BIOAUGMENTATION IN PRAWN (PENAEUS MONODON FAB) FARMING: EFFECTS OF ACCELOBAC AG ON THE WATER QUALITY AND SURVIVAL OF VARIOUS GROWTH STAGES OF PRAWNS REARED UNDER SEMI-CONTROLLED CONDITIONS[1]

By

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ABSTRACT

Three experiments were conducted to determine the specific benefits derived from the application of **Accelobac AG** in a semi-controlled culture system with growing prawns.

In the first experiment, **Accelobac AG** was applied at 1.0, 0.5 and 0.3 g/cu.m. in the rearing medium of prawn postlarvae, PL18. Significant improvements in the water quality were obtained in the bacteria-inoculated lots over the control. The bacterial population and total solids content of the water were also monitored. Survival of prawn postlarvae after 20 days was 85.3% with the application of **Accelobac AG** compared with the control, which was 54.3%.

The second experiment demonstrated the efficiency of **Accelobac AG** inoculation in the growing media of older prawns (Stage I: 37-day old; Stage II: 50-day old; Stage III: 76-day old; Stage IV: 96-day old). Nitrite and ammonia levels were reduced significantly with bacterial inoculation (NO2 = 0.156 to 0.213 ppm; NH3 = 0.073 to 0.093 ppm) compared to the control (NO2 = 0.483 to 0.563 ppm; NH3 = 0.413 to 0.674 ppm). Regardless of age of prawns, water quality parameters were maintained within optimum limits set for the organism. Percent survival for bacteria-inoculated lots were 96.7% (Stage II) and 86.7% (Stage III) compared to the control which were set at 43.3% and 40% for stages II and III respectively.

Various concentrations of **Accelobac AG** were tested for growing 61-day old and 100-day old prawns. Application of **Accelobac AG** at only 0.5 g/cu.m. was equally effective as the higher concentrations of 1.0 and 1.5 g/cu.m. Good water quality and high prawn survival were obtained with bacterial inoculations.

1 Research commissioned by G.L. Whang of Nu-Genes Technologies, Inc.
This paper was presented to the "International Symposium on Application of Biotechnological Methods and Recent Accomplishments of Economic Value in Asia" in Bangkok, Thailand.

2 Consultants, Nu-Genes Technologies, Inc.

BIOGRAPHICAL SKETCH

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Introduction

Research and development efforts are greatly needed to sustain the growth of the prawn industry in the Philippines. In 1987, prawn production showed a growth rate of 31% in volume (15,500 MT) valued at P3.2 Billion. Employment generated in this sector is tremendous considering that at least 20,000 hectares of brackish water ponds have been converted to prawn monoculture. However, the industry suffered a major setback due to the low market price of prawns. For these reasons, support and assistance in terms of industry-oriented researches on ways and means of improving production efficiency must be conducted at the soonest time possible.

One area of research and development is the use of specially selected and adapted strains of microorganisms inoculated in the ponds to improve water quality and survival rates of prawns. It is in this context that bioaugmentation of these bacterial strains in the new and innovative product, **Accelobac AG**, were tested under semi-controlled conditions. Experimental results will provide baseline information for the eventual commercial application of **Accelobac AG** in prawn culture systems.

First Experiment: Effects of **Accelobac AG** on the quality and Survival of Prawn Postlarvae, PL 18.

Materials and Methods

Five thousand pieces of prawn postlarvae (PL 18) obtained from Calatagan, Batangas were transported to the experimental site in double plastic bags with oxygen. Young postlarvae were stocked at 5,000 pcs/bag with 30 liters water during transport. Upon arrival, the transported postlarvae were placed in 100-liter capacity tanks containing 20 liters of water with salinity of 23 ppt. The set-up was provided with moderate to vigorous aeration. They were then conditioned to their new environment with the gradual addition of 20 ppt seawater.

After two days, 50 pcs each of the post-larvae were stocked in 12 units of 100-liter capacity tanks containing 30 liters of water at 20 ppt. or an effective stocking density of 1,666.7 per square meter.

The required amount of **Accelobac AG** was applied by preparing a stock solution of 1 gram of **Accelobac AG** in one liter of water. Aliquot amounts were added in each tank, i.e. 30 ml, 15 ml, and 9 ml each for treatment I, II and III. The un-inoculated lots served as control.

	Dosage	
Treatment	**g/cu.m.**	**g/30 l.**
I	1.0	0.030
II	0.5	0.015
III	0.3	0.009
IV	0	0

Prawn postlarvae were fed with SMI-President starter feed given at 3% of body weight (BW). The feed was prepared by mixing one part of feed to 5 parts of water osterized and seived through 80 microns plankton nets. Feed ration for the day was divided into 6 equal parts and fed to the prawns at every four-hour interval.

