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Assessment on Impact of Dietary Probiotic Supplementation on Growth Indices of Mono-Sex Tilapia (*Oreochromis niloticus*) Cage Culture at Dakatia River, Chandpur, Bangladesh

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Abstract: This study was carried out in experimental cages in the Dakatia River, located about 3 km southeast of the Bangladesh Fisheries Research Institute (BFRI), Riverine Station (RS), Chandpur campus, to evaluate the effect of dietary probiotic supplementation on growth performance and survival of male mono-sex Tilapia (*Oreochromis niloticus*) during April 2013 to August 2013. A commercial probiotic named "Biotics" used at the rate of 2 and 3 g/kg feed for treatments 2 and 3 respectively with no probiotic for treatment 1. Nine net cages were divided into three treatments each having three replicates. Average stocking weight was $33.66\pm6.23g$ and stocking density was $50/m^3$ in each cage. Final weight in probiotic treated cages (T-2 and T-3) was significantly (P<0.05) higher than control (T-1). Final weight gain, Average Daily Gain (ADG) and production per cage were the highest in T-2, 237.82 ± 13.69 , 1.98 ± 0.11 and 476.79 ± 27.41 kg respectively and found to be significantly different from both T-1 and T-3. FCR was the best in T-2 (1.11 ± 0.047) and significantly different from T-1 but not from T-3.

Key words: Probiotic Supplementation • Oreochromis niloticus • Growth Indices • Survival • Net Cage • Dakatia River

INTRODUCTION

Tilapia, because of their enormous adaptability to a wide range of physical and environmental conditions, ability to reproduce in captivity, relative resistance to handling stress and disease-causing agents compared to other cultured finfish species, good flesh quality, feed on a low trophic level and excellent growth rate on a wide variety of natural and artificial diets, are the most abundantly cultured species worldwide. They are presently cultured in virtually all types of production systems, in both fresh and salt water and in tropical, subtropical and temperate climates [1]. They are the second most cultured freshwater fish in the world (After carps). However, they are increasingly recognized as the species of choice for intensive aquaculture and are likely to become the most important cultured fish in the world [2]. Several species of tilapia are being cultured commercially, but Nile tilapia, Oreochromis niloticus and

various hybrids are the predominant culture species worldwide. Among the various types of culture practices of tilapia, cage culture is recently found to be very common worldwide.

In Bangladesh, the Bangladesh Fisheries Development Corporation (BFDC) was the pioneer in experimental cage culture of Nile tilapia in Kaptai Lake, Rangamati in the 1980s. However, no production data is available. Subsequently CARE, Bangladesh conducted grow-out trials of Bangladesh Fisheries Research Institute (BFRI) Super GIFT strain in cages in the Meghna River lagoon area near Munshiganj in 1990s. CARE also implemented a CAGES project for more than 5 years with limited success as potential livelihood options in different sites in Bangladesh. Recently, cage culture of mono-sex tilapia is being practiced successfully in Dakatia River of Chandpur district and in some channels of the Meghna River in Laxmipur district.

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A major problem associated with cage culture operations in the aforementioned areas of Bangladesh is the increased susceptibility of fish to infectious diseases. This problem found to be very much serious over the last couple of years which caused huge losses for the cage culture farmers of the region. Traditionally, antibiotics and chemicals have been used to treat diseases in aquatic animals [3]. However; the use of antibiotics to cure bacterial infection and prevent fish mortality in aquaculture is becoming limited as pathogens develop resistance to the drugs [4, 5]. Further, beneficial bacterial flora are killed or inhibited by orally administered antibiotics. As a result the use of antibiotics in animal production, including aquaculture, is increasingly under public scrutiny and criticism in most developed countries. Consequently, there have been considerable interests in recent years to evaluate the feasibility of using nonnutrient dietary additives, particularly probiotics which are a live microbial feed, beneficially affects the host animal by improving its intestinal balance as supplements to improve growth [6, 7] and in some cases as a mean of replacing antimicrobial compounds [8]. On the other hand their effectiveness to mitigate the effects of stress, resulting in a greater production [9]. Current probiotic applications and scientific data on mechanisms of action

indicate that non-viable microbial components act in a beneficial manner and this benefit is not limited just to the intestinal region [10]. Multiple ways exist in which probiotics could be beneficial and these could act either as a singly or combination with several probiotics. These include: inhibition of a pathogen via production of antagonistic compounds, alteration of enzymatic activity of pathogens and nutritional benefits such as improving feed digestibility and feed utilization [11-13]. The present study is an endeavor to find out the effect of dietary probiotics on growth performance, feed utilization efficiency and survival of cage reared mono-sex tilapia.

