

# ROLE OF PROBIOTICS IN SUSTAINABLE SHRIMP FARMING ALONG BALASORE COAST, ORISSA

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## **INTRODUCTION**

The Probiotics is "a live microbial feed supplement which beneficially affect the host animal by improving its intestinal microbial balance" (Fuller, 1992). The scientific shrimp culture started during 1980s. There is about 1.2 million hectare of brackish water available for shrimp culture in India. This shrimp culture is carried out by traditional, extensive, semiintensive and intensive method. The out break of disease occurred during 1995 and by 1996 it had severely affected in East Asia and south East Asia which has intensive in Andhra Pradesh. The shrimp culturist beard a heavy loss during that time and the export of shrimp to other country suffered a lot. The researcher attempted to come out from this situation with the application of different antibiotics which was not successful due to antibiotics resistance of organism. Now shrimp culturists are using probiotics to overcome this precarious situation and adopt sustainable and eco-friendly practices. The term sustainability can be defined as environmental friendly, social acceptable, economical viable and technically feasible so in this context the periodic interval application of probiotics play a secrete energy for shrimp throughout the culture and maintain culture sustainable year after year. The use of probiotics in human and animal were documented (Sission, 1989; Fuller, 1992; Maeda and Liao, 1992, 1994; Douillet and Langdon, 1994; Austin et al., 1992, 1995; Garriques and Arevalo, 1995; Moriarty, 1997, 1998; Moulder, et al., 1997; Rengpipat et al., 2000; Rinkinen et al., 2003; Balcazar et al., 2006). Gatesoupe (1999) have studied use of Probiotics in aquaculture and most attempts to purpose by isolating and selecting strains from aquatic environment.

**ABSTRACT** The present study has been undertaken to know the role of probiotics in sustainable shrimp farming along the coastal Balasore of Orissa. The hydrological parameters like water temperature, pH, salinity, transparency and alkalinity were observed weekly where as hardness was observed every 20 days of culture (DOC) for a period of six month and dissolved oxygen was observed every day in the early morning and evening throughout the culture beginning from 30 days of culture. The highest water temperature was observed to be 30.3°C and the lowest was 25.33°C. The highest water pH was observed be 8.53 and the lowest was 7.26. The highest salinity was observed to be 24.66 ppm and lowest was 11.66 ppm. The highest transparency was observed to be 43 cm and lowest was 17 cm. Likewise, the highest carbonate was observed to be 57.33 ppm and lowest was 11.33 ppm. The highest bicarbonate alkalinity was observed being 285 ppm and lowest was 100 ppm. The hardness varied from 8443 ppm to 3533 ppm. The highest and lowest dissolved oxygen was observed to be 4.53 ppm and 3.53 ppm during culture period. Five different commercial brands of Probiotics of unique composition were used for different ponds during every month. Though bacterial load present inside the culture ponds throughout the culture period it does not show any problem due to application of probiotics which helped for suppression and maintaining of a clean and hygienic environment for sustainable shrimp culture.

> Rengpipat et al., (2000) have studied immunity enhancement in black tiger shrimp (*P.monodom*) by probiont bacterium (Bacillus-11). Gatesoupe (2002) have studied probiotic and formaldehyde treatments of Artemia nauplii as food for larval Pollack Pollachius pollachius. Abraham (2004) has studied antibacterial marine bacterium deter luminous vibriosis in shrimp larvae and suppressed the activity of V.harvayi M3 and reduced mortality of P.monodon. Burgents et al., (2004) have observed the disease resistance of pacific white Shrimp Litopenaeus vannamei, following the dietary administration of a yeast culture food supplement. So far application of probiotics in shrimp culture in India is concerned Dalmin et al., (2001) studied the effect of probiotics on bacterial population and health status of shrimp in culture pond eco-system. Lakshman and Sundarpandian (2008) have studied effect of commercial probiotics on large scale culture of black tiger shrimp Penaeus monodon (Fabricus). Sundarapandian and Sankar (2008) have studied effect of Probiotics on the survival and production of black tiger Shrimp Penaeus monodon (Fabricus). No or little work has been done on application Probiotics in Orissa in general and Balasore coast in particular. Therefore the present study has been under taken to study the role of probiotics for sustainable shrimp farming in Balasore coast.