The physico-chemical parameters of the rearing water were closely monitored. These consisted of nitrite, nitrate, ammonia, hydrogen sulfide, dissolved oxygen (D.O.), pH, total solids, and bacterial count. Samples were taken initially and every two days thereafter using standard methods.

No water change was done for the entire duration of the experiment (20 days). Initial and final length and final survival of the test animals were recorded.

Results and Discussion

Nitrite and Nitrate Levels of the Water (Figs. 1 & 2)

The nitrite levels in the growing media of young postlarvae, PL 18, were maintained at very low levels in all treatments with bacterial inoculations (0.3, 0.5 and 1.0 g/cu.m.). Values obtained on the 20th day of culture period were below 0.2 ppm NO2 compared with the un-inoculated control, which reached 0.592 ppm and 0.648 ppm on the 18th and 20th day of culture respectively. These results demonstrate the effectiveness of **Accelobac AG** in reducing the nitrite level of water even at a very low concentration of 0.3 g/cu.m. for young postlarvae. The same trend was observed in the nitrate level of the water (Fig. 1 and 2).

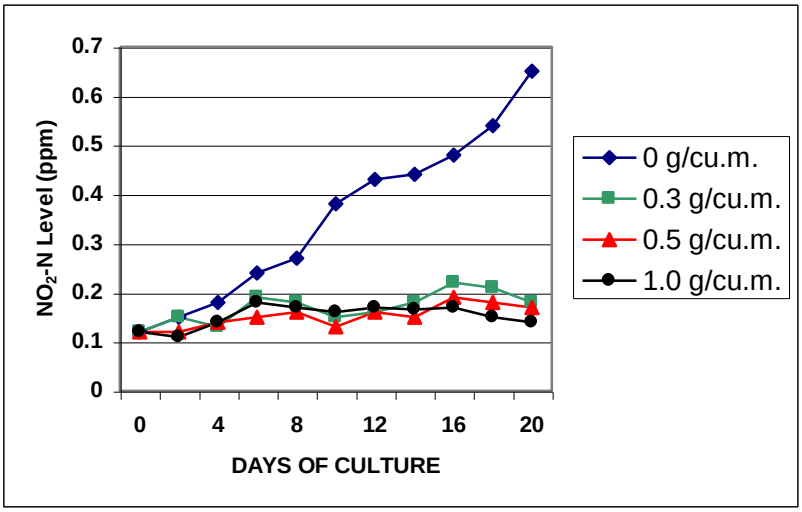


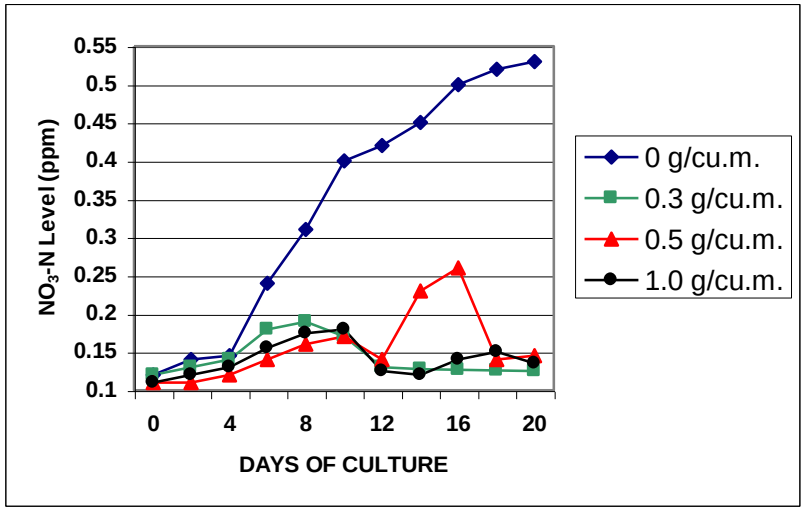
Fig. 1. Nitrite content of the water inoculated with varying concentrations of **Accelobac AG**