Methodology

Rearing Conditions and Experimental Design: The experiment was carried out in Dakatia River, Chandpur Sadar, Bangladesh. Nine net cages were installed in a suitable place near the bank of Dakatia River, located about 3 km southeast of the Bangladesh Fisheries Research Institute (BFRI), Riverine Station (RS), Chandpur campus (Fig. 1). Sizes of net cages were $6\times3\times2$ m each, made of plastic net (Mesh size 2.5cm) and knotless polyethylene net (Mesh size 1.1 cm). Frames of the cages were made by GI pipe and were floated by means of 250 liters size plastic drums.

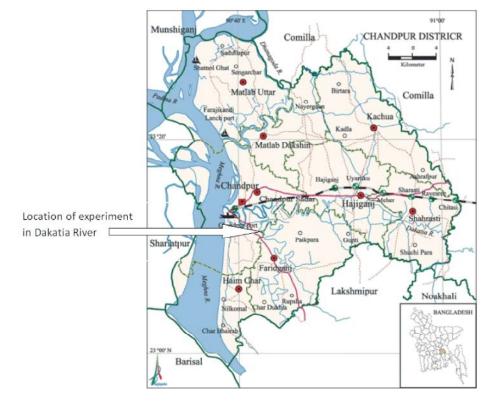


Fig. 1: Location of experimental cages (Source: Banglapedia)

Feed type	Protein (Min %)	Fat (Min %)	Fiber (Max %)	Ash (Max %)	Moisture (Max %)	Pellet size (mm)
Starter	31	4	6.5	11	11	1
Grower	28	4	7	11	11	2
Finisher	26	4	8	11	11	3

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Table 2: Feeding strategy for different types of feeds					
Culture period	Types of supplied feed	Feeding frequency	Feed rate (% of body weight)		
1-30 days	Starter	2 times	4		
31-60 days	Grower	2 times	3		
61-90 days	Grower	2 times	2		
91-120 days	finisher	2 times	2		

Table 3: Composition of per kg "Biotics"

Ingredients	Quantity
Sacchromyces cerevisiae (Min)	1.75×1011 CFU
Lactobacillus acidophilus	1.5×109 CFU
Bacillus subtilis	2.1×109 CFU
Aspergillus oryzae	1.5×109 CFU
amylase	3042 IU
Protease	11950 IU
Cellulase	500 IU
Lipase	870 IU
Beta-glucanase	1957 IU
Xylanare	739 IU
SiO ₂	25%
Al ₂ O ₃	4%
CaO	1%
Na ₂ O	0.025%
MgO	0.5%
Fe ₂ O ₃	0.5%

Each cage was covered at the top by another piece of tire cord net (7-7.5cm mesh size) to prevent escaping of fish and predation by bird. The whole structure was fixed with bamboo raft and tied with anchors at both side by nylon rope to facilitate easy floating of cages depending on water level.

cages were divided into three Nine net treatments each having three replications. Mono-sex (All male) tilapia (Oreochromis niloticus) having an average weight of 33.66±6.23g were stocked at a density of 50/m3 in each cage and reared for 120 days.

