



# Use of Probiotics in Shrimp Farm: A study from Nagapattinam southeast coast of India

Murugesan Venkatesan\*, Radhakrishnan Jayaprakash, Selvendhiran Sudhakar

Centre of Advanced Study in Marine Biology, Faculty of Marine Sciences, Annamalai University, Parangipettai – 608 502, Tamil Nadu, India

\* Corresponding author E-mail: [venkatesa2003@yahoo.co.in](mailto:venkatesa2003@yahoo.co.in)

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## Article Info

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## ABSTRACT

*Shrimp is one of the most traded seafood commodities, and aquaculture of shrimp is considered to be one of the success stories of modern aquaculture. Antibiotic resistance in aquaculture has demanded an alternative and ecofriendly disease management approaches for the sustainable development of aquaculture sector and safe food production. Information was collected from different secondary sources, compiled systematically and arranged chronologically. Probiotics besides being beneficial bacteria also possess antiviral activity. Exploitation of these probiotics in treatment and prevention of viral diseases in shrimp aquaculture is a novel and efficient method. The benefits of probiotics and their criteria for selection in shrimp aquaculture and their role in immune power enhancement toward viral diseases are studied.*

**Keywords:** Probiotics; Shrimp Aquaculture, Nagapattinam

## 1. INTRODUCTION

Aquaculture is one of the important economic activities in many countries and offers number of opportunities to contribute to poverty alleviation, employment and community development, and reduction of over exploitation of natural resources and food security in tropical and sub-tropical regions (Ramanathan et al., 2005). Aquaculture is the culture of organisms in controlled aquatic environments (Food and Agriculture Organization, 2016). Marine shrimp aquaculture has been the mainstay of aquaculture industries due to shrimp's richness in protein supply (Food and Agriculture Organization, 2016). Shrimp aquaculture is a common practice in developing nations in Asia, Latin America and throughout the tropical

world supporting nonurban communities with means of survival, and consequently reduced poverty (Food and Agriculture Organization, 2016). It contributes to over 50% of global shrimp production as cached shrimp cannot serve the export demand for shrimp worldwide and is regarded as the most valued aquaculture business. Black tiger shrimp (*Penaeus monodon*) is the most widely cultured species in many countries, but due to the emergence of viral pathogens, many farmers have switched to the imported Pacific whiteleg shrimp, *Litopenaeus vannamei* which has been genetically selected and has since 2003 become a domesticated stock (Moriarty, 1999; Flegel, 2012). Most developing countries have embraced shrimp farming and are serving as a major export commodity (in international trade) to the

developing world. The industry is rapidly growing with 7 million metric tons annual production, generating billions of dollars every year in trade (currently worth around US\$10 billion or 16% of all fishery exports), employs millions of people globally and set for a period of strongly growing demand (Joseph, 2017). Recently, attention has focused on the use of probiotics in aquaculture. Probiotics are used as dietary supplements in aquaculture, and their role in intestinal microbial balance, growth, nutrition, health status and resistance against infectious agents are already established (Gatesoup, 1999). Probiotics may prevent potential pathogens from colonizing the gut by production of antimicrobial compounds (Robertson et al., 2000; Balcazar et al., 2006). Probiotics that have been examined for use in shrimp aquaculture include bacteria, yeasts and microalgae (Ajitha et al., 2004; Balcazar et al., 2006b). Lactobacillus, a genus of lactic acid bacteria (LAB) has been used as probiotic in shrimp aquaculture (Phianphak et al., 1999)

India is a large country with great potential for aquaculture, but the application and development of the probiotics in Indian aquaculture is very meager when compared to other countries. In recent years, the diseases of shrimp hindered the development of shrimp culture. Based on the previous research results on probiotics suggest that the use of probiotic bacteria in aquaculture has tremendous scope and the study of the application of probiotics in aquaculture have a glorious future. The role of probiotics bacteria in small culture is studied but commercial level is not that much reported especially in giant tiger shrimp, *P.monodon*. Hence the beneficial effect of probiotics on the commercial culture of Indian major candidate shrimp, *P.monodon* is very much need of the hour. Therefore, the present study was aimed to examine the effect of a probiotics; Superbiotic (water probiotic) and super PS (soil probiotic) (cp aquaculture India pvt ltd) Eco force and Exo rich (feed probiotic) on the shrimp *P.monodon* culture was studied.

## 2. MATERIALS AND METHODS

### Study area

#### Location of the farm {Farm "A" (Raja aqua farm) & Farm "B" (Senthil aqua farm)}

The farms are located on the northern bank of Villiyaru estuary in Vellakanni. The farm is situated about 12 km away from Nagapattinam. The southern

side of the farm is elevated to a height of 3.5 m from Uppanar estuary. The total area (farm "A") covered is 4.2 ha of which water spread is about 3.0 ha. Totally six pond is there, each culture pond size is .5 ha. One pond act as reservoir (1.0ha). Farm "B" also same like farm "A". The layout is given in fig.2

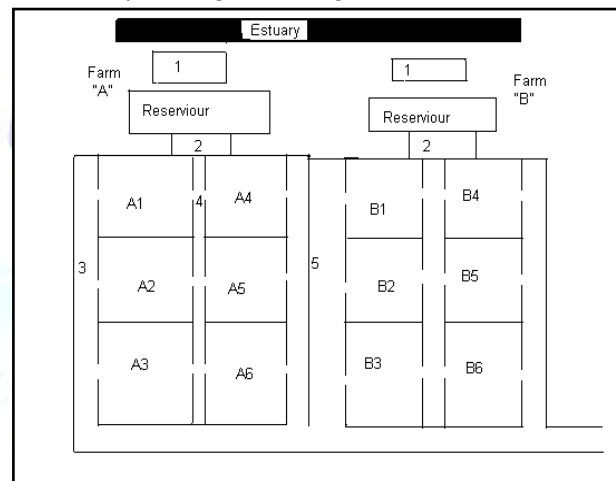


Fig. 2. Farm layout 1&2- pump house, 3&5 – drainage channel, 4 – inlet channel A1,2,3,4,5,6 & B1,2,3,4,5,6 -.5 ha

### Farm components:

#### Pump house and farm shed

The pump house is located 25 m away from the estuary and the type of pump is horizontal pump (30 HP). Farm shed is located at the other end of the farm, and nearby another shed is used for the storage of feed and other materials.