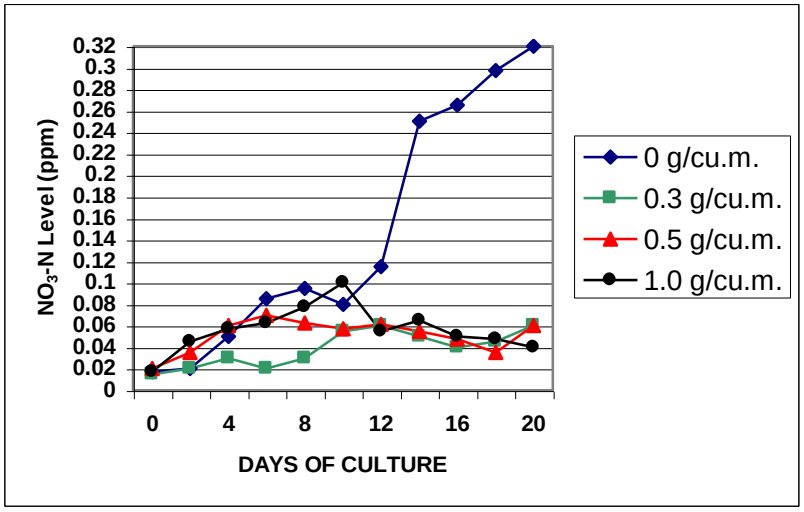
Statistical analysis showed that bacterial inoculations of various levels provided a lot of beneficial effects and are significantly different from the control without bacterial inoculations.

Fig. 2. Nitrate level of the water inoculated with varying concentrations of **Accelobac AG**

Ammonia content of the water (Fig.3)

Bacterial inoculation was also very effective in maintaining the ammonia level of the rearing water of young prawn postlarvae within optimum levels. This effect was most





pronounced starting on the 12th day of culture when the un-inoculated control lots showed drastic increase in the ammonia level. However, the conditions are unlike in older prawns where the ammonia accumulation in the control reached toxic levels of up to 3.2 ppm (see results of 2nd and 3rd experiments). With young PL18, the highest value of NH3-N in the control without bacterial inoculation is only 0.316 ppm which is still within the optimum

Fig. 3. Ammonia level of the water inoculated with varying concentrations of **Accelobac AG**

limits set for prawn. This result can be explained by the fact that young postlarvae are given less feeds compared with older prawns.

Dissolved oxygen in the water (Fig.4)

Mean dissolved oxygen levels of both control and treated lots were high during the experimental period. Values ranged from 6.28 to 7.02 ppm after 2 days of rearing. Dissolved oxygen (D.O.) readings were maintained within these high values up to 20 days without change of water and no photosynthesis. The D.O. is well within the optimum limit for prawns which is set at a value higher than 3.5 ppm. The higher values of D.O. obtained in the control compared to the treated lots may be due to the consumption of oxygen by the bacteria during the decomposition of uneaten feeds and fecal matter excreted by the growing postlarvae.

The Duncan's Multiple Range Test showed that no significant difference was observed among the treated and controlled lots.

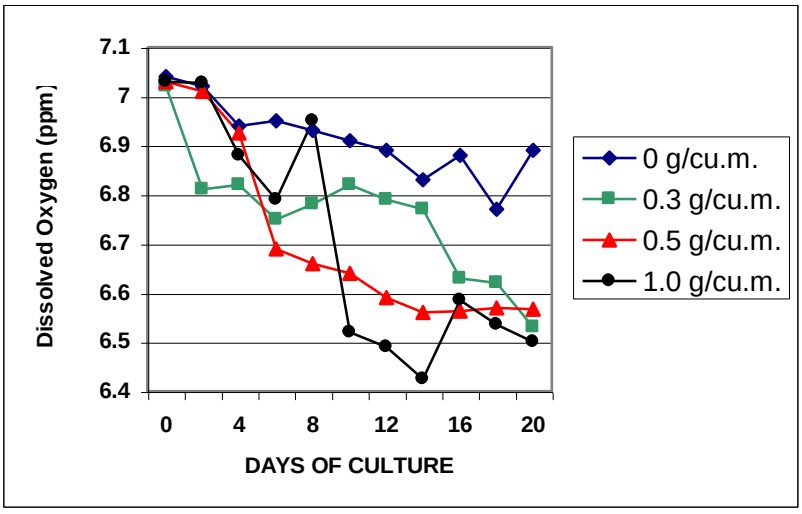


Fig. 4. Dissolved Oxygen levels of the water inoculated with varying concentrations of **Accelobac AG**

pH of the water (Fig.5)

The same trend was observed for both treated and controlled lots. Values started at pH 7.0 gradually becoming lower (pH 6.4 etc) as the experiment progressed. The range of pH for both control and treated lots up to the 16th day of culture without water change remained within the optimum limit set for prawn (pH 6.5 to pH 8.7).

