the survival rate, growth performance and improved feed efficiency in *C. gariepinus* culture. Use of *B. subtilis* and *Lactobacillus* sp. with the concentration of 10^5 cells/g feed and 10^7 cells/mL in water were effective and efficient to stimulate fish production.

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