#### Diffuser tank

A concrete diffuser tank was constructed which divided in to two portions. It is square in shape however; water from the delivery point discharged through a PVC pipe in to this tank and allowed to flow in to the feeder canal. The opening of the tank is provided with two 40 micron meshes prevent the entry of predators and pests.

#### Peripheral dyke and Partition dyke

The height of peripheral dyke is 2m, the top width is of 3 m and bottom, which is of 9 m, the side slope is 1:2, the earthen dyke runs to a length of 500 m and width about 400 m. The partition dyke with a height of 2m, the top width of 1.5 m and bottom width is of 4 m the slope of the partition dyke is 1:2.



### Feeder canal and feeder canal dyke

It is trapezoidal designed water run from diffuser tank to feeder canal is through gravitational flow. The feeder canal is about 500 m in length and 0.5 m height. The feeder canal dyke is of 1.5 m depth and is lined with breaks with the size 80 cm in length and of 45 cm width.

### Inlets

There are about 3 pairs of inlets provided in each pond. The water from the feeder canal is let into the pond through 6- inch diameter PVC pipes. Inlet PVC pipes length and diameter was 1.5 m and 16 cm respectively. A filter net of 40 micron mesh size was fixed at the tip of mouth in each inlet pipes.

### Catwalks and check trays

Catwalks were made up of caesarians poles in the length of which is 4 m from the peripheral dyke and partition dyke, and each pond has 4 catwalks. The check trays are suspended in water from the catwalks (made up of iron rod in one meter round in shape.). Four check trays being provided per pond. The bases of the check trays were covered with 40 number nylon mesh.

### Culture pond and outlets

The ponds are rectangular in shape. All twell (farm "A"& Farm"B") ponds are semi intensive type with stocking densities of 10pls/m<sup>2</sup>. The depth of pond was 1.2 m and pond bed slope 30 cm from inlet point towards outlet. Monk type outlet was constructed. The outlet was constructed opposite to the inlets. The dimension of the sluice was 2m long, 0.7 m width and 2m heights. The shutter was made out of wooden planks, whereas the filter is made up of nylon mesh fitted in wooden frames.

### Aerators

Four numbers of paddle wheel aerators of 1 hp (team) Taiwan made were provided per pond. Aerators placed 5 meters from the dike, about 30 –40 m distance from each other. They were used to create the water current for the accumulation of black soil and waste in the center of the pond and also to increase the dissolved oxygen in the water column.

### Pond preparation

#### Soil culture

Initially all the ponds of the present study was allowed to dry and crack to increase the capacity of

oxidation of hydrogen sulphide and to eliminate the fish eggs, crab larvae and other predators. Then pond bottom was scrapped 2 to 4 cm by using a tractor blade to avoid topsoil. Then the pond bottom was ploughed horizontally and vertically a depth of 30 cm to remove the obnoxious gases, oxygenate the bottom soil, discolouration of the black soil to remove the hydrogen sulphide odour and to increase the fertility. The soil pH was recorded in the ponds with the help of cone type pH meter. The average pH was calculated from the collected data and required amount of lime was applied to neutralize the acid soil condition and increases the availability of nutrient (Table 1).

Table 1. Details of the lime application.

Pond	Initial soil pH	Lime applied (kg/ha)	Corrected soil pH	Type of lime applied
A1	6.6	300	7.3	Ca (OH) <sub>2</sub>
A2	6.5	400	7.4	Ca (OH) <sub>2</sub>
A3	6.3	700	7.3	Ca (OH) <sub>2</sub>
A4	6.5	400	7.2	Ca (OH) <sub>2</sub>
A5	6.6	300	7.2	Ca (OH) <sub>2</sub>
A6	6.8	200	7.1	Ca(OH) <sub>2</sub>
B1	6.6	500	7.5	Ca (OH) <sub>2</sub>
B2	6.9	200	7.3	Ca (OH) <sub>2</sub>
B3	6.2	700	7.4	Ca (OH) <sub>2</sub>
B4	6.4	600	7.3	Ca (OH) <sub>2</sub>
B5	6.8	350	7.3	Ca (OH) <sub>2</sub>
B6	6.4	500	7.2	Ca(OH) <sub>2</sub>

### Water culture

Water culture is one of the important processes during the culture period. Indeed, if the PLs are stocked in to a pond with poor algal populations, they will become stressed. That not only greatly reduces PL growth, but weakens the animals, making them much more prone to disease and subsequent death. For blooming, the pond is fertilized with inorganic or organic fertilizers.

### Chlorination

The initial water levels in all ponds were maintained at 120 cm level (Twelve culture pond and two reservoir pond). After filling need one day for sedimentation. After sedimentation process speared the chlorine with water in the pond every ever (dosage for 1 ha 1meter water level / 600 kg chlorine). After 72 hour completion, need to start the blooming process.

## Blooming

The organic fertilizers such as rice bran; groundnut oil cake, dry cow dung and yeast were soaked overnight and applied the extract to the all ponds (Table 2). The same procedure was continued for three days. After three days the water colour turned to light green. Then water level was maintained to 120 cm of the ponds and added urea and super phosphate to improve the primary production. Fertilization enhanced the optimal algal bloom in the ponds and the transparency in the ponds ranged from 33 to 36 cm. During the culture period four types of lime was used to maintain the pH and algal bloom (Table 3). During the water culture chain dragging was done daily before stocking of seeds.