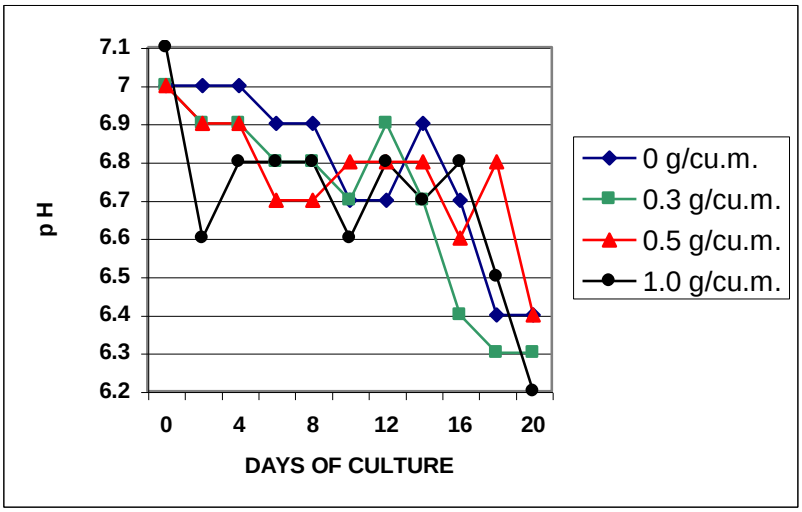


Fig. 5. pH of the water inoculated with varying concentrations of **Accelobac AG**

Survival of postlarvae (Fig.6)

After 20 days, higher percent survival was obtained in all treatments inoculated with **Accelobac AG** compared with the control. The highest bacterial concentration applied (1.0 g/cu.m.), gave a high percent survival of 85.3% compared to the uninoculated control which was 54.3% only. Duncan's Multiple Range Test showed significant difference between control and treated lots.

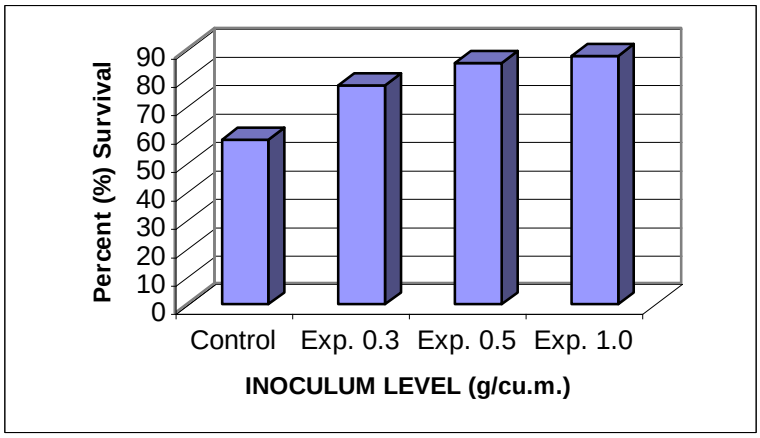


Fig. 6. Percent survival of prawn post larvae (PL18) grown in media inoculated with varying concentration of **Accelobac AG**

Bacterial population (Fig.7)

The bacterial population were significantly higher in the inoculated lots than the control. This effect was most apparent on the 14th day of culture. This may be explained by the fact that the inoculated bacteria multiplied in number because of the accumulation of organic matter in the rearing media. On the other hand, the non-specific, indigenous bacterial population in the control did not show significant increase in number.

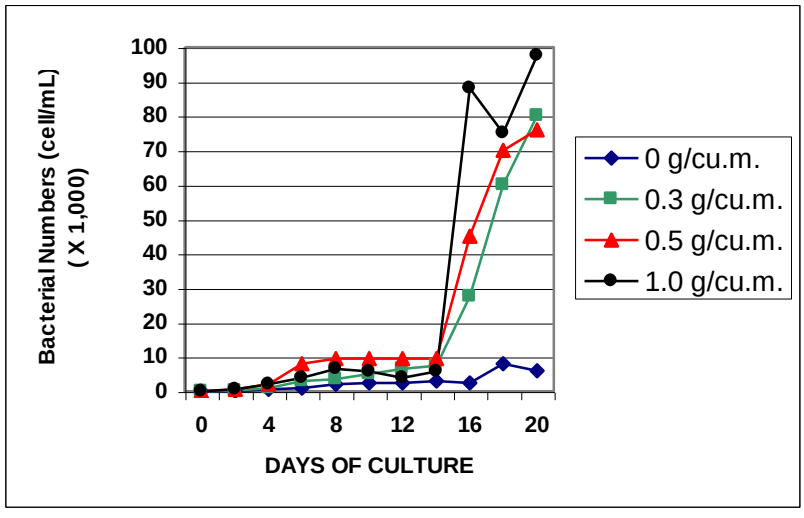


Fig. 7. Bacterial population in the water inoculated with varying concentrations of **Accelobac AG**

Total Solids (Fig.8)

The efficiency of bacterial inoculation was reflected in the total solids content of the medium. In the control lots, there was a build up of organic matter in the form of feces and uneaten feeds. With bacterial inoculation, degradation was going on efficiently in the system such that the total solid contents of the media remained low. This effect was most apparent starting on the 12th day of culture.

