

Research Article

The Role of Probiotics in *Penaeus vannamei* Aquaculture from the Culture Ponds of Gollalavalasa, Andhra Pradesh, India

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Abstract

The present work is carried out in commercial shrimp farms located at Gollalavalasa of Srikakulam District, Andhra Pradesh, India, over a period of two consecutive years i.e. 2018-2019. Modified extensive shrimp farms were selected for this research work. The data was recorded from both control and experimental ponds in summer and winter crops. In the present study the growth and survival of the shrimps in all the experimental ponds showed better performance than control ponds in two different crops during the study period. The important point of consideration in the study is that the application of feed probiotics along with the immunostimulant showed significantly better results than the feed probiotics alone given in the feeds to the shrimp. The slow growth rate in the control ponds in the selected study areas was due to high pathogenic bacterial loads, delayed moulting and stunted growth.

Keywords: Growth, Survival, *P. vannamei*.

Introduction

Probiotic is a live non-pathogenic microorganism and resist to the actions of pathogenic microorganisms. So that it can be used to treat some diseases. According to Fuller (1992) probiotics as feed supplements can be improve intestinal micro flora balance of the host. Farzanfar (2006) provided information about the lactic acid bacteria and *Bacillus* species as ecofriendly agents; these can be compete with harmful bacteria for resources and promotes the growth in cultivable organisms such as shellfish and finfish. According to Verschuere *et al.*, (2000) as live microorganism, probiotics could show positive effects on the host by changing the host associated microorganisms community, there by improved feed consumption and enhance the nutritive value of the feed and response of the host towards disease.

The usage of probiotic in shrimp farming has been increased with the demand for eco-friendly aquaculture as reported by Wang *et al.*, (2005), Balcazar *et al.*, (2007) and Kesarcodi-Watson *et al.*, (2008). Ferreira *et al.*, (2015) reported about the microbial biofloc as a source of probiotic bacteria in *P. vannamei* farming. The role of probiotics in disease control and usage of probiotics was described by Newaj-Fyzul *et al.*, (2014). Kumar *et al.*, (2016) provided information about the importance of probiotics in aquaculture, methods of application and mechanism of their action in the culture system. Brown (2011) described about recent advances and mode of action in probiotics. Abdullah *et al.*, (2011) also evaluated the use of *Lactobacillus acidophilus* as probiotic for controlling pathogenic bacteria and haematological parameters in *Clarias gariepinus*. Hostins *et al.*, (2017)

evaluated the efficacy and differences in bacterial population densities in the gut of *P. vannamei*. Franco *et al.*, (2017) also evaluated two important probiotics used in *P. vannamei* culture system. Many studies have been conducted on effect of probiotics in shrimp farming. The feed conversion ratio and growth rates were significantly higher in probiotic fed shrimps than those shrimps were not fed with probiotics as reported by Ziaei-Nejad *et al.*, (2006). The purpose of the present study is to evaluate the role of probiotic Pro-2 along with immunostimulant 1,3 β -Glucan, a commercial brand β -ADVANTAGE to assess the impact on growth and survival of shrimp *P. vannamei*.

Material and Methods

The present work is carried out in commercial shrimp farms located at Gollalavalasa of Srikakulam District, Andhra Pradesh, India, over a period of two consecutive years i.e 2018-2019. Modified extensive shrimp farms were selected for this research work. The data was recorded from both control and experimental ponds in summer and winter crops. For studies on growth and survival feeding was followed according to the specifications given by the feed manufacturers in both crops during the study period. The feed used during the study was C.P. branded semi-intensive feed. The stocking density of all the ponds was uniformly followed during the study period in both control and experimental ponds. The stocking density was done uniformly at the rate of 1, 13,000 seeds per hectare pond i.e. 13 pieces/sq.mt. Four check trays were arranged in four corners of the pond. Feeding procedure was monitored according to body weight sampling after check tray observation. The feeding procedure was followed as follows: 25% at 6 am, 20% at 11 am, 30% at 6 pm and 25% at 10 pm. After 15 days of stocking, sampling of shrimp was done weekly during early hours of the day with a cast net and weights are recorded and tabulated. The Survival rate and average body weight (ABW) of the shrimp were estimated and condition of shrimp health was observed.