**Table 2. Details of fertilizer dosage.**

Fertilizer	Dosage (Kg/ha)
<b>II. ORGANIC FERTILIZER</b>	
Rice bran +Ground nut oil cake+Dry cow dung +Yeast+Mixed with 200 liter water and soak over night	10+8+10+200g
<b>III. INORGANIC FERTILIZER</b>	
Urea + Super phosphate	7 + 5

**Table 3. Types of lime used during culture.**

Types of Lime	Chemical formula
Calcium carbonate	$\text{CaCO}_3$ (Agricultural lime)
Calcium magnesium carbonate	$\text{Ca Mg} (\text{CO}_3)_2$ (Dolomite)
Calcium oxide	$\text{CaO}$
Calcium hydroxide	$\text{Ca} (\text{OH})_2$

## STOCKING

The *P. monodon* (PL16 pass the PCR test and stress test) seeds were purchased from Tropical hatchery, Marakkanam and were transported in oxygenated double-layered polythene bags with crushed ice. Packs between inner and outer covers of the bag and packed in a carton. The seeds were brought to the farm site and bags were kept in the pond water for some time to adjust the temperature. Then the pond water was added slowly into the seed bag to adjust the salinity and pH. Subsequently the seeds were released slowly in to the ponds. The stocking density per pond was 10/m<sup>2</sup> (50,000 PLs / pond).



**PCR CHECKED SEEDS**

## Application of probiotics

Farm "A" Ponds (A1, A2, A3, A4, A5, A6) was treated with both water and soil probiotics (Superbiotic – water, super Ps – soil, manufactured by Charoen pokhpond aquaculture India Pvt. Ltd, Chennai). For Farm "B" ponds (B1, B2, B3, B4, B5, B6,) without probiotic the study was carried out (Table 4 & 5). For water quality management 1.5 kg Superbiotic mixes with 200liter water (8 hour fermentation process aerating through aquarium aerator). It was broadcasted throughout the pond during morning hours in an interval of 15 days (Probiotic application after 50 th doc 10 days once).For bottom soil quality management 10 liter super Ps probiotic was Mixed in 50 kg of dry sand then it was broadcasted throughout the pond during morning hours in an interval of 15 days (Probiotic application after 50 th doc 10 days once). For shrimp health and proper moulting, gut probiotic was given through mixing with feed {(Ecoforce and Exo rich) manufactured by Tablets India pvt ltd}.

**Table 4. Dosage of water and soil probiotic**

Days of culture	Ponds (A1,A2,A3,A4,A5,A6) (water probiotic) Superbiotic	Ponds (A1,A2,A3,A4,A5,A6) (Soil probiotic) Super ps
Before stocking	1.5 kg	10 lit
1	1.5 kg	10 lit
15	1.5 kg	10 lit
30	1.5 kg	10 lit
45	1.5 kg	10 lit
55	1.5 kg	10 lit
65	1.5 kg	10 lit
75	1.5 kg	10 lit
85	1.5 kg	10 lit
95	1.5 kg	10 lit
105	1.5 kg	10 lit
115	1.5 kg	10 lit
125	1.5 kg	10 lit



**Table. 5 Dosage of feed probiotic (1or 2 meal / day / sugar or banana binder)**

DOC	Ecoforce	Exorich
15-20	20 grm / kg	
20-30	10 grm / kg	
31-40	10 grm / kg	10 grm / kg
41-50		10 grm / kg
51-60		10 grm / kg
61-70		10 grm / kg
71-80	10 grm / kg	10 grm / kg
81-90	10 grm / kg	10 grm / kg
91-100	10 grm / kg	
101-110		
110-120	10 grm / kg	10 grm / kg
120-140	10 grm / kg	10 grm / kg

### Microbiological Analysis

For microbial analysis, the water and sediment samples were collected separately from different parts of the ponds in sterile conical flask and were mixed to make a single sample. This procedure was repeated for every pond and the final samples were brought to the

laboratory immediately and were analyzed for microbial counts. It was then transferred to a sterile conical flask (150-ml) containing 99ml of sterile diluents and serial dilution was performed to get  $10^{-1}$ ,  $10^{-2}$ ,  $10^{-3}$ ,  $10^{-4}$ , and  $10^{-5}$  suspension samples. For enumeration of Total Heterotrophic Bacteria (THB), Zobell marine agar medium (Hi-media, Mumbai) was used (Table 6). For enumeration of *Vibrio* spp TCBS MEDIA WAS OBTAINED FROM Hi-media, Mumbai.

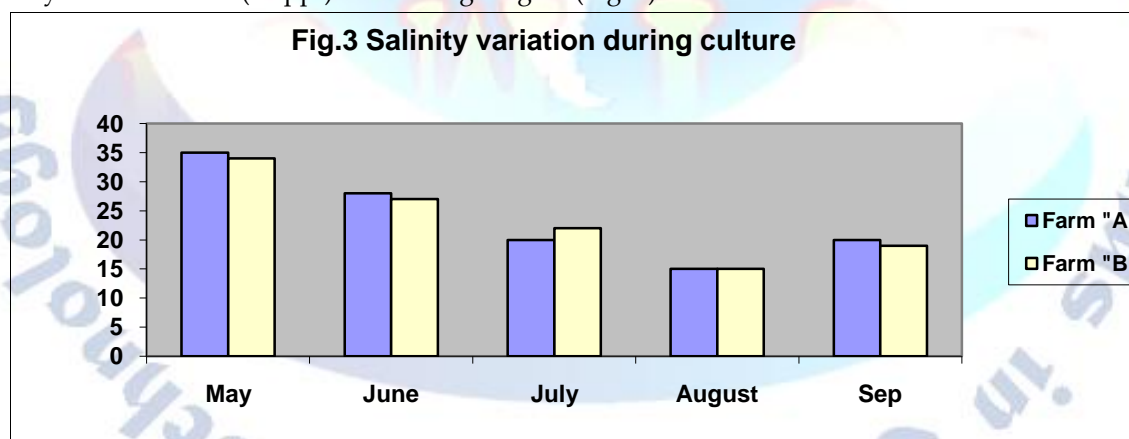
**Table 6. Composition of Zobell marine agar medium.**