The total solids content of the rearing media were significantly lower in the inoculated lots compared to the control.

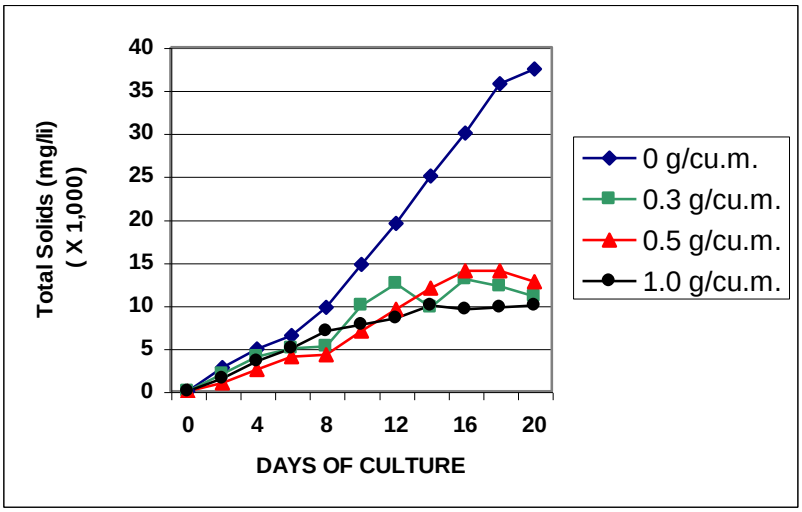


Fig. 8. Total solids content of the medium inoculated with varying concentrations of **Accelobac AG**

Second Experiment: Effect of **Accelobac AG** in four growth stages of prawn (37, 50, 76, and 96 day-old juveniles)

Materials and Methods

Transport of older prawns was done using the usual double plastic bags (50 x 70 cm.) which were filled with oxygen. Water taken from the pond where the prawn juveniles were grown was used. Bigger prawns (i.e. 76 and 96 day-old), were transported at 25 pcs./bag while the smaller ones (i.e. 37 and 50 day-old) were transported at 50 pcs./bag. To each bag were added 5 grams of **Accelobac AG** to maintain good water quality during the transport since the prawns were not properly conditioned (i.e. not starved for at least 12 hours) prior to transport. No mortality during transport and after 24 hours was observed. Transport time up to the experimental site was 3 hours and 10 min. coming from Nasugbu, Batangas and 4 hours and 15 min. coming from Pagbilao, Quezon.

Opon arriving at the experimental site, the transported prawns were placed in 100 liter capacity tanks containing 40 liters of pond water with salinities of 20 and 18 ppt for prawns coming from Nasugbu, Batangas and Pagbilao, Quezon respectively. The set-up was provided with a moderate aeration system. The experimental animals were allowed to be conditioned to its new environment for 24 hours. They were then gradually acclimated to a water salinity of 18 ppt prior to the conduct of the experiment.

Prawn representing four growth stages as indicated below were stocked at 10 pcs. each in 24 units of 100-liter capacity tanks containing 20 liters of water at 18 ppt or an effective stocking density of 42 per square meter. The different stages of growth were designated as follows:

Stages of growth		**Source**
Stage I	37 day old	Nasugbu, Batangas
Stage II	50 day old	Pagbilao, Quezon
Stage III	76 day old	Nasugbu, Batangas
Stage IV	96 day old	Pagbilao, Quezon

Accelobac AG were added at the rate of 1.19 g/tank or a dosage of 5 g/cu.m. Control lots were those without bacterial inoculation.

The experimental set-up was provided with moderate aeration and pieces of nets to serve as sanctuary for the prawn during molting.

The experiment was conducted in a randomized Complete Block Design with three replicates per treatment.

Prawns were fed with SMI-President Feeds as in the first experimental given at the following rates:

Stage of Growth	**% Body Weight**	**Type of Feed**
Stage I	20% BW	Starter
Stage II	20% BW	Starter
Stage III	12% BW	Grower
Stage IV	5% BW	Grower

The Designated feeding rates were divided into six equal parts and fed to the prawns at every four hour interval.