The body weight and survival rate of the shrimp was estimated by adopting the formula

Average Body Weight of the shrimp (ABW)

Weight gain = Final weight of the shrimp-Initial weight of the shrimp/ Initial weight of the shrimp X 100

% Survival rate of the shrimp

Survival rate = Number of shrimps survived/ Number of shrimps stocked x 100

Application of feed probiotic

In the present study feed probiotic Pro-2 was applied along with the immunostimulant 1, 3 β -Glucan, a commercial brand β -ADVANTAGE for both summer and winter crops. The feed probiotic applied at the rate of 5g/kg and 10g/kg with 5g/kg immunostimulant in the experimental ponds at two different study areas. The application of feed probiotic and immunostimulant was followed every day for both the seasons i.e. summer and winter during study period i.e. 2018 to 2019.

Statistical analysis

One-way ANOVA was carried out to check the effect of days on the growth and survival rate in control and experimental farms of the winter crop and summer crops at Gollalavalasa during the years 2018 to 2019. These analyses were done by using IBM SPSS Version 22.0. Bar graphs were drawn by using mean values and SD of growth and survival rates in MS Excel 2016. All values were represented as Mean \pm SD.

Results

It is evident from the results in summer crop of year 2018 at Gollalavalasa, that the growth in grams of *P. vannamei* was noticed as 3.62 ± 0.19 at 30 days of culture in control pond where as in the experimental pond, the growth of 4.28 ± 0.22 was observed at 30 days of culture. Similarly the highest growth in grams was noticed as 21.86 ± 0.28 , 23.89 ± 0.62 in control and experimental pond at 120 days of culture respectively (Table 1 & 2, Figure 1 & 2).

Similarly in the winter crop of year 2018 the growth in grams of *P. vannamei* was noticed as 4.22 ± 0.22 at 30 days of culture in control pond, where as in the experimental pond, the growth of 4.29 ± 0.26 was observed at 30 days of culture. Similarly the highest growth in grams was noticed as 24.35 ± 0.61 , 24.92 ± 0.54 in control and experimental pond at 120 days of culture respectively (Table 1 & 2, Figure 1 & 2).

It is evident from the present results in summer crop of year 2019 at Gollalavalasa, that the growth in grams of *P. vannamei* was noticed as 4.10 ± 0.18 at 30 days of culture in control pond and this pond harvested due to white spot disease at 20.52 g on 112nd day, whereas in the experimental pond, the growth of 5.17 ± 0.29 was noticed at 30 days of culture. Similarly the highest growth in grams was noticed as 24.86 ± 0.63 in experimental pond at 120 days of culture (Table 1 & 2, Figure 1 & 2).

Similarly in the winter crop of year 2019 the growth in grams of *P. vannamei* was noticed as 3.99 ± 0.26 at 30 days of culture in control pond, where as in the experimental pond, the growth of 4.28 ± 0.24 was observed at 30 days of culture. Similarly the highest growth in grams was noticed as 22.20 ± 0.44 , 23.64 ± 0.61 in control and experimental pond at 120 days of culture respectively (Table 1 & 2, Figure 1 & 2).

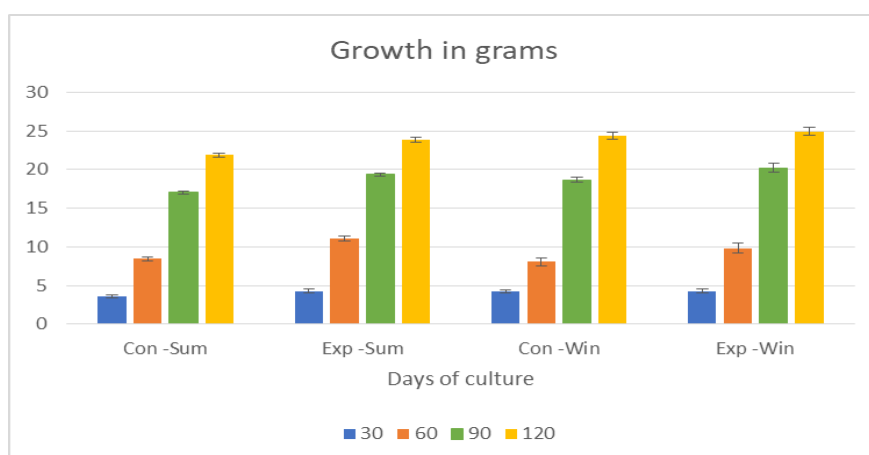


Figure 1. Growth of *P. vannamei* (in grams) at Gollalavalasa during the year 2018.

Table 1. ANOVA for Growth of *P. vannamei* (in grams) at Gollalavalasa during the year 2018.