### MATERIALS AND METHODS

The study was carried out for one culture operation *i.e.*, June – 2009 to November 2009. The ponds are located on Gudupai of Balasore district of Orissa named as Eastern India Fisheries and Agro Product Pvt. Ltd. The farm is located 12 km away

from Balasore town towards east nearer to Chandipur coast. The farm is having a total of eleven number of pond out of which 5 ponds are selected for the experimental purposes. Each pond was having an area of 0.8 ha and stocking density was 12 nos/m<sup>2</sup>. The water quality parameter like temperature, pH, salinity, transparency and alkalinity were tested in every week. dissolved oxygen was tested twice a day in every morning and evening *i*.e. at 5 am and 5 pm The temperature was observed in field using a centigrade thermometer of 0.1°C accuracy. pH was tested using digital pH meter. Salinity test was conducted by field refractometer. Likewise, transparency was tested by sacchi disc in field. The parameters like alkalinity, dissolved oxygen and hardness was tested following standard method of APHA (2005). Five commercial brand of pond probiotic were selected for application in all five ponds like Ecomax, Super biotic, Environ AC, Prolig HQ, Bioremid -Aqua of different companies like Biosolution Thiland, CP Aqua Pvt. Ltd, Biostad India Ltd and Neosparck Pvt. Ltd. A Gut probiotic named Ecoforce of Tablet India Ltd was selected for application in all the five ponds. The wave brand feed of Water base Ltd was selected as shrimp feed throughout the culture period. For microbial analysis, the water sediment was collected separately from different part of the ponds in sterile conical flask and was 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 the microbial counts. It was then transferred to a sterile conical flask (150 mL) containing 99 mL of sterile diluents and sterile dilution was performed to get 10<sup>-1</sup>, 10<sup>-2</sup>, 10<sup>-3</sup> and 10<sup>-4</sup> suspension sample for enumeration of total vibrio(Kannan, 1993). The TCBS (Tri ethyl citrate bile salt sucrose) agar media (Hi-media, Mumbai) was used to know the significance between the parameters of probiotics treated pond. The analysis of vibrio was made at an interval of 30 days of culture (DOC) during whole culture period. The vibrio colony estimation was made by serial dilution method and the vibrio colony was expressed as colony forming unit per mL (cfu/mL).The biomass was calculated taking average harvested shrimp from each pond and expressed in kg. The food conversion ratio (FCR) and survival percentage was calculated for each pond by the following formula (Cruz, 1991).

> FCR = Total Biomass Harvested Total Feed Consumed Survival% = Total Biomass Harvested Average Body Weight

### RESULTS

The study was conducted for 130 days of culture which started in last week of June and harvested in first week of November 2009.The pond area was about 0.8 hectare each having stocking density of 96,000 number of seed in each pond. The average water quality parameter of five different ponds during different days of culture was given in Table 1 to 5. The highest water temperature was observed in pond 1 on 20 days of culture being 30.3°C. The lowest water temperature was observed in pond 1 and 5 after 120 days of culture being 25.33°C (Table 1 to 5). The highest water pH was observed in pond 3 on 20 days of culture being 8.53 and the lowest was 7.26 in pond 4 after 120 days of culture (Table 3 and 4) .The highest salinity was observed to be 24.66 ppm in pond 5 on 20 days of culture and lowest was 11.66 ppm in pond 4 after 120 days of culture (Table 4 and 5). The highest transparency was observed to be 43 cm in pond 5 on 20 days of culture and lowest was 17.33 cm in pond 2 on 20 days of culture (Table 2 and 5). Likewise, the highest carbonate was observed to be 57.33 ppm in pond 3 on 20 days of culture and lowest was 11.33 ppm in pond 4 on 80 days of culture (Table 3 and 4). The highest bicarbonate alkalinity was observed being 283.33 ppm in pond 2 on 120 days of culture and lowest was 100 ppm in pond 1 after 80 days of culture .The total hardness varied from 8443 ppm in pond 1 after 80 days of culture and lowest was observed to be 3533 ppm in pond 4 after 80 days of culture(Table1 and 4). The highest and lowest dissolved oxygen was observed to be 4.53ppm and 3.53 ppm on 20 and 120 days of culture in all the ponds. The vibrio colony forming unit was highest being 26×10<sup>8</sup> in pond 4 when applied with Prolique-HQ brand and lowest was 1.4×10<sup>8</sup> pond 1 when applied with Ecomax brand (Table 6). The biomas was highest in pond 1 being 2,432 kg when total feed consumption was 4465.7 kg (Table 7). Likewise the highest survival was found in pond 3 being 75% and the lowest survival was found in pond 4 being 61%.