To determine the activity of **Accelobac AG**, bacterial counts of the rearing water were taken initially and every two (2) days thereafter through the Most Probable Number (MPN) Method with Nutrient Agar as medium.

The physico-chemical parameters of the water like pH, D.O., nitrite, ammonia, and total solids (fecal matter and unconsumed feeds) were likewise taken initially and every two days thereafter using the standard methods (APHA, 1976).

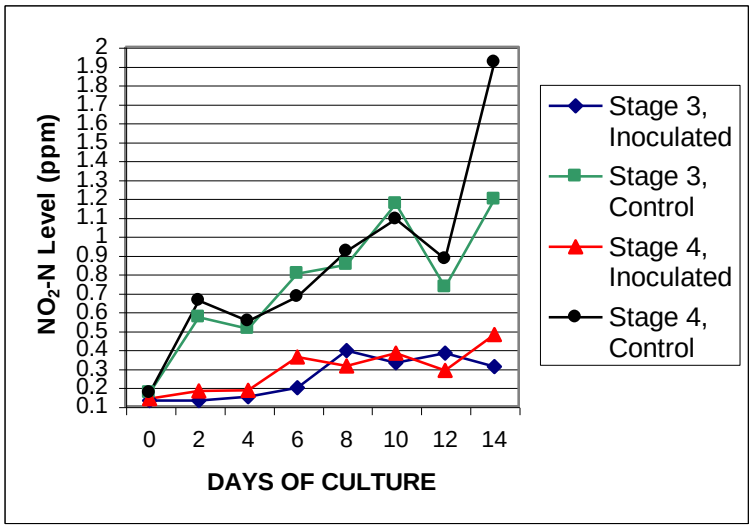
No water change was done for the entire duration of the experiment which lasted for 14 days.

Results and Discussion

Nitrite level of the water (Fig.9-A&B)

Inoculation of the **NS Series in the rearing** medium with prawns at various stages of growth improved water quality. This is evident as early as during the first two (2) days where the nitrite levels were considerably lower (0.156 – 0.213 ppm) than in the uninoculated control (0.483 – 0.563 ppm). The nitrite level was maintained at a significantly lower value in the treated lots than in the control for the duration of the experiment which lasted for two (2) weeks. (It should be noted that there was no water change during the experimental period). On the other hand, the nitrite level in the uninoculated increased tremendously. In general, the inoculated bacteria was effective in reducing the nitrite level in the rearing medium with prawns by as much as **481%**. This became apparent on the 14^{th} day when the nitrite level reached 2.133 ppm in the control while the medium with the bacteria had only 0.493 ppm. These data were observed in the medium where 96 day-old prawn (Stage IV) were stocked.

The effectiveness of the **Accelobac AG** in the inoculated medium was further demonstrated by the fact the irregardless of age of prawn, the low nitrite level was maintained. This means that despite the high amount of feeds given to older prawns, the inoculated bacteria were able to



degrade the uneaten feeds and metabolites such as there were no accumulation of the toxic nitrite in the medium. As the prawns got older, the higher rate of feeding resulted in greater amounts of uneaten feeds and

Fig. 9-A

higher rate of nitrite level in the water. This was true for both the inoculated and the uninoculated rearing media. However, it should be emphasized that the nitrite level in the prawn growing medium inoculated with **Accelobac AG** remained below the toxic level, (For prawn, the toxic level is greater than 5 ppm). On the other hand, the uninoculated control reached a value of 2.13 ppm.

