Article

Efficiency of probiotics (Ecoforce) in the growth and survival of *Peneaus monodon*

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Abstract

Probiotic supplementation of live microorganisms in aquaculture aids in preventing disease, thereby increasing production and decreasing economic loss. Application of probiotics bacteria in aquaculture systems plays significant role that determines the fate and success rate of culture. The present study was carried out to evaluate the performance of commercially available probiotics (The major active ingredients include *Streptococcus faecalis, Streptococcus faecium, Bacillus mesentericus, Bacillus subtilis, Bacillus natto, Clostridium butyricum, Saccharomyces cerevivisiae*, Alkaline Protease and Lipase) in the growth and survival of *Peneaus monodon* in grow-out conditions. At the end of the culture, the survival rate of Pond I (treatment pond) was 76%, whereas for Pond II (control pond) it was only 59%. Pond I (29.2g) where as shrimps in the Pond II showed the least average body weight of 22.6g. It is evident that application of probiotics has improved the growth and survival of *P. monodon* and which in turn paved way to reap better profit for the farmers. This investigation showed that administration of mixed *Bacillus* probiotics significantly improved growth and survival of shrimp, increased beneficial bacteria in shrimp culture and enhanced water quality for parameter., it seems likely that the use of probiotics will gradually increase in the days to come that will open new vistas in the arena of aquaculture.

Keywords semi-intensive culture; probiotics; growth rate; survival; FCR.

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1 Introduction

The 21st century marine fisheries scenario is facing a pathetic situation in recent years due to declining yield of the target fish species in terms of catch per unit effort. A recent study revealed the shocking conclusion that the way the fishing is conducted now is not sustainable and predicted the end of fisheries industry by 2050 (Boris, 2006). During fiscal year 2009-2010, according to officials at India's Marine Exports Product Development

Authority (MPEDA), total aquaculture production reached 106,000 metric tons, an increase of more than 30% over the previous fiscal year. Tamil Nadu having about 56,620 ha of brackish water lands and the potential area for shrimp culture is estimated about 3, 684.18 ha.

Probiotics-supplemented functional food will contain single or mixed cultures of micro-organisms which capable of improving the health of the host (Fuller, 1992). The concept of probiotics first used in animal husbandry (Fuller, 1989) has been exploited for use in aquaculture systems (Moriarty, 1998). The research on probiotics for aquatic animals is increasing with the demand for environment friendly aquaculture. In recent years, there is a great interest in the use of probiotics bacteria in aquaculture to improve disease resistance, water quality and growth of farmed shrimps. Application of commercial microbial products (probiotics) in aquaculture ponds is rapidly increasing as a disease management strategy (Gatesoupe, 1999).

A feed probiotics is defined as "a living microbial supplements that positively affects hosts by modifying the host-associated microbial community, improves food degradation by enhancing its nutritional value, and improves the quality of the environmental parameters" (Verschuere et al., 2000). Rengpipat et al (1998a, b; 2000) emphasized that probiotics bacteria administered in the culture proved to improve the growth and survival of *Penaeus monodon* by the way of enhancing the immunity. Probiotics applied through feed beneficially act upon shrimp growth, ultimately increasing production. However, the beneficial effect of using such microbial products in aquaculture is still debatable as their efficacy is yet unclear. In India, several imported microbial products are being used in shrimp farms but, little attempt has been made to verify the performance of these products. In this backdrop, an attempt has been made in the present study to investigate the effects of the commercial available feed probiotics on the growth and survival of farmed shrimps.

2 Materials and Methods

The present study was conducted at Clinton aqua farms, Sivanarpuram during from November 2007 to February 2008. Total experiments of 2 ponds, each of 0.5ha water spread area, were selected. The feed probiotics selected for the present study are Ecoforce, produced by Tablets India Ltd. Pond I, the application of probiotics commenced from the 15th day of culture and Pond II was treated as control. The water quality parameters in the pond are strictly monitored on daily basis at different corners of the pond, the water salinity was measured by using a hand refractometer (Erma-Japan). The pH of the pond water was measured by using electronic pH pen (Erma-Japan). The dissolved oxygen was estimated by modified Winkler's method as described by Strickland and Parsons (1972). Transparency was quantified based on the measurement of light penetration, using a Secchi disc.

