CIBTech Journal of Zoology ISSN: 2319–3883
Online, International Journal, Available at http://www.cibtech.org/cjz.htm
2022 Vol.11, pp.205-208/Jayaprakash
***Research Article** (Open Access)*

EFFECT OF PROBIOTIC (SUPER BIOTIC) ON THE TOTAL VIBRIO LOADS IN THE CULTURE PONDS OF *PENAEUS VANNAMEI*

***Jayaprakash Ronanki**
Department of Zoology, Govt. Degree College (Men),Srikakulam, A.P.-532001
**Author for Correspondence*

ABSTRACT
The main objective of the present study is to evaluate the effect of probiotic (SUPER BIOTIC) on the total vibrio loads in the culture ponds of *P. vannamei*. The present study was conducted at Amplam in Srikakulam district of Andhra Pradesh, India. This study was conducted in two different crops i.e. summer and winter crops both in control and experimental ponds. In the experimental pond of summer crop the salinity ranged from 18 to 21 ppt, pH ranged from 7.8 to 8.6, temperature ranged from 29 to 31^0C and TVC values fluctuated between $0.25\times10^2\pm0.26$ to $3.85\times10^2\pm0.40$ and at 120 days of culture in the experimental ponds the TVC levels were reduced when compared to control ponds and recorded as $2.37\times10^2\pm0.40$. In the experimental pond of winter crop the salinity ranged from 15 to 19 ppt, pH ranged from 7.8 to 8.5, temperature ranged from 25 to 28^0C and TVC values fluctuated between $0.11\times10^2\pm0.10$ to $2.90\times10^2\pm0.38$ and at 120 days of culture in the experimental ponds the TVC levels were reduced when compared to control ponds and recorded as $2.01\times10^2\pm0.30$.

Keywords: *Control pond, experimental pond, P. vannamei,Total Vibrio Count (TVC)*

INTRODUCTION
In the shrimp culture the application of probiotics started in 1986 and that of organic acids in 1990's and gained importance continuously till now. The concept of usage of organic acids and probiotics in shrimp culture took its pace in the last ten years and the results are highly promising in the successful productions. The main factors addressed by these two are prevention of pathogenic bacterial colonies in the ecosystem thereby preventing the incidence of diseases. At the same time avoid the usage of Antibiotics in the aquaculture; these were administered in the pond ecosystem and also in the feeds. The available products of probiotics mostly using in aquaculture practices are related to the genus *Bacillus* and *Bifidobacterium* species. The *Bacillus subtilis*, *Bacillus licheniformis* and *Bacillus circulans* are comes under *Bacillus* species, *B. bifidum, B, lactis, B. thermophilum* are comes under *Bifidobacterium* species. Lactic acid bacteria i.e. *Lactobacillus* spp, *Carnobacterium* spp. and yeast *Sachharomyces cerevisiae* are also well studied (Lee *et al.*, 1999; Sanders and Klaenhammer, 2001). In Indian aquaculture practices numerous studies were conducted to evaluate the efficiency of antagonistic properties of commercially available probiotics they are Top-25, Epicin, MIC-sanolife, Hi-5, Korag bios, Inve-Pro-W, Biomin, and Bactocell-100. The main bacterial species present in these products are *Lactobacillus* spp, *Bacillus* spp. *Pseudonomonas* spp, *Pediococcus* spp as microbial ingredients. The available reports on the effect of Super Biotic are very limited from the coastal ponds of Srikakulam. Hence in the present investigation various experiments were conducted to evaluate the effect of probiotic (SUPER BIOTIC) on the total vibrio loads in the culture ponds of *P. vannamei*.

MATERIALS AND METHODS
Modified semi intensive farms were used for the present study at Amplam in Srikakulam district located at Latitude 18^o18'N and Longitude 83^o 54'E. For studies on the role of probiotics, the feeding of shrimp was followed according to the specifications given by the feed manufacturers for both crops during the study period. The feed used during the study was C.P. semi-intensive feed. The stocking density of the ponds (both control and experimental) was uniformly followed for both summer and winter seasons. The

stocking density was done uniformly at the rate of 3, 50,000 seeds per hectare pond i.e. 35 pieces/sq.mt. The application of probiotics in the experimental ponds was followed uniformly in two different crops. The probiotic used for this study is SUPER BIOTIC which is a composition of water probiotic having the strength of 10 million colony forming units (CFU) i.e. 109 cfu/g. The probiotic strains in this SUPER BIOTIC were *Bacillus spp.* they are *Bacillus subtilis*, *Bacillus licheniformis*, *Bacillus megatirium* and *Bacillus polymixa*. The probiotic was applied at the rate of 2.5kgha^{-1} in the experimental pond at the frequency of every 15 days during the study period.