#### DISCUSSION

The important water quality parameters monitored during the Penaeus monodon culture ponds were water temperature, pH, salinity, alkalinity, transparency, total hardness and dissolved oxygen. There was no significant difference (p>0.05) between physical parameter such as temperature, dissolved oxygen, pH, salinity and transparency among the ponds and these were all at optimum level for P.monodon culture (Table 1 to 5). Water temperature fluctuation showed similar pattern among the ponds *i.e.* it is high in the starting of culture and it is gradually decreased at the end of the culture which may be due to hot climate in beginning of the culture period and cold climate at the last part of the culture. Generally dissolved oxygen and pH values in all the ponds reached critical levels (Table 1 to 5) which may be due to aeration. According to Neori et al., (1989) the dissolved oxygen level and pH was maintained with in acceptable limit using aeration device. The pH value in present study varied from 7.2 to 8.5 and this result is inagreement with the result of Ramanathan et al., (2005) and Reddy (2000) as the optimum range of pH was 6.8 to 7.8 maintained for maximum growth and production of penaeid species. Transparency was high in beginning of the culture period because there was no much plankton in the ponds. But at the end of the culture period the transparency was less because of the growth of the phytoplankton in the ponds due to addition of feed materials in the ponds. Reddy (2000) has observed the transparency from culture ponds of Nellore Andhra Pradesh and reported that the value varied from 25-50cm. Similarly Anonymous (2006) at Tamilnadu has observed the Sacchi disc value in culture ponds between 30-40 cm. Yosoff et al., (2002) have observed that the transparency was less i.e. 30 cm due to high density of phytoplankton and the transparency was more *i.e.* 50 cm due

Days of culture	Water Temp <sup>o</sup> C	рН	Salinity ppm	Alkalinity pp Carbonate	m Bi Carbonate	Transparency cm	Total Hardnes ppm	s Dissolved Oxygen p	pm
								5 AM	5PM
20	$30.33 \pm$	$8.39 \pm$	$21.16 \pm$	$32\pm$	$197.33 \pm$	$36.33 \pm$	$5190 \pm$	$4.533 \pm$	$5.50 \pm$
	1.154	0.011	1.285	6.928	99.30	7637	65.574	0.305	0.201
40	$28.33 \pm$	$8.27 \pm$	$17.3 \pm$	$39.33 \pm$	$202 \pm$	$30.33 \pm$	$5716.66 \pm$	$4.10 \pm$	$5.4 \pm$
	2.001	0.070	2.081	14.6	72.74	5.033	104.083	0.101	0.556
60	$27.33 \pm$	$8.4 \pm$	$15.00 \pm$	$36.00 \pm$	$245.33 \pm$	$20.00 \pm$	$6616.66 \pm$	$4.266 \pm$	$4.65 \pm$
	0.577	0.257	0.001	4.00	8.08	0.001	812.91	0.208	0.519
80	27330 ±	$8.4\pm$	$14.3 \pm$	$39.3 \pm$	$100 \pm$	$21.00 \pm$	$8443.33 \pm$	$4.10 \pm$	$4.15 \pm$
	1.00	0.034	0.577	2.05	17.32	1.001	1294.38	0.100	0.608
100	$25.66 \pm$	$8.24 \pm$	$13.3 \pm$	$27.33 \pm$	$170 \pm$	$23.66 \pm$	$5373.33 \pm$	$3.766 \pm$	$4.6 \pm$
	0.577	0.063	0.577	8.00	17.32	1.527	799.77	0.152	0.115
120	$2533 \pm$	$791 \pm$	$12.6 \pm$	$25.33 \pm$	$270 \pm$	$20.66 \pm$	$4283.33 \pm$	$3.533 \pm$	$4.55 \pm$
	0.577	0.432	0.57	5.03	17.32	1.154	125.83	0.351	0.351