Fig. 9-B

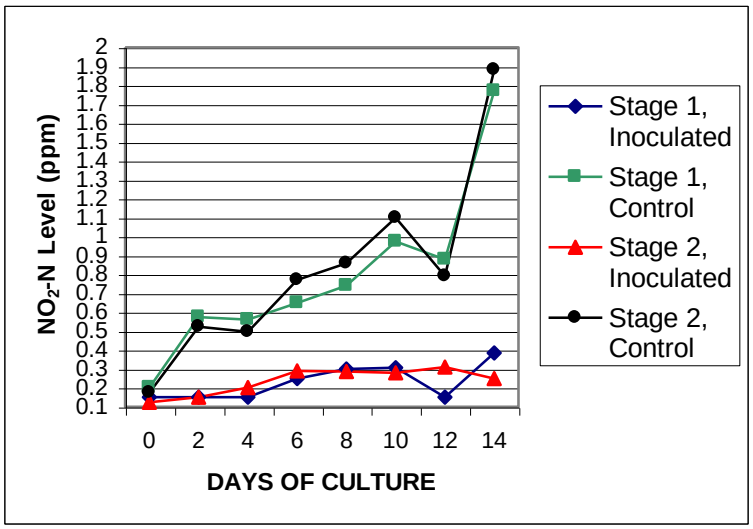
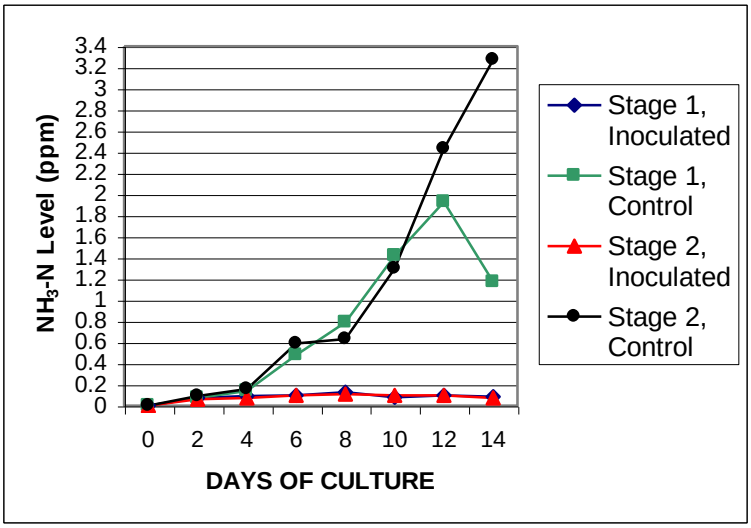


Fig. 9. A&B Nitrite level of the rearing medium of four growth stages of prawns inoculated with **Accelobac AG**

Statistical analysis showed the significant differences between the treated and control lots were obtained from the 2nd to the 14th day of culture for all growth stages of prawns.

Ammonia Content of the water (Fig.10 A & B)

The same trend was observed on the effectiveness of **Accelobac AG** in maintaining good water quality for prawn farming. The ammonia levels up to the 6th day of culture were relatively low and safe for the growing of prawns for both control and bacterial-inoculated lots. The values ranged from 0.073 to 0.093 ppm for the inoculated growing media with prawns at stages I to IV while that in the control without bacterial inoculation had higher values ranging from 0.413 to 0.674 ppm. Starting on the 8th day and up to 2 weeks, ammonia accumulated at very high levels for all control lots. During the same periods, the bacteria inoculated media maintained an ammonia level below 1 ppm which is still considered tolerable for prawns. Except for the treated lots I which had 37 day old prawns, the ammonia levels remained well below the stress values (0.23-0.54 ppm). In contrast, the uninoculated control lots had ammonia accumulating in the media at extremely high levels of up to 3.2 ppm.



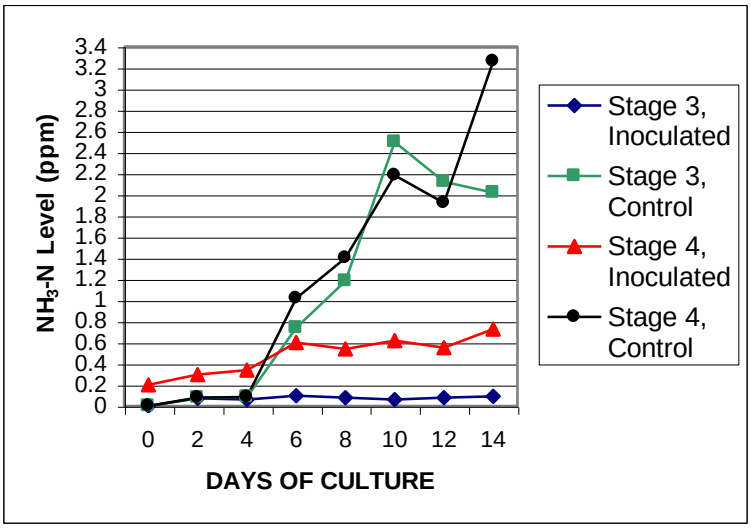
Earlier reports of Chiu (1987) stated that prolonged exposure of prawns to 0.1 ppm NH3-N can have adverse effects.

Fig.10-A

The ammonia content of the water is dependent on pH and temperature. At higher pH values, ammonia toxicity problems become more serious. During the experiment, there was not much increase in the pH level for both control and treated lots (Figs. 11 A & B). However, in general, in the media inoculated with bacteria, the pH values were lower than in the control. This again is attributed to no water change and the subsequent degradation of organic matter into CO2, H20 and cell tissue by the inoculated bacteria in **Accelobac AG**.