Composition	Amount (g)
Peptone	5.0
Yeast extract	1.0
K <sub>2</sub> HPO <sub>4</sub>	0.5
Feso <sub>4</sub>	Trace
Agar	15
50% seawater	1000ml
pH	7.2

## 3. RESULTS

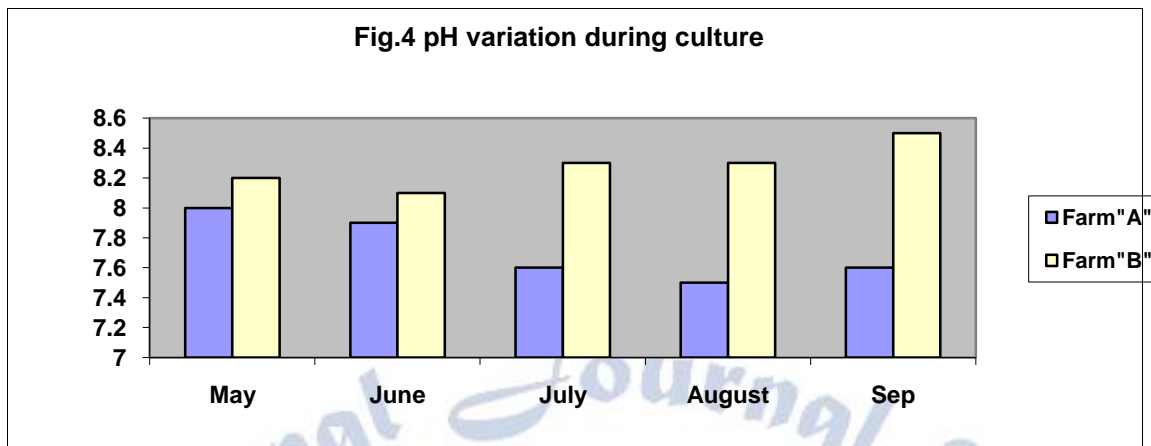
### Salinity

Water quality parameters in the culture ponds (Table. 7). The salinity was recorded maximum (35ppt) in the month of May and minimum (15 ppt) was during August (Fig. 3).



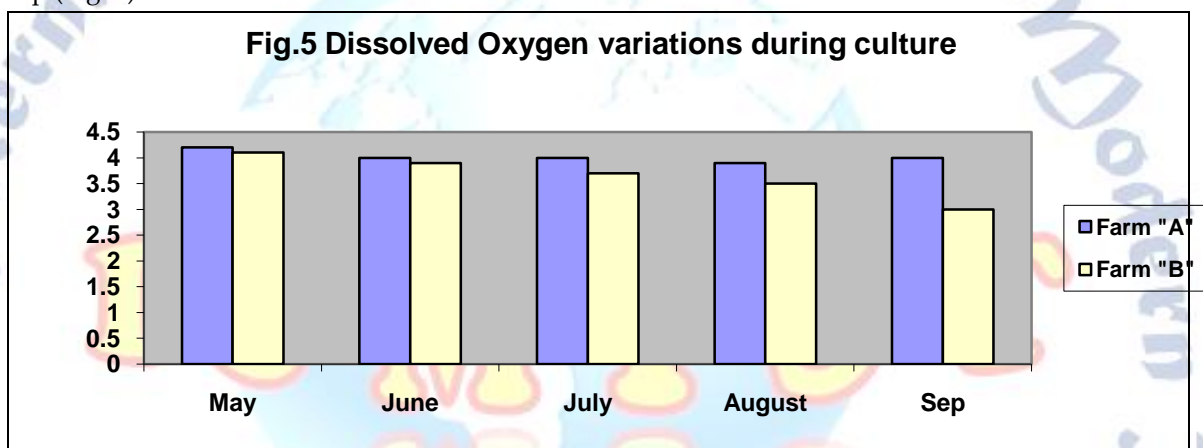
### Water pH

The pH was recorded during the culture period from May to Sep 2007. The average pH was between 7.5 to 8.2 (Fig. 4)



#### Dissolved oxygen

The dissolved oxygen was recorded maximum (4.2 ppm) during the month of May and minimum (3.0 ppm) was during Sep (Fig. 5).



#### Ammonia

The ammonia was recorded maximum (0.4) in the month of September and minimum (0.1) was during June (Fig. 6)