Table 1: Average water quality parameter of pond-1 during different days of culture at E.I.F.A. (Eastern India Fisheries and Agro product Pvt LTD)

Table 2: Average water quality parameter of pond-2 during different days of culture at E.I.F.A. (Eastern India Fisheries and Agro product Pvt LTD)

Day of	Water	pН	Salinity	Alkalinity in	PPM	Transparency	Total Hardness	Dissolved	Oxygen
culture	Temp <sup>o</sup> C		ppm	Carbonate	Bi Carbonate	cm	ppm	ppm	
								5AM	5PM
20	$29.56 \pm$	$8.45 \pm$	$22.33 \pm$	$45.33 \pm$	$138 \pm$	$38.33 \pm$	$4230\pm$	$4.533 \pm$	$5.5 \pm$
	0.404	0.155	0.577	4.61	41.56	7.094	306.104	0.305	0.201
40	$27.66 \pm$	$8.29 \pm$	$19.16 \pm$	$38.66 \pm$	$214.66 \pm$	$32.22 \pm$	$4916.66 \pm$	$4.100 \pm$	$5.4 \pm$
	0.577	0.132	2.598	18.90	87.32	2.516	1277.04	0.100	0.556
60	$26.16 \pm$	$8.44 \pm$	$16.50 \pm$	$44 \pm$	$230 \pm$	$24.00 \pm$	$6424.66 \pm$	$4.266 \pm$	$4.65 \pm$
	0.288	0.049	0.501	24.33	17.32	1.00	1020.26	0.208	0.519
80	$26.00 \pm$	$8.31 \pm$	$16.00 \pm$	$31.33 \pm$	$180 \pm$	$22.33 \pm$	$4133.33 \pm$	$4.100 \pm$	$4.15 \pm$
	1.00	0.045	1.001	8.08	1.100	1.154	208.16	0.100	0.608
100	$26.06 \pm$	$8.28 \pm$	$14.00\pm$	$32 \pm$	$183.3 \pm$	$22.33 \pm$	5216.66 <u>+</u>	$3.766 \pm$	$4.6 \pm$
	0.901	0.063	1.002	6.920	25.16	2.516	897.68	0.152	0.115
120	$25.6 \pm$	$7.91 \pm$	$11.83 \pm$	$16.66 \pm$	$283.33 \pm$	$17.33 \pm$	$4283.33 \pm$	$3.533 \pm$	$4.55 \pm$
	0.577	0.270	0.763	5.77	25.16	6.429	125.83	0.351	0.351

Table 3: Average water quality parameter of pond-3 during different days of culture at E.I.F.A. (Eastern India Fisheries and Agro product Pvt LTD)

Day of culture	Water Temp <sup>o</sup> C	рН	Salinity ppm	Alkalinity ppn Carbonate	n Bi Carbonate	Transparency cm	Total Hardness ppm	Dissolved C ppm	Dxygen
								5AM	5PM
20	$27.33 \pm$	$8.53 \pm$	$22.33 \pm$	$57.33 \pm$	$177.33 \pm$	$40.66 \pm$	$4570 \pm$	$4.533 \pm$	$55 \pm$
	0.763	0.011	0.577	9.237	12.70	5.686	403.360	0.305	0.210
40	$28.16 \pm$	$8.28 \pm$	$19.33 \pm$	$53.33 \pm$	$187.33 \pm$	$31.66 \pm$	$4599 \pm$	$4.1 \pm$	$5.4 \pm$
	0.763	0.5057	2.081	16.289	31.64	6.350	300.0101	0.1.00	0.556
60	$26.33 \pm$	$8.30 \pm$	$15.66 \pm$	$30.33 \pm$	$196.66 \pm$	$25.33 \pm$	$6600 \pm$	$4.35 \pm$	$465 \pm$
	1.154	0.110	0.577	13.051	35.11	2.516	813.535	0.208	0.519
80	$26.166 \pm$	$8.35 \pm$	$14.66 \pm$	$32 \pm$	$186.66 \pm$	$23.00 \pm$	$4566.66 \pm$	4.1±	$4.15 \pm$
	0.763	0.086	0.288	6.920	30.55	2.645	351.18	0.1.00	0.608
100	27±	8,35±	$14.00 \pm$	$33.33 \pm$	$170.66 \pm$	$22.33 \pm$	$4350 \pm$	$3.85 \pm$	$4.6 \pm$
	0.866	0.089	0.001	7.023	44.06	2.516	1143.460	1.527	0.115
120	$26.4 \pm$	7.71±	$12.3 \pm$	$24.66 \pm$	$246.66 \pm$	$22.33 \pm$	$4246.66 \pm$	$3.533 \pm$	$4.55 \pm$
	1.216	0.572	1.101	15011	49.32	2.081	332.46	0.351	0.351