Estimation of bacterial load of water

Water samples from the selected culture ponds were collected in sterile glass bottles and brought to the laboratory in cold condition and bacterial loads were estimated within one hour of collection, by employing standard pour plate method. The samples were prepared by serial dilution method. One ml of diluted water sample was taken asceptically into sterile, dry petridish with the help of a pipette. The nutrient agar medium (Himedia, Bombay) in lukeworm state was poured onto the sample contained in the petridish, and then petridish was rotated gently in both clock and anti-clockwise directions for uniform distribution of sample solution, triplicate sets were maintained for each experiment. The petridishes were inverted after medium got solidified. The total *Vibrio* count analysis in the laboratories were done in the ZMA (Zobell Marine Agar) and TCBS (Thiosulphate Citrate Bilesucrose Agar) Medium under strict and hygienic microbiological laboratory conditions. The recorded values were tabulated.

RESULTS AND DISCUSSION

Table 1. Total *Vibrio* Count of culture ponds at Amplam during summer 2020

Control pond

S/N	Days of culture	Salinity ppt	pH	Temp ^{0}C	TVC Water cfu/ml
1	Before stocking	19	7.9	29	NIL
2	30	20	8.4	31	$0.33\times10^2\pm0.22$
3	60	20	8.3	30	$1.61\times10^2\pm0.31$
4	90	18	8.5	32	$4.57\times10^2\pm0.29$
5	120	-	-	-	-

Control pond harvested due to white spot disease at 21.07 g on 105th day

Experimental Pond

S/N	Days of culture	Salinity ppt	pH	Temp ^{0}C	TVC Water cfu/ml
1	Before stocking	19	7.9	28	NIL
2	30	20	8.5	31	$0.25\times10^2\pm0.26$
3	60	21	8.1	29	$0.87\times10^2\pm0.32$
4	90	19	8.6	31	$3.85\times10^2\pm0.40$
5	120	18	7.8	30	$2.37\times10^2\pm0.40$

The major water quality parameters such as salinity, pH and temperature were recorded in both control and experimental ponds during the year 2020. In the control pond of summer crop the salinity ranged from 18 to 20 ppt, pH ranged from 7.9 to 8.5, temperature ranged from 29 to 32^{0}C and TVC values fluctuated between $0.33\times10^2\pm0.22$ to $4.57\times10^2\pm0.29$.

In the experimental pond of summer crop the salinity ranged from 18 to 21 ppt, pH ranged from 7.8 to 8.6, temperature ranged from 29 to 31^{0}C and TVC values fluctuated between $0.25\times10^2\pm0.26$ to $3.85\times10^2\pm0.40$ and at 120 days culture in the experimental ponds the TVC levels were reduced when compared to control ponds and recorded as $2.37\times10^2\pm0.40$.

Table 2. Total *Vibrio* Count of culture ponds at Amplam during winter 2020
Control pond

S/N	Days of culture	Salinity ppt	pH	Temp ^{0}C	TVC Water cfu/ml
1	Before stocking	16	7.0	26	NIL
2	30	17	7.2	27	0.18 ×10²±0.31
3	60	18	8.1	25	2.24 ×10²±0.22
4	90	16	8.0	26	3.09 ×10²±0.49
5	120	-	-	-	-

Control pond harvested due to white spot disease at 15.5 g on 90th day
Experimental Pond

S/N	Days of culture	Salinity ppt	pH	Temp ^{0}C	TVC Water cfu/ml
1	Before stocking	15	7.8	27	NIL
2	30	17	8.0	28	0.11 ×10²±0.10
3	60	16	8.2	26	0.81 ×10²±0.52
4	90	19	8.3	25	2.90 ×10²±0.38
5	120	18	8.5	26	2.01×10²±0.30

The major water quality parameters such as salinity, pH and temperature were recorded in both control and experimental ponds during the year 2020. In the control pond of winter crop the salinity ranged from 16 to 18 ppt, pH ranged from 7.0 to 8.1, temperature ranged from 25 to 27^{0}C and TVC values fluctuated between 0.18 ×10²±0.31 to 3.09×10²±0.49.

In the experimental pond of winter crop the salinity ranged from 15 to 19 ppt, pH ranged from 7.8 to 8.5, temperature ranged from 25 to 28^{0}C and TVC values fluctuated between 0.11×10² ±0.10 to 2.90 ×10² ± 0.38 and at 120 days culture in the experimental ponds the TVC levels were reduced when compared to control ponds and recorded as 2.01×10² ± 0.30.