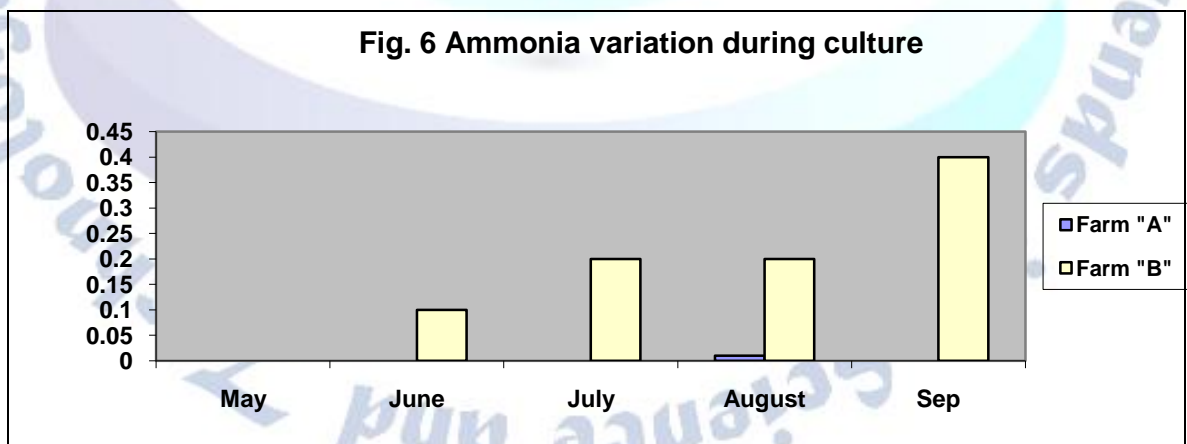
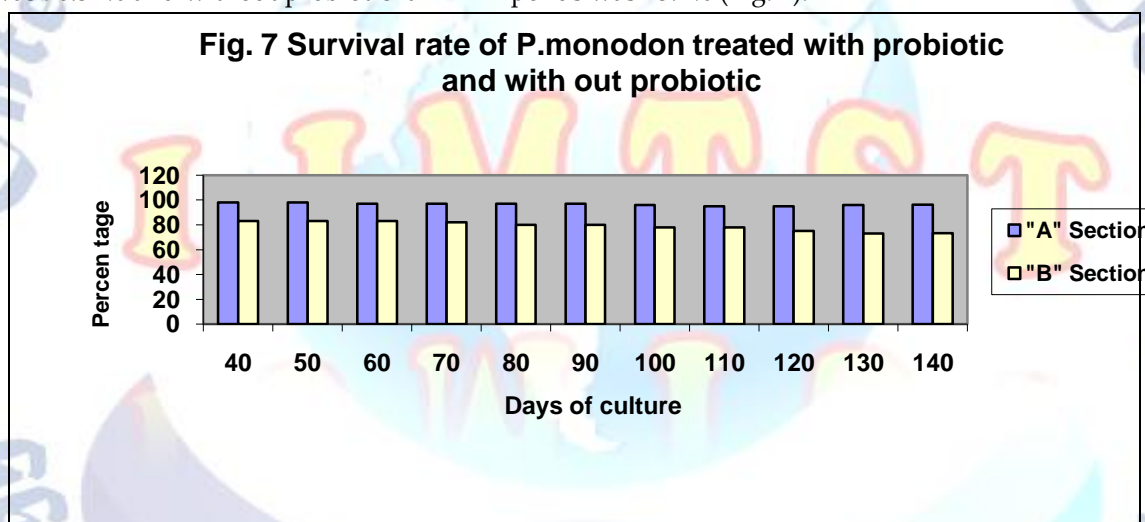


Table 7. Water quality parameters in the culture ponds

Parameters	Farm "A"	Farm "B"
Salinity	15 – 35	15 - 35
pH	7.5 - 8.0	8.1 - 8.5
Dissolved oxygen (ppm)	3.9 - 4.2	3.0 – 4.1
Temperature (°C)	28 - 30	28 - 30
Transparency (cm)	30 - 45	25 - 45
Ammonia	.01	.1 - .4

#### Survival rate

Survival rate of both the sections ("A&B" farm ponds) which was applied with probiotic was higher than that of without probiotic ponds. At the end of culture period the survival rate of farm "A" ponds with probiotic dosage was 96.34% and without probiotic farm "B" ponds was 73.2% (Fig. 7).



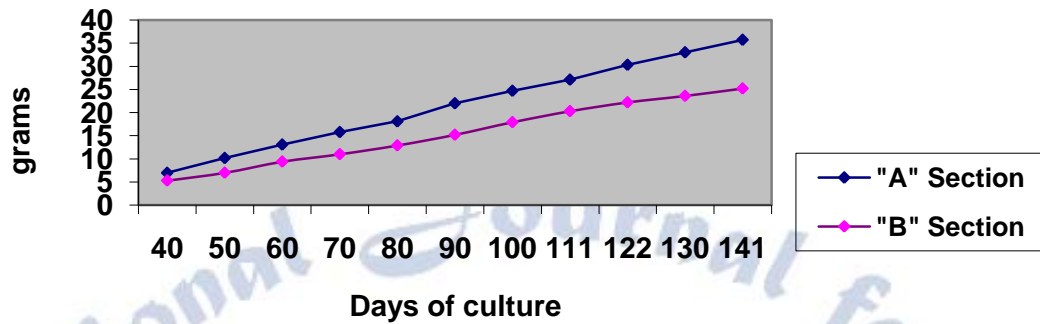
#### Growth rate

Maximum growth was observed in the "A" section ponds treated with probiotic during each sampling interval and by the end of the experiment. Average weight gained for the shrimps that were supplied with probiotic was approximately 25% greater than that of the "B" section ponds. (Fig. 8; Table. 8)

Table 8. Average body weight of the P.monodon reared in the "A & B" section ponds.

PONDS	Days of culture (DOC)										
	0	5	6	7	8	9	10	11	12	30	41
Average body weight (ABW) (g)											
"A"SECTION	0.2	1.1	3.1	5.8	8.1	2.2	4.7	7.1	0.3	3.0	5.7
"B"SECTION	.3	.7	.9	1.4	2.9	5.2	7.92	0.3	2.2	3.6	5.2