to low density of phytoplankton. The present result is in partial agreement with the result of Reddy (2000) and Anonymous (2006) which may be due to different in phytoplankton production and the turbidity of water in different culture ponds. *Penaeus monodon* is a euryhaline species and can be raised in lower salinities. Salinity is the most important factor commonly affecting the growth of the penaeid shrimp (Gunter, 1961; Gunter *et al.*, 1964; Willams, 1965). For better growth of the shrimp the salinity should not be less then 5 ppm. Under laboratory condition Nair and Krishnankutty (1975) observed that the growth rate of *Penaeus indicus* is significantly

high and salinities of 10 ppm for post larval stage. The salinity varied from 11 to 24 ppm during the present study period. The fluctuation of salinity during culture period was with in range of optimal level for the growth of Penaeid shrimp which was inagreement with the work of Nair and Krishnankutty (1975) and Hudinga(1942). The bicarbonate alkalinity is more in all the ponds and this was due to heavy rain in mid of the culture period and addition of fresh water by pumping of ground water in to the ponds. This result is inagreement with the result of Putheti and Leburu (2009). The carbonate and bicarbonate values were with in range of 16 to57 ppm and

Day of culture	Water Temp <sup>o</sup> C	рН	Salinity ppm	Alkalinity pp Carbonate	om Bi Carbonate	Transparency cm	Total Hardness ppm	Dissolved ( ppm	Dxygen
								5AM	5PM
20	2866± 2.081	$\begin{array}{c} 8.44 \pm \\ 0040 \end{array}$	4.33 ± 0.577	41.33± 16.165	146± 95.84	40.5± 6.264	4300± 1081.66	$4.533 \pm 0.305$	5.5± 0.210
40	$27.00 \pm 0.50$	8.22± 0.190	19.00± 0.001	20± 18.330	175.33± 41.19	29.66± 3.785	5100± 435.88	4.10± 0.10	5.4± 0.556
60	26.4± 1.442	$8.30 \pm 0.075$	16.03± 1.401	35± 14.524	170± 17.32	24.33± 2.081	6299± 700.011	$4.35 \pm 0.208$	4.65± 0.519
80	26.96± 1.078	8.24 ± 0.110	14.9± 0.655	11.33± 1.154	183.33 ± 20.81	33 ± 1.154	3533.33± 907.37	4.10± 0.101	4.15± 0.608
100	26.66± 1.527	8.34± 0.017	12.33± 0.577	17.33± 309	193.33 ± 30.55	21.33 ± 2.309	4166.66± 945.16	3.85± 1.527	4.6± 0.115
120	$25.4 \pm 0.692$	7.26± 0.210	11.66± 0.577	$\begin{array}{c} 12.66 \pm \\ 6.429 \end{array}$	193.33± 45.09	$\begin{array}{c} 21.66 \pm \\ 2.886 \end{array}$	4300± 721.110	3.533± 0.351	4.55± 0.351

Table 4: Average water quality parameter of pond-4 during different days of culture at E.I.F.A. (Eastern India Fisheries and Agro product Pvt LTD)

Table 5: Average water quality parameter of pond-5 during different days of culture at E.I.FA (Eastern India Fisheries and Agro product Pvt Ltd)