Growth in grams					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	32.532	3	10.844	.166	.919
Within Groups	2878.856	44	65.429		
Total	2911.388	47			

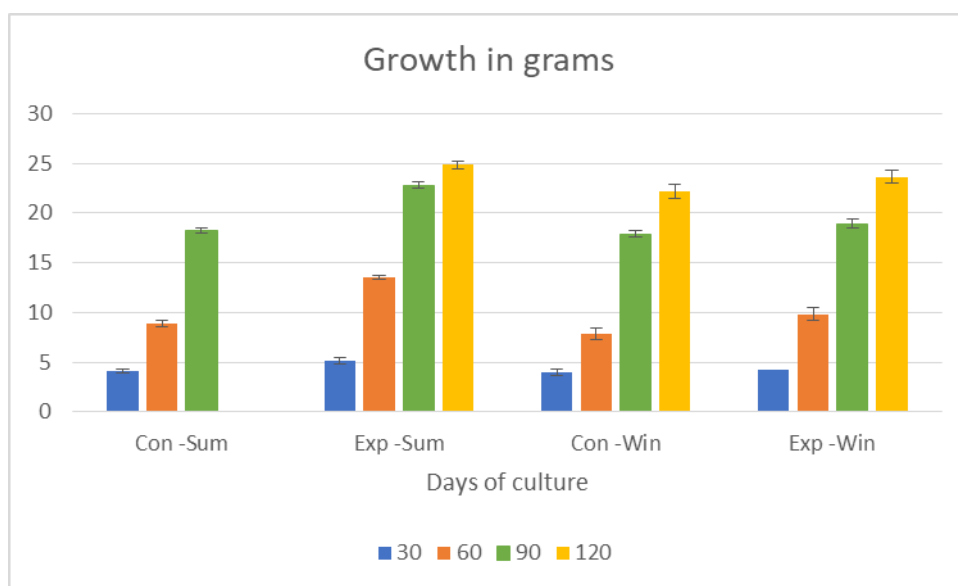


Figure 2. Growth of *P. vannamei* (in grams) at Gollalavalasa during the year 2019.

Table 2. ANOVA for Growth of *P. vannamei* (in grams) at Gollalavalasa during the year 2019.

Growth in grams					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	205.856	3	68.619	1.176	.331
Within Groups	2392.669	41	58.358		
Total	2598.525	44			

Table 3. Percentage survival rate of *P. vannamei* in the culture ponds at Gollalavalasa during the years 2018 to 2019.

S/N	Station	2018				2019			
		Summer Crop		Winter Crop		Summer Crop		Winter Crop	
		C.P.	E.P.	C.P.	E.P.	C.P.	E.P.	C.P.	E.P.
1	Gollalavalasa	72.89	80.49	70.87	79.86	80.42	87.53	81.53	86.54

Note: C.P. = Control Pond; E.P. = Experimental Pond

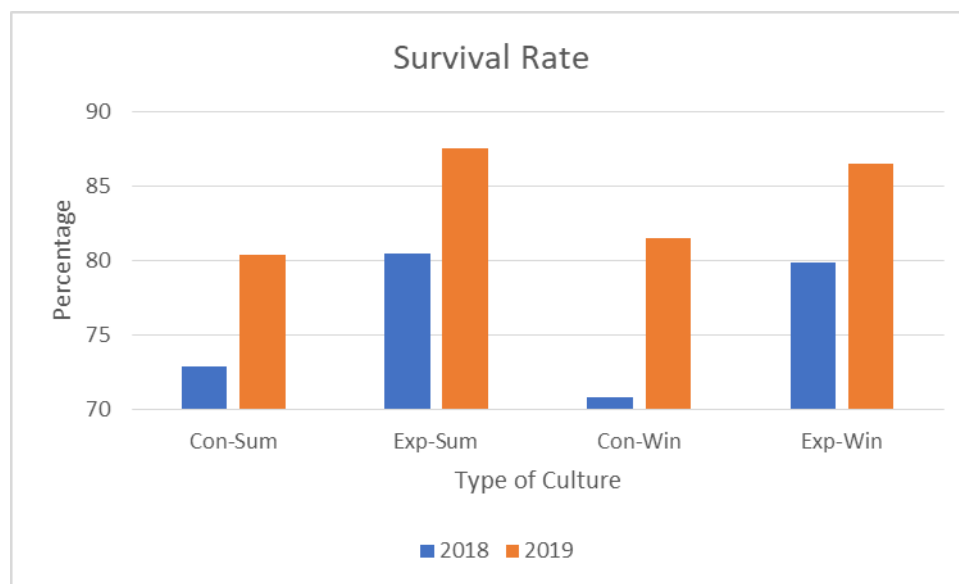


Figure 3. Percentage survival rate of *P. vannamei* in the culture ponds at Gollalavalasa during the years 2018 to 2019.