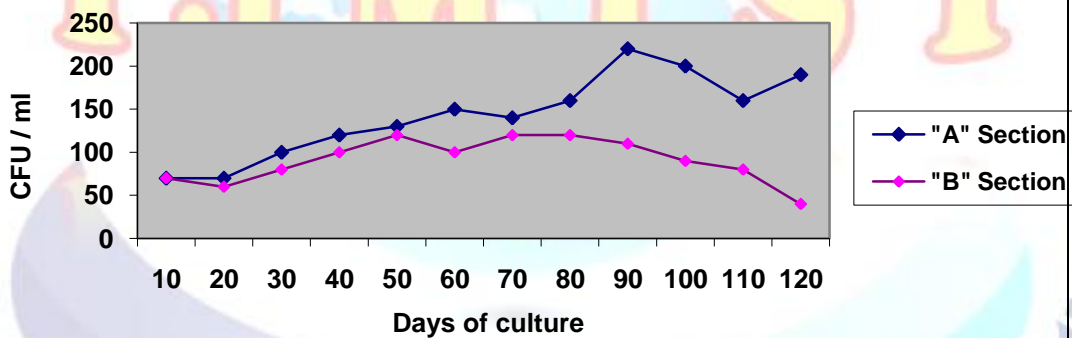
**Fig. 8 Average bogy weight of P.monodon treated with probiotic and with out probiotic**



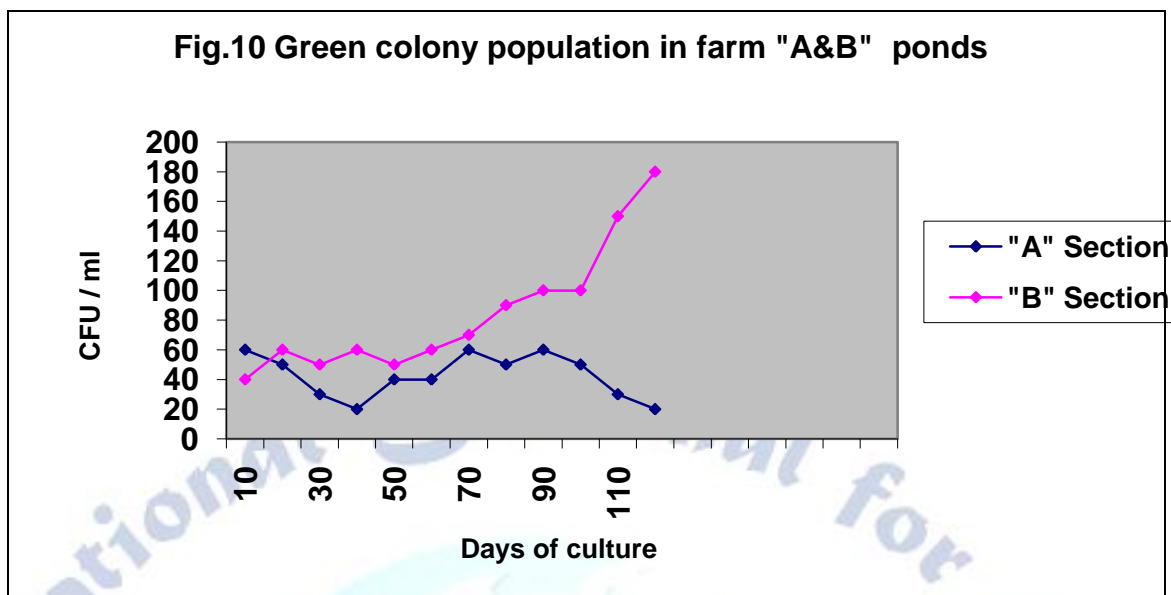
#### Microbial dynamic

The bacterial population changed during every sampling. In general, the bacterial population of sediment was higher than that of water in both "A" and "B" section ponds. The bacterial population decreased at the end of culture in farm "A" ponds, but the load of *Vibrio spp* when compared with THB in the farm "B" ponds was not showing a significant decrease. *Vibrio spp* was contributing much in the THB load of the control ponds. However among the farm "A" ponds (probiotic ponds) was showing a reduced count of *Vibrio Spp* (Fig. 9 & 10)

**Fig.9 Yellow colony population in farm "A&B" ponds**







#### ECONOMICS

Maximum net profit was obtained from farm "A" (Rs.2, 48, 516) and minimum was in the farm "B" (Rs56, 600). However the net profit from all ponds was Rs. 3, 05, 116 (Table 9).

**Table 9. Average Harvest details and economics of *P. monodon* culture ponds.**

IV. PARTICULARS	Farm	
	A	B
Pond area (m <sup>2</sup> )	5000	5,000
Stoking density/m <sup>2</sup>	10	10
Initial stock	50,000	50,000
Average daily growth	0.26	0.18
Culture period (Days)	140	141
Total Production (Kg)	1,719	915
Production Ton/ha)	3,438	1,830
Size of harvest (g)	35.7	25.20
Survival rate (%)	96.34	73.2
Feed intake	2,406	1,428
FCR	1:1.4	1:1.56
Income (Rs)	4,53,816	1,83,000
Total seed cost (Rs)	15,000	15,000
Total feed cost (Rs)	1,20,300	71,400
Other expenses (Pond preparation, water culture, probiotics & electrical charges etc.)	70,000	40,000
<b>Net Profit (Rs)</b>	<b>2,48,516.0</b>	<b>56,600.0</b>
	<b>0</b>	<b>0</b>

#### 4. DISCUSSION

There has been a considerable increase in the culture of brackish water shrimp due to its taste, market demand both national and international markets. In order to

prevent many problems due to shrimp culture, sustainable shrimp farming is the need of the hour. Ideal pond size for shrimp culture was 1 or less than 1 ha (Ramanathan *et al.*, 2005). In the present investigation also 12 ponds were used for shrimp culture and each

pond size was 0.5 ha. Even though shrimps are bottom dwelling organisms, the depth and volume of water in a pond has certain physical and biological consequences.