Day of	Water	рН	Salinity	Alkalinity ppr	n Ri Carbonata	Transparency	Total Hardness	Dissolved Oxygen	
culture	remp* C		ррт	Carbonale	BI Carbonale	cm	ррт	5AM	5PM
20	30.33 ± 0.577	8.34± 0.017	24.66 ± 1.154	43.33 ± 5.773	146± 22.53	43.66± 4725	4266.66± 971.253	$4.53 \pm 0.305$	$5.50 \pm 0.210$
40	27.66 ± 1.527	$7.88 \pm 0.409$	$\begin{array}{c} 19.00 \pm \\ 0.00 \end{array}$	23.33 ± 14.189	174.66 <u>+</u> 78.01	35.33± 4.04	4133.33± 503.322	4.100± 0.100	$5.4 \pm 0.556$
60	27.66 <u>+</u> 1527	8.51 <u>+</u> 0.334	16.66± 1.527	40.66± 11.015	204 ± 22.53	$24.66 \pm 5.03$	6100± 458.257	$4.35 \pm 0.208$	4.65± 0.519
80	25.73 <u>+</u> 1.101	$8.29 \pm 0.092$	14.33± 1.154	31.33 ± 14.742	188.66 <u>+</u> 22.53	25.00± 0.01	5083.33 ± 1153.617	4.100± 0.100	4.15± 0.608
100	26.83 ± 1.607	$8.23 \pm 0.092$	12.00± 0.010	29.33 ± 3.055	176.66± 15.27	22.33± 1.15	5933.33 ± 461.88	3.85± 0.152	4.6± 0.115
120	25.33± 0.577	8.23± 0.017	$12.00 \pm 0.101$	20.00± 0.101	216.66± 49.32	$\begin{array}{c} 22.00 \pm \\ 2.00 \end{array}$	6150± 726.292	3.533± 0.351	$4.55 \pm 0.351$

#### Table 6: Vibrio colony estimation in different ponds of different days of culture in colony forming unit (Cfu/mL)

Pond No.	Eco-Max		Superbiotio	2	Environ AC		Prolique	HQ	Bioremid	
and Brands										
Colony Cfu / mL	Y.C.	G.C.	Y.C.	G.C.	Y.C.	G.C.	Y.C.	G.C.	Y.C.	G.C.
Vibrio before stocking	1 x 10 <sup>8</sup>	1.4 x 10 <sup>6</sup>	9 x 10 <sup>7</sup>	2 x 10 <sup>7</sup>	1.5 x 10 <sup>7</sup>	5 x 10 <sup>7</sup>	2 x 10 <sup>8</sup>	26 x 10 <sup>8</sup>	1 x 10 <sup>8</sup>	3 x 10 <sup>7</sup>
After Doc 30	6 x 10 <sup>7</sup>	_	3.5 x 10 <sup>7</sup>	1 x 10 <sup>6</sup>	1 x 10 <sup>8</sup>	_	2 x 10 <sup>7</sup>	1.6 x 10 <sup>6</sup>	5 x 10 <sup>7</sup>	_
After Doc 60	5 x 10 <sup>7</sup>	2 x 10 <sup>6</sup>	3 x 10 <sup>7</sup>	4 x 10 <sup>6</sup>	1 x 10 <sup>6</sup>	2 x 10 <sup>6</sup>	5 x 10 <sup>7</sup>	2 x 10 <sup>6</sup>	1.4 x 10 <sup>7</sup>	1 x 10 <sup>6</sup>
After Doc 90	6.4 x 10 <sup>7</sup>	4 x 10 <sup>6</sup>	9 x 10 <sup>7</sup>	6 x 10 <sup>6</sup>	2.6 x 10 <sup>8</sup>	4 x 10 <sup>6</sup>	3 x 10 <sup>8</sup>	9 x 10 <sup>6</sup>	2.6 x 10 <sup>8</sup>	1.6 x 10 <sup>7</sup>
After Doc 120	1.5 x 10 <sup>8</sup>	6 x 10 <sup>6</sup>	1.6 x 10 <sup>8</sup>	2 x 10 <sup>6</sup>	1.5 x 10 <sup>8</sup>	1 x 10 <sup>6</sup>	5 x 10 <sup>7</sup>	3 x 10 <sup>6</sup>	1 x 10 <sup>7</sup>	2 x 10 <sup>6</sup>
The analysis of vibrio is	done in every :	30 days of culture i	in interval i.e. ir	n Doc 1, 30, 60, 9	1, 121 and their	colony estimati	on in fu is giv	ven in Table 6		