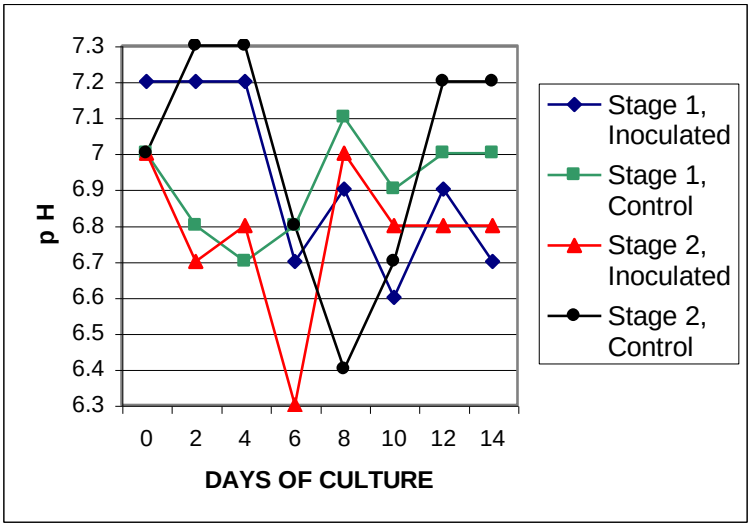
The Duncan's Multiple Range Test showed significant differences between control and treated lots from the 4th to the 14th day of culture.

Fig.10-B



Figs. 10- A & B. Ammonia content of the rearing medium of four growth stages of prawns inoculated with **Accelobac AG**

Fig. 11-A



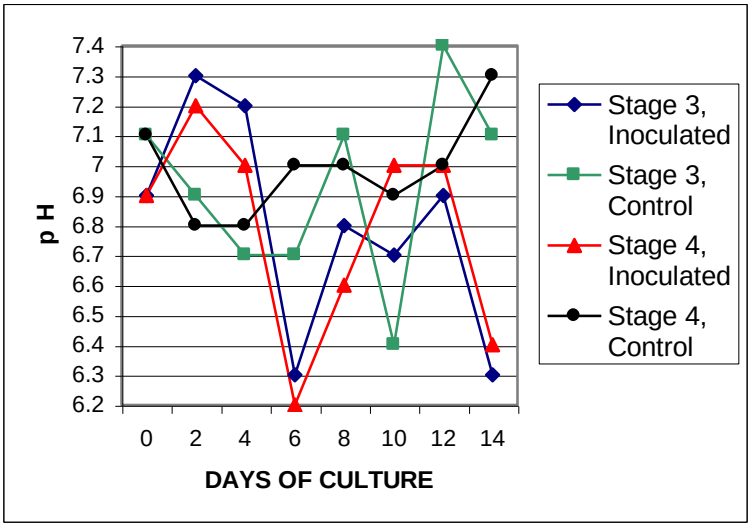


Fig. 11-B

Fig. 11 A & B. pH levels of the rearing medium of four growth stages with inoculation of **Accelobac AG**

Effect on the dissolved oxygen (Figs. 12 A & B)

Both control and treated lots showed high D.O. values favorable for the growth of prawns. It should be noted however that the experiment was conducted under a semi-controlled system. This being the case, there

was no photosynthetic activity going on which would increase the D.O. content in the water. Moreover, the inoculated bacteria which break down organic matter in the system, consume oxygen in the process. This would explain the lower D.O. values in the **Accelobac AG** inoculated treatments.

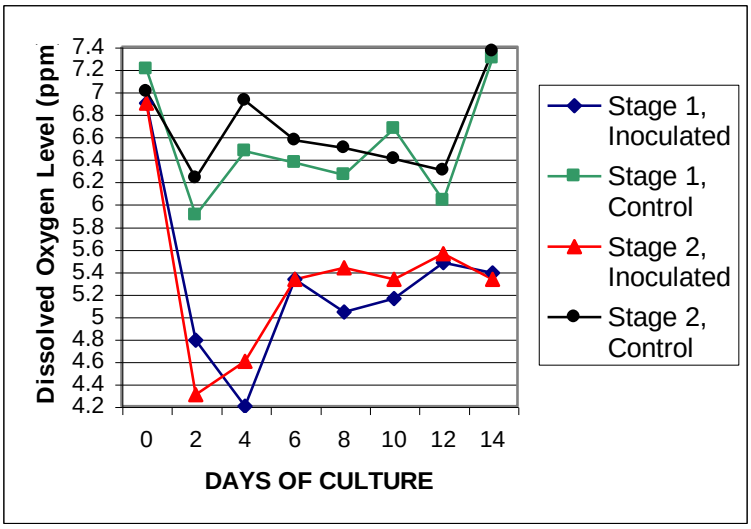


Fig. 12-A