Feeding and Probiotic Supplement Preparation: Prior stocking in experimental cages tilapia fries were nursed for 45 days in a nursery pond at BFRI, RS campus. The Mono-sex (All male) tilapia (Oreochromis niloticus) fries were purchased from hatchery of CP Bangladesh Co. Ltd. a renowned multinational company. Commercial floating feed named "Ruposhi Bangla" was used for the experiment. The proximate compositions of different types of "Ruposhi Bangla" feed are given in Table 1 and feeding strategy is showed in Table 3.

probiotic "Biotics" А commercial named manufactured by Anova Pharma Joint Stock Company, long An Province, Vietnam was used for the experiment and its composition is given in Table 3. Prior applying feed to the cages, the probiotic was mixed well with the feed at a rate of 2 g/kg feed for treatment 2 and 3 g/kg feed for treatment 3. In case of treatment 1 no probiotic was applied. For mixing the probiotic with the feed, firstly the powder formed probiotic was diluted in water. Then the probiotic containing water was splashed over the feed stack as it could be mixed uniformly with the whole feed stack (Table 4). Finally the probiotic mixed feed stack was kept in a dry and sun lighted place for half an hour to become dried.

Determination of Growth Indices and Water Quality: Sampling was done at every 15 days interval by using a scoop net to observe the growth of tilapia. During each sampling fifty individuals were collected from each cage and weight of each tilapia was measured in nearest gm by using a digital precision balance to assess differential growth of mono-sex tilapia. The sampled fishes were handled very carefully to avoid handling stress.

Indicators of growth include Weight gain, Average daily gain (ADG), SGR (% per day), FCR and survival rate were expressed as following:

Weight gain=Mean final weight- Mean initial weight

RESULTS

Mean final weight-Mean initial weight

 $SGR (\%/day) = \frac{100 \times (lnw_2 - lnw_1)}{Duration of culture in days}$

Here, W2 = Mean final weight and W1= Mean initial weight

Food conversion Ratio (FCR) = ------Total weight gain

Water quality data *viz*. Temperature, pH, Dissolved Oxygen (DO), CO₂, Alkalinity, Hardness and Ammonia of the culture area in the Dakatia River were taken at monthly interval during the study period. Water quality sampling was done according to APHA [14] methods.

One way analysis of variance (ANOVA) was performed to test the significance of difference among the treatment means by using SPSS software package. Water quality parameters of the area adjacent to the experimental cages did not show any deviation from acceptable limit throughout the culture period shown in Table 4.

The growth rate of mono-sex (All male) tilapia fed with different levels of probiotics treated feed were recorded 15 days interval and the results have been presented in the Table 5. For the evaluation of growth indices of fish in different treatments such as weight gain, average daily gain, specific growth rate (SGR % per day), food conversion ratio (FCR), survival (%) and production (kg/cage) were calculated and are shown in Table 6.

In this study, there were no significant differences (P<0.05) in initial mean weight 33.66 ± 6.23 gm among treatments (Table 1). Final weight in probiotic treated cages (Cages belong to T-2 and T-3) was significantly (P<0.05) higher than control (T-1). Average weight was recorded higher in fish fed with probiotic supplement. Weight gain was found to be highest (Table 5) in T-3 which was significantly different from both T-1 and T-3. In case of ADG the result was as same as weight gain (Table 5). SGR was significantly higher in T-2 than that of T-1 and T-3. FCR was the best in T-2 and significantly different from T-1 but not from T-3 (Table 5). Survival rate and production per cage was also highest in T-2 and in case of production per cage it was significantly different from both T-1 and T-3.

Table 4: Water quality parameters of the area adjacent to the experimental cages							
Dissolved O2(mg/l)	Free CO ₂ (mg/l)	pH	NH ₃ (mg/l)	Total alkalinity	Total hardness	BOD (B) (mg/l)	BOD (N) (mg/l)
7.29±1.04(6.35-8.02)	3.18±0.79(2.62-5.97)	8.25±0.35(7.5-8.5)	0.10±0.14(0-0.14)	132.98±16.23(80.5-189.0)	164.16±21.51(66.57-203.0)	5.85±2.50(2.52-7.62)	7.69±1.62