#### Table 7: Biomass harvested, total feed consumption, FCR, ABW and feed brand used in different pond

Pond	S.D.	Biomass in Kg.	Total feed consumed	FCR	ABW Gm.	Brand of feed			
1	1 lakh	2.432	4465.7	1.51	32	Wave			
2	1 lakh	2.094	3300.4	1.57	30	Wave			
3	1 lakh	2.497	3802.8	1.52	33	Wave			
4	1 lakh	1.830	2909.7	1.59	30	Wave			
5	1 lakh	1.999	3084.5	1.55	32	Wave			
Likewise the amount of feed consumed in different pond throughout the culture and their FCR Biomass harvested. ABW as well as brand of feeds is given in Table 7									

Likewise the amount of feed consumed in different pond throughout the culture and the 100 to 283 ppm respectively in the ponds. This result is inagreement with result of Boyd (1999). The total hardness was maintained up to its optimum level for 60 days of culture but after that it exceeded quite to its optimum level which may be due to application of feed and increased in size of shrimp inside the pond but it has not affected so much to culture operation and it was maintained by the exchange of water. Addition of some commercial prepared probiotics was

reported to be effectively fruitful with these substances and that way helped in maintaining water quality parameters thereby improving growth rate, weight gain and survival rate with an attractive FCR in farmed organisms (Sission, 1989). In the present study higher survival rate 60-75% was recorded and it was achieved due to application of probiotics and required stocking density of 96,000/ha and this result was inagreement with the work of Krantz and Norris (1975) that



Figure 1: Changes in green colony count at different ponds during the culture period

survival rate was 60-80% to be expected for P.monodon under suitable rearing condition. The water guality parameters of ponds, which were applied with microbial supplement through probiotics, was good because of various roles played by microbes. The pond that treated with Ecomax, Super biotic, Environ-Ac, Prolig-HQ, Bioremid-Aqua and Ecoforce was abundant with Bacillus sps, which showed a low level of Ammonia, which was converted into nitrate through nitrite. The result presented in Fig. 1 and 2 indicated that vibrio load was found in almost all days of culture. The vibrio load was very low during 30 days of culture which may be due to use of feed and planktonic dead material inside the pond. The load was more in rest of the culture period because of different problem like bloom crash, gill chock, vibriosis, bioluminescence, one month mortality and snail inside the pond. Though the reduction of vibrio load was observed which may be due to application of probiotics in 1<sup>st</sup> 50 days, It did not influence as much as on rest of the culture and it may be due to the occurrence of vibrosis in Doc 76 in pond No. 4, occurrence of vibriosis in Doc 90 in Pond No. 5 which is inagreement with the result of Dalmin et al., (2001) that Bacillus sp. contributes 20% and vibriosis contributes around 40% of total heterotrophic bacteria in coastal ecosystems of culture pond.

Manipulation of bacterial species composition will lead to keep the pathogenic bacteria like vibriosis under control which was confirmed by various workers on different species like Altermonas sp. supress activity of V.harvayi (Abraham, 2004). Marine pseudomonas 1-2 strain (Chythanya and Karunasagar, 2002) Bacillus S11 a probiant bacterium for immunity enhancement in black tiger shrimp (Rengpipat et al., 2000). Bacillus sps. used as probiotics on digestive enzyme activity, survival and growth of Indian white shrimp Fenneropenaeus indicus (Nejad et al., 2006) Lactobacillus brevis and Lactic acid reduced the load of Vibrio alginolyticus(Villamil et al., 2003). Yeast culture supplement as disease resistant to Pacific white shrimp Litopenaeus vannamei (Burgents et al., 2004). Among these priority was given to Bacillus sps. The Bacilli inhibit the pathogenic vibriosis by competing for nutrients and damaging the lime layer of vibriosis through secreting enzyme that degraded slime. The Bacillus also secrete enzyme that could break down the organic wastes and excess nutrients in pond eco-system. Though the vibrio load was found throughout the culture period in all the ponds, in the present investigation the probiotic applied to pond showed good water quality as it was evident by optimum



Figure 2: Changes in yellow colony count at different ponds during the culture period

transparency by regular weekly interval exchange of water. Hence regular use of probiotics in culture pond will definitely help to suppress the pathogenenic of vibrosis, which will not affect the pond and the culture was successful to obtain good crops.

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