The present study was undertaken to ascertain the efficiency of probiotic (Superbiotic, Super ps, Ecoforce, Exo rich) on the growth and survival of the most important cultivable shrimp species, *P.monodon* in addition to its influence on important water quality parameters. Important water quality parameters monitored during the study were, temperature, salinity, oxygen, pH, and ammonia levels. The volumes of water behave like a buffer, which prevents weather fluctuations from influencing the environment in which shrimp lives. The ideal water depth is between 0.8 to 1.5 m depending upon the stage of culture. It is recommended that a minimum depth of 1m be maintained at operational level. In the present study 100 cm water level was maintained in all ponds throughout the culture period. When a pond is ready for operation, the optimum stocking density of seeds in a pond determined in accordance with the production capacity of the pond and the culture system, which included the soil and water quality, food availability and seasonal variations, target production and farmers experience (Ramanathan, 2000; Gunalan, 2006). The stocking density between 10-20PLs/m<sup>2</sup> is ideal for successful shrimp farms (Ramanathan, 2005; Gunalan 2006). In the present study the seeds were stocked at the stocking density of 10/ m<sup>2</sup> in all ponds.

The maintenance of good water quality is essential for optimum growth and survival of shrimps. The levels of physical, chemical and biological parameters control the quality of pond waters. The level of metabolites in pond water can have an adverse effect on the growth. Good water quality is characterized by adequate oxygen and limited level of metabolites. Excess feed, faecal matter and metabolites will exert tremendous influence on the water quality of the shrimp ponds. Hence critical water quality parameters are to be monitored carefully as adverse conditions may be disastrous effect on the growing shrimps (Ramanathan, 2005; Soundrapandian and Gunalan, 2008).

Salinity is important parameters to control growth and survival of shrimps. Even though *P. monodon* is euryhaline animals it is comfortable when exposed to optimum salinity. At high salinity the shrimps will grow slowly but they are healthy and resistance to diseases. If

the salinity is low the shell will be weak and prone to diseases. The salinity of the present study was maintained 15-35 ppt in all ponds (Table 7). Muthu (1980), Gunalan (2006) and Karthikeyan (1994) recommended a salinity range of 10-35 ppt was ideal for *P. monodon* culture. While Chanratchakool *et al.* (1994) and Rajalakshmi maintained the salinity of 10-30 ppt and 15-20 ppt respectively. Chen (1980) opined that salinity ranges of 15-20 ppt are optimal for culture of *P.monodon*. There are few reports (Shivappa and Hambry, 1997; Ramakrishna Reddy, 2000; Collins and Russel, 2003; soundrapandian and Gunalan, 2008), which stated that *P. monodon* adapted quite well in freshwater conditions also because of its wide range of salinity tolerance.

Water pH is one of the vital environmental characteristics, which decides the survival, and growth of shrimp under culture; it also affects the metabolism and other physiological process of shrimps. The optimum range of pH 6.8 to 8.7 should be maintained for maximum growth and production (Ramanathan *et al.*, 2005). In the present study pH was ranging between 7.5 to 8.2 for the farm "A" ponds as well as farm "B" ponds. Saha *et al.* (1995) noticed the pH of 8.11 to 8.67 in low saline ponds. Ramakrishnareddy, 2000; Gunalan, 2006; Soundrapandian and Gunalan, 2008; was recommended pH of 7.5 to 8.5 for *P.monodon* culture. The pH of pond water is influenced by many factors, including pH of source waters and acidity of bottom soil and shrimp culture inputs and biological activity. The most common cause of low pH in water is acidic bottom soil, liming can be used to reduce soil acidity. In most common cause of high pH is high rate of photosynthesis by dense phytoplankton blooms. When pH is high water exchange will be better choice (Boyd, 2001).

Dissolved oxygen plays an important role on growth and production through its direct effect on feed consumption and maturation. Oxygen affects the solubility and availability of many nutrients. Low levels of dissolved oxygen can cause damages in oxidation state of substances from the oxidized to the reduced form. Lack of dissolved oxygen can be directly harmful to shrimps and cause a substantial increase in the level of toxic metabolites. Low-level of oxygen tension hampers metabolic performances in shrimp and can reduce growth and mounting and cause mortality (Gilles Le Molluae, 2001). The dissolved oxygen in all the culture



ponds in the present study was ranging between 3.0 to 4.2 ppm (Table, 7).

Water temperature is probably the most important environmental variables in shrimp cultures, because it directly affects metabolism, oxygen consumption, growth, moulting and survival. In general, a sudden change of temperature affects the shrimp immune system. The optimum range of temperature for the black tiger shrimp is between 28 to 30°C (Ramanathan *et al.*, 2005). The temperature in the present study was 28 to 30°C and the low temperature 28°C were observed due to cloudy weather (Table 7). The optimum range of temperature of *P. monodon* was between 26 to 33°C (Mayaule, 1997; Gunalan, 2006) and temperature range of 28 to 33°C supports normal growth (MPEDA) as observed in the present study.

The transparency is mainly depends on the presence of phytoplankton. The secchi disc reading should be between 30-40 cm (MPEDA, 1986). The optimum range of secchi disc reading is between 30 to 60 cm to the juvenile stage and between 25 to 40 cm to the sub adult and final stage. The transparency of the present study is 25 to 45 cm (table 7). Ramakrishnareddy (2000), Gunalan (2006) also observed similar transparencies (25-50 cm) for his study. The reading less than 30 cm mean that the phytoplankton density is high. If it is more than 40 cm indicates, low population of phytoplankton. For the growth of phytoplankton adequate quality of sunlight is needed. Due to low intensity of light during the culture period, the plankton bloom was less. Hence, the transparency was more.

Ammonia is the principal end product of protein catabolism of organisms and it is excreted through gills. It is also formed by decay of organic matter. Under anaerobic condition, sulphate is also reduced to ammonia. In present study, there was no such change because the culture medium was continuously aerated. Collapse of cyanobacteria bloom also leads to increase ammonia. When the ammonia concentration in the culture medium increase, excretion of ammonia by cultured organisms decreases. Under farm conditions, the ammonia level should be less than 1 ppm. In present study in the farm "B" ponds, the level was well below this mark. But in the farm "A" ponds, very low level ammonia problem recorded. This is mainly due to the microorganisms (nitrosomonas) present in the probiotics which initiate nitrification. Due to this process ammonia

is converted into nitrite which is further acted upon by the nitrobacteria and converted as nitrate through the process nitrification. In farm "B" ponds the shrimps were affected by black gill disease, tail rot disease however this is absents in probiotic used farm "A" ponds. This is mainly due to the absence of probiotic in the farm "B" ponds. Ravi *et al.*, (1998) described the benefits of probiotics in maintaining water quality and enhancing growth rate in India Whit Prawn, *P. indicus*.