Fuller (1986) stated that, the *Lactobacillus* spp, *Bacillus* spp, and *Saccharomyces* spp are routinely used microorganisms as probiotics in animal nutrition. According to the study of Kumar *et al.,* (2007) presence of *V. harveyi* in *P. monodon* hatchery operations is very common. Luminous *Vibiro* usually occur in natural seawater; therefore seawater is a major source of luminous *Vibrio* into the hatchery facilities as per the observations of previous workers (Nair *et al.,* 1979; Ramaiah and Chandramohan, 1987; Ramesh *et al.,* 1987; Lapota *et al.,* 1988). The hatcheries vary from small-scale low-input backyard hatcheries to large-scale and high-tech hatcheries that can produce billions of post larvae (PLs) per year under strictly controlled conditions (Rosenberry, 1997). It is very important to procure quality seeds for culture operations though a *P. vannamei larva is* PCR screened at various stages in hatchery and nursery operations. The use of probiotics in culture of aquatic organisms has attained special interest in recent days towards maintaining ecofriendly practices (Gatesoupe, 1999). According to FAO/WHO (2001), the probiotics may be defined as live microorganisms which contribute major benefit to the host organism when supplemented in appropriate quantities. According to the studies of Balcazar, (2003) and Balcazar *et al.,* (2006) adequate intake of probiotics will give promising results in maintaining microbial balance, proper absorption of food, enhance the digestive enzyme activities and also to minimize the pathogenic problems in the gut of animal. The available reports on the beneficial effect of probiotics stating that, the use and applications of probiotics in aquaculture practices has quiet advantageous, because they influence the enzymatic activity which contributes good digestion, minimize the pathogen effect, to add the growth promoting factors and also involve in improvement in nutritional value of food, immune responses and maintaining good water quality parameters (Verschuere *et al.,* 2000; Ziaei-Nejad *et al.,* 2006; Wang, 2007). This is well in agreement with the results of present study that the SUPER BIOTIC applied in the experimental ponds produced promising results with reference to TVC values. According to this study the TVC levels in the experimental ponds both in summer and winter crops were minimized when compared to control ponds.

REFERENCES

Balcazar JL (2003). Evaluation of probiotic bacterial strains in *Litopeaneus vannamei*. Final Report National Center for Marine and Aquaculture Research, Guayaquil, Ecuador.

Balcázar JL, Blas I, Zarzuela-Ruiz I, Cunningham D, Vendrell D, Músquiz JL (2006). The role of probiotics in aquaculture. *Veterinary Microbiology* **114** 173-186.

FAO/WHO (2001). Report of a joint FAO/WHO expert consultation on evaluation of health and nutritional properties of probiotics in food including powder milk live lactic acid bacteria. Cordova. Argentina.

Fuller R (1986). Probiotics. *Journal of Applied Bacteriology Symposium* **15** 1-7.

Gatesoupe FJ (1999). The use of probiotics in aquaculture. *Aquaculture* **180** 147-165.

Kumar MS, George R, John KR, Jeyaseelan MJP (2007). Molecular typing of bacteria *Vibrio harveyi* and *V. alginolyticus* from shrimp farming systems. *Indian Journal of Marine Science* **36** 43-50.

Lapota D, Galt C, Losee JR, Huddell HD, Olzech JK, Nealson KH (1988). Observations and measurements of planktonic bioluminescence in and around a milky sea. *Journal of Experimental Marine Biology and Ecology* **119** 55-81.

Lee YK, Nomoto K, Salminen S, Gorbash SL (1999). Handbook of probiotics. New York, Wiley.

Nair GB, Abraham M, Natarajan R (1979). Isolation and identification of luminous bacteria from Porto Novo estuarine environs. *Indian Journal of Marine Science* **8** 46-48.

Ramaiah N, Chandramohan D (1987). Distribution and Species composition of planktonic luminous bacteria in the Arabian Sea. *Indian Journal of Marine Science* **16** 139-142.

Ramesh A, Nair GB, Abraham M, Natarajan R, Venugopalan VK (1987). Seasonal distribution of luminous bacteria in the tropical Vellar estuary. *Microbios* 52 151-159.

Sanders ME, Klaenhammer TR (2001). Invited review: the scientific basis of *Lactobacillus acidophilus* NCFM functionality as a probiotic. *Journal of Dairy Science* **84** 319- 331.

Verschuere L, Rombaut G, Sorgeloos P, Verstraete W (2000). Probiotic bacteria as biological control agents in aquaculture. *Microbiology and Molecular Biology Review* **64** 655-671.

Wang YB (2007). Effect of probiotics on growth performance and digestive enzyme activity of the shrimp *Penaeus vannamei*. *Aquaculture* **269** 259-264.

Ziaei-Nejad S, Rezaei M, Takami G, Lovett D, Mirvaghefi A, Shakouri M (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.