2.1 Ecoforce

The major active ingredients of Ecoforce for a 500 g pack are detailed as below:

Streptococcus faecalis	T-110	$10g (3.5X10^9)$
Streptococcus faecium	T-120	ſ
Bacillus mesentericus	TO-A)
Bacillus subtilis	TO-A	$10g (1.5X10^8)$
Bacillus natto	TO-A	J
Clostridium butyricum	TO-A	$10g (1.0X10^8)$
Saccharomyces cerevivisiae		5g
Alkaline Protease		5g (500,000 units)
Lipase		5g (20,000 units)

Apart from this, various micro and macronutrients and common fish attractants are also included.

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2.2 Feeding frequency

The application dosage was calculated as 10g of probiotics per kg of feed. Initially the probiotics are diluted with 500-1000 ml freshwater and are subjected to mild aeration for 30 minutes. Then the liquid is sprinkled over the feed, mixed well and subjected for 30 minutes in shade drying. After 30 minutes, the egg or gels were used as binder to keep the feed intact and allow the feed for shade drying at room temperature. Then the feeds are uniformly broadcasted in the growout.

The shrimps were fed with CP feed (Charoen Pokhpond Aquaculture India Pvt. Ltd.). The feeding schedule was based on the feed chart given by the CP Company. The total feed used per day was rationed at the rate of 18%, 23%, 18%, 23% and 18% in the morning (6:00 AM), noon (10:00 AM), afternoon (2:00 PM), evening (6:00 PM) and night (10:00 PM) time respectively. The feed were broadcasted following the rope method with the aid of floats so as to ensure that the feed is spread over a wider area. The type of feed changed (proximate composition and size) as the days of culture progressed and is detailed in Table 1.

S.No	DOC	TYPE OF FEED
1	1-7	CP Irawan no 1
2	8-21	CP Irawan no 2
3	22-28	CP Irawan nos 2+3
4	29-60	CP Irawan no 3
5	61-64	CP Irawan nos 3+4
6	64- end	CP Irawan no 4s

 Table 1 Type of feed used during different days of culture.

Healthiness, survival rate, Average Body Weight (ABW) of the animals was estimated with the help of cast net sampling. Sampling was done in all ponds every fortnight during the early hours of the day with a cast net of known diameter (3.2m). The total number of shrimps caught per haul and individual weight of the shrimps were recorded, The Average Body Weight were calculated based on these data

2.3 Survival

Survival (%) = Nos. of animals survived/ Nos. of animals stocked $\times 100$

2.4 Average Body Weight (ABW)

Weight Gain (%) = (Final weight – Initial weight)/Initial weight $\times 100$

2.5 Food Conversion Ratio (FCR)

FCR = Dry weight of ingested food/Wet weight of produced shrimp

3 Results

The results of survival rate of shrimps fed with probiotics in Pond I are detailed in Fig. 1. The growout subjected to probiotics application showed higher survival rate than that of control pond, as a final point pond I, 76% whereas the survival rate of Pond II was only 59%. The results of growth performance based on Average Body Weight are detailed in Fig. 2. The average body weight of shrimps at 15 DOC was 2.8 g in Ponds I & II. The growout Pond I subjected to probiotics application showed the ABW 29.2g from pond I and least average body weight of 22.6g from control pond.



ABW - Experimental and control pond

Fig. 1 ABW experimental and control pond.



Survival - Experimental and control pond

Fig. 2 Percentage of survival experimental and control pond.

The Food Conversion Ratio (FCR) of shrimps grown was calculated separately for each culture pond the details are depicted following. Ratio was 1:1.47 and 1:1.80 experimental and control pond. Among the grow-out ponds, Pond I showed a comparatively good FCR than that of control pond (Fig. 3).



FCR - Experimental and control pond

Fig. 3 Food conversion ratio between experimental and control pond.

4 Discussion

The use of probiotics in the culture of aquatic organisms is increasing with the demand for more environmentfriendly aquaculture practices (Gatesoupe, 1999). Probiotics are micro-organisms or their products that ensures health benefit to the host which are used in aquaculture as a means of diseases control, improve immunity of cultured animals to pathogenic microorganisms and positively affects the growth and survival. In the present study an attempt has been made to compare the performance of commercially available feed probiotics in the growth and survival of *P. monodon*. Probiotics entitled "Ecoforce" was applied in pond I, and pond II was treated as control.