	Treatments	Treatments				
Growth parameters	 T-1	T-2	Т-3			
Mean initial weight(gm)	33.66±6.23	33.66±6.23	33.66±6.23			
Mean final weight(gm)	207.90±7.01ª	271.48±13.69 ^b	240.07±6.35°			
Weight gain(gm)	174.24±7.01 ^a	237.82±13.69 ^b	206.41±6.35 °			
Average daily gain (ADG) (gm)	1.45±0.058 °	1.98±0.11 ^b	1.72±0.05 °			
Specific Growth Rate (SGR) (% per day)	1.52±0.028 ª	1.74±0.042 ^b	1.64±0.021 ^a			
Food conversion Ratio (FCR)	1.41±0.057 ^a	1.11±0.047 ^b	1.29±0.12 ^{ab}			
Survival rate (%)	95.76±0.86 ª	97.54±0.65 ^b	96.94±0.40 ^{ab}			
Production (kg/cage)	357.62±14.01 °	476.79±27.41 ^b	418.94±12.65 °			

Letters with different superscripts at the same row differ significantly by P<0.05 by means separation using the Turkey test.

DISCUSSION

Numerous trials were conducted with microorganisms as probiotics to improve cultivability of food species and to improve human health and welfare. Appropriate probiotics applications were shown to improve intestinal microbial balance, thus leading to improve food absorption [7, 15]. According to the results of this study, the tilapia fishes reared in the cages fed with commercial probiotic supplemented feed, showed improved growth rate. This may be attributed to activity of Probiotics that improve digestion by stimulating production of digestive enzymes or through other alterations in the gut environment [1]. The gut microbial population is also important to the nutrition of fish by increasing nutrient uptake and facilitating utilization of enzymes, amino acids, short-chain fatty acids, vitamins and improved digestion [1, 16, 17].

The mean final body weight, ADG and SGR were increased among *O. niloticus* with probiotics application treatments compared with probiotic free treatment. So we might assume to be considered as a growth promoter/enhancer in fish aquaculture. These results showed resemblances with Rad *et al.* [18] and Mohamed *et al.* [19] which were reported that the probiotics treated group of *O. niloticus* and *O. mossambicus* fingerling reared in concrete tanks exhibited enhanced growth rate. Survival of *O. niloticus* and *O. mossambicus* was significantly greater in probiotic treated groups than that of the control group

The commercial probiotic used in this experiment contains a significant portion of *Sacchromyces cerevisiae* that is likely to be a constituent, contributing to the exhibition of improved growth and FCR for the fishes fed with probiotic treated feed. Similarly [20] reported that weight gain and specific growth rate of tilapia increased significantly (P<0.05) with the increase in dietary yeast *(S. cerevisiae*) levels. Fish fed the diet containing *S. cerevisiae* exhibited the greatest increase in growth performance, which was attributed to significantly higher activity of alkaline phosphatase [21, 22]. Lara-Flores *et al.* [23] revealed that *S. cerevisiae* supplementation produced significantly higher weight gain and feed utilization efficiency in tilapia.

The results of the present study indicated clearly that the cage under T-2 (2.0 g probiotic/kg feed) showed enhanced growth performance as measured by final weight, SGR and feed utilization of tilapia. Nevertheless, It is difficult to draw concrete conclusions and provide specific recommendations on the effects of dietary probiotics on growth performance of cage reared tilapia, so that the studies vary widely with regard to fish age and size, stocking density, diet composition, dietary probiont concentration, feed allowances, feeding duration and of course, type and source of probiont [1]. The stimulatory effect of dietary probiotics immune parameters and resistance of tilapia to infectious pathogens need further investigations.

CONCLUSION

The application of probiotics in aquaculture ponds appears bright. There is an over-increasing demand for aquaculture production and a similar increase in the search for alternative to antibiotics, the application of probiotics intended for culture systems is now attracting considerable attention and a number of commercial products are available, particularly directed at the culture organisms. Krishna et al. [24] explained that the use of probiotics in biological systems in details and they give details report on sustainable aquaculture management. In the present study all of the growth parameters were significantly higher in cages with probiotic treated feed than control cages. The improved survival and growth parameters in treatments over controls strongly suggests that adding the probiotics during the cage culture and continuing its administration throughout the farming stages could be a noteworthy practice to maximize survival and growth in cage culture system. Though there is a paucity of study on effect of probiotics on cage aquaculture the general conclusion obtained from the present study is that the probiotics plays a vital role in survival and disease resistance of the cage reared fish species like tilapia.

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