Survival rate

The percentage survival rates of *P. vannamei* in control and experimental ponds were tabulated and the data was evaluated. It is evident from the present results that, the survival rate of *P. vannamei* during the year 2018 at Gollalavalasa in summer crop the percentage of 72.89% and 80.49% was observed for both control and experimental ponds respectively. Similarly in the winter crop of 2018 these values were observed as 70.87% and 79.86% for both control and experimental ponds respectively (**Table 3, Figure 3**).

In the same way the survival rate of *P. vannamei* during the year 2019 at Gollalavalasa in summer crop the percentage of 80.42% and 87.53% was observed for both control and experimental ponds respectively. Similarly in the winter crop of 2019 these values were observed as 81.53% and 86.54% for both control and experimental ponds respectively (**Table 3, Figure 3**).

Discussion

Probiotics can help the host for proper utilization of nutrients for its metabolic requirements. For instance 20% increased digestibility was noticed in *P. indicus* when the shrimp fed with lactic acid bacteria as probiotic as reported by Fernandez *et al.*, (2011). In a study revealed that when the shrimps were fed with probiotics changes occurs in the growth performance by means of feed efficiency, and feed conversion efficiency as reported by Boonthai *et al.*, (2011) and Zokaeifar *et al.*, (2012, 2014). In addition to capability of probiotics in enhancing the growth performance, they influence on the enzymes present in the gut of the host is believed to be one of significant contribution. According to Becerra-Dorame *et al.*, (2012) the nutritional condition of the cultivable animals effected for both given feed and their digestive physiology. Digestive enzymes are very much essential to breakdown the complex compounds into simple compounds and absorbable molecules that can be utilized by the host animal as reported by Lazado *et al.*, (2012). Actually utilization of feed related material is

greatly depends on whether they can be easily absorbed or not by the host digestive physiology.

In some times the host digestive physiology is influenced by several factors, such as types of feed, environment and health of the host. In view of probiotic applications, enzymatic influence could be induced either by stimulation or direct contribution. According to Kureshy and Davis (2002) probiotic is main limiting nutrient for shrimp growth, so that protease producing probiotic is considered as an important component. In a study of Ochoa-Solano and Olmos-Soto, (2006) the increased digestibility is related to probiotic application has been connected to efficiency of the microorganisms in facilitating the protein digestibility by means of their enzymes and those influence on the host endogenous proteolytic activity.

Zhang *et al.*, (2011) observed the decreased population of vibrio bacteria in the gut of *P. japonicus* when fed with probiotics. These observations represent the competitive elimination mechanism of probiotics in modifying the microbial population of the gut, specially reduce the pathogenic bacterial counts. According to Xiongfei *et al.*, (2005) and Taoka *et al.*, (2006) application of probiotics on the shrimp *P. vannamei* and fish *Paralichthys olivaceus* cultured in recirculating system observed better growth performance and high survival rates in experimental groups than control group. To improve water quality in shrimp hatcheries recently usages of probiotics were increased (Gomez and Shen, 2008; van Hai *et al.*, 2009). Routine application of probiotics in farming and hatchery systems, higher feed conversion ratios and improvement of growth of the shrimp was observed by Liu *et al.*, (2009).

Molting frequency has been improved in shrimp larvae with the application of probiotics *Bacillus fusiformis* as reported by Guo *et al.*, (2009) and Zhou *et al.*, (2009). According to Decamp *et al.*, (2008) synthesis of viable probiotic is not an easy task and full scale trials may require as well as development of keen monitoring tools and controlled production. The main aim of using probiotics is to maintain or restore a favorable conditions between friendly and pathogenic microorganism that represent the bacterial flora of skin mucus of aquatic animals. Prosperous of probiotics is assumed to have specific properties in order to validate a beneficial effect as reported by Ali (2000).

According to Vaseeharan and Ramasamy (2003) and Guo *et al.*, (2009) antagonistic activity of *B. subtilis* against *V. harveyi*, bestowed on protection to *P. monodon*. And they also reported about the antagonism of *B. foraminis*, *B. cereus*, and *B. fusiformis* against *Streptococcus iniae* and *Photobacterium damsela* sub sp. piscicida and increases larval survival of *P. vannamei* in vitro and vivo conditions. Ajitha *et al.*, (2004) reported about the antigenic components in *Pseudomonas* species that show antagonism against *Aeromonas hydrophobia* and extracts of *L. acidophilus*, *S. cremoris*, *L. bulgaricus* also revealed negative effect on growth of the *V. alginolyticus* in agar plate incubations.

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