Shrimp aquaculture production in much of the world is depressed by disease, particularly caused by luminous *Vibrio* and/or viruses. Antibiotics, which have been used in large quantities, are in many cases ineffective, or result in increases in virulence of pathogens and, furthermore, are cause for concern in promoting transfer of antibiotic resistance to human pathogens. Probiotic technology provides a solution to these problems. The microbial species composition in hatchery tanks or large aquaculture ponds can be changed by adding selected bacterial species to displace deleterious normal bacteria. Virulence of luminous of *Vibrio* species can be controlled in this manner. Abundance of luminous *Vibrio* strains decreased in ponds and tanks where specially selected, probiotic strains of *Bacillus* species were added. A farm on Negros, in the Philippines, which had been devastated by luminous *Vibrio* disease while using heavy doses of antibiotic in feed, achieved survival of 80-100% of shrimp in all ponds treated with probiotics (Moriarty, 1998).

Ruangpan and Kitao (1991) reported in their study that the high abundance of luminescent *Vibrio* is consistent with occurrence of disease and poor or zero harvest results. *V. harveyi*, a pathogen of *P. monodon* that causes severe losses (Baticados *et al.*, 1990). The farm, which used the Super biotic probiotic bacteria, had either a very low abundance or a complete absence of luminous *Vibrio* in pond water and very good harvest result. This consistent and high productivity occurred, even though the proportion of luminescent *Vibrio* in the pond water was high in the sea water source, and the abundance of total green colony in the pond water was higher than in the water source. Furthermore, luminescent *Vibrio* were completely absent at all stages of grow out from the pond sediment in the presence of the Super biotic *Bacillus* species. Shrimp were healthier in ponds with super biotic, super ps, Ecoforce and Exorich.



The probiotic treated (Farm "A") ponds in the present study had either a very low abundance or a completed absence of luminous and very good survival was achieved. This result is comparable with the study of Dalmin et al. (2001). Colonization of the gastrointestinal tract of animals by probiotics is only after birth, and before the definitive installation of a very competitive indigenous micribita.

After this installation, only the addition of doses of probiotic provokes its artificial and temporary dominance. In mature animals, the population of probiotic organisms in the gastrointestinal tract shows a sharp decrease (Fuller, 1992). Application of microbial supplement in the probiotic ponds hindered the growth of *Vibrio* spp, like *V. alginolyticus* and *V. harveyi* because of the colonization of the beneficial microbes like *Bacillus* sp., *Pseudomonas* sp., *Lactobacillus* sp, and *Saccharomuces* sp, in the shrimp gut. Since the shrimp in the farm "B" ponds were dominated with green colony, which caused Vibriosis can be attributed as the reason for low survival in the farm "B" ponds when compared with the farm "A" ponds. This was evident from the presence of higher load of green colony, in the water and sediment of without probiotic ponds than in the probiotic used ponds. The occurrence of green colony in the farm "B" ponds was concluded by presence of luminescence in the nighttime and occurrence of dead animals in the check tray. In the present study white gut disease was reported in farm "B" ponds, which ultimately leads fungal diseases, naturally animal's activities slowed down and become sluggishness finally leads to fungal disease. The white gut and fungal disease are not observed in probiotic treated (farm "A") ponds.

The general conclusion obtained from the present study is that the probiotics plays a vital role in growth, survival and disease resistance of the animal by maintaining good water quality parameters throughout the culture period. It is clear from the microbial load data that green colony is dominant in the farm "B" (without probiotic) ponds. Besides green colony, the shrimps in the farm "B" ponds also affected by black gill, white gut and fungal diseases.

## 5. CONCLUSION

The culture of shrimp received maximum importance due to its unique taste, high nutritive value and persistent demand in the world market. In recent

years, the diseases of shrimp hindered the development of shrimp culture. In the present study an attempt has been made to culture the giant tiger shrimp, *P. monodon* in (two farms) 12 ponds each with 0.5ha near Vellakanni village of, Nagai district, Tamilnadu. Hence, the use of probiotic bacteria in aquaculture has tremendous scope and the study of the application of probiotics in aquaculture has a glorious future. In the present study the probiotics, Superbiotic, super ps, Ecoforce and Exorich was applied in culture ponds of *P. monodon* with different dosages which is compared with farm "B" (without probiotics). Growth and survival rate of the ponds which was applied (farm "A" ponds) with probiotic was higher than that of farm "B" ponds. Concentration of was nil for the probiotic treated ponds than the farm "B" ponds. Chlorophyll was observed maximum in probiotic ponds rather than farm "B" ponds. The bacterial population decreased at the end of culture in farm "A" ponds, but in farm "B" ponds the bacterial population was not showing a significant decrease. Black gill, white gut and fungal diseases were recorded in farm "B" ponds. But these diseases were not that much reported in probiotic treated (farm "A") ponds. The general conclusion obtained from the present study is that the probiotic plays a vital role in growth, survival and disease resistance of the animal by maintaining good water quality parameters throughout the culture period. It is clear from the bacteria colony data that green colony dominant only in the farm "B" ponds. So it can be recommended that the probiotics Superbiotic, super ps, Ecoforce and Exorich can be very well utilized for the shrimp ponds to get reasonable growth and survival.

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## Conflict of interest statement

Authors declare that they do not have any conflict of interest.

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