The results of survival revealed that shrimps in ponds treated with probiotics exhibited fairly higher survival than that of control pond. Pond I which used an Ecoforce showed higher survival percentage than that of control pond. A similar trend was also noticed in the case of growth in such a way that *P. monodon* grown in pond I, that supplied with the probiotics performed high Average Body Weight than that of control pond. Similar results were obtained by Ziaei-Nejad et al. (2006) in which the *Bacillus* probionts showed positive effect on digestive enzyme activity, survival and growth in the shrimp *Fenneropenaeus indicus*. Moriarty (1998) and Rengpipat et al. (2000) examined an increase in survival rate of shrimps fed with probiotics. Similarly, Rengpipat et al. (2000) examined an increased survival and growth of *P. monodon* fed with probiotits *Bacillus* S11 for 90 DOC.

In Indian shrimp industry the average food conversion ratios were varying between 1.5 and 1.85 (Paul Raj, 1999). Saha et al. (1999) observed that the food conversion ratios of 1.31 to 1.58 in low saline ponds where as Ramakrishna (2000) recorded the FCR of 1.35 to 1.68 in high saline ponds. In the present study the food conversion ration varied between 1:1.47 and 1:1.80 among which the good FCR were recorded in ponds applied with probiotics.

In the case of probiotics Ecoforce the active strains included are *Streptococcus faecalis, Streptococcus faeciu, Bacillus mesentericus, Bacillus subtilis, Bacillus natto, Clostridium butyricum* and *Saccharomyces cerevivisia* were the active beneficial microbial strains. Studies have shown that when these bacterial strains were administered as probiotics in the *P. monodon* culture, its growth and survival were improved and

immunity was enhanced (Rengpipat et al., 1998a, b, 2000). Administration of *Bacillus* spp. as probionts has been shown to increase shrimp survival by enhancing resistance to pathogens by activating both cellular and humoral immune defenses in shrimp (Rengpipat et al., 2000). The higher survival in probiotics fed shrimps might be attributed to the presence of *Bacillus* bacteria which are able to out-compete other bacteria for nutrients and space and can exclude other bacteria through the production of antibiotics (Moriarty, 1998; Verschuere et al., 2000).

From the present study it is evident that application of probiotics has improved the growth and survival of *P. monodon* and which in turn paved way to reap better profit for the farmers. A detailed investigation is found wanting on the selection of probiotics strains and its positive roles in the immunity and growth enhancement of shrimps. It is safe to conclude that probiotics are welcome addition to the armament of disease prophylaxis in aqua farms although the technology and science behind it is still very much in a development phase, it seems likely that the use of probiotics will gradually increase in the days to come that will open new vistas in the arena of aquaculture.

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References

- Boris. 2006. Estimates that half of the world's seafood demand will be met by aquaculture in 2020. Food and Agricultural Organization (FAO), Italy
- Fuller R. 1989. Probiotics in man and animal. Journal of Applied Bacteriology, 66: 365-378
- Fuller R. 1992. History and development of probiotics. In: Probiotics: The Scientific Basis (Fuller R, ed). Chapman and Hall, New York, USA
- Gatesoupe FJ. 1999. The use of probiotics in aquaculture. Aquaculture, 180: 147-165
- Moriarty DJW. 1998. Control of luminous *Vibrio* species in penaeid Aquaculture ponds. Aquaculture, 164: 351-358
- Paul Raj BB. 1999. Ecofriendly feed and management system for sustainable shrimp culture. Fisheries World, 13-17
- Ramakrishna R. 2000. Culture of the tiger shrimp *Penaeus monodon* (Fabricus) in low saline waters. MSc Dissertation. Annamalai University, India
- Rengpipat S, Phianphak W, Piyatiratitivorakul S, et al. 1998. Effects of a probiotic bacterium on black tiger shrimp *Penaeus monodon* survival and growth. Aquaculture, 167: 301-313
- Rengpipat S, Rukpratanporn S, Piyatiratitivorakul S, et al. 2000. Use *Carnobacterium sp.* as probiotic for Atlantic salmon (*Salmo salar* L.) and rainbow trout (*Oncorhynchus mykiss* Walbaum). Aquaculture, 185: 235-243
- Saha SB, Bhattacharyya C, Choudhury A. 1999. Preliminary observations onculture of *Penaeus monodon* in low saline waters. Naga, 22(10): 30-33
- Verschuere L, Rombaut G, Sorgeloos P, et al. 2000. Probiotic bacteria as biological agents in aquaculture. Microbiology and Molecular Biology Reviews, 64(4): 655-671
- Ziaei-Nejad S, Rezaei MH, Takami GA, et al. 2006. The effect of *Bacillus* spp. bacteria used as probiotics on digestive enzyme activity, survival and growth in the Indian white shrimp *Fenneropenaeus indicus*. Aquaculture, 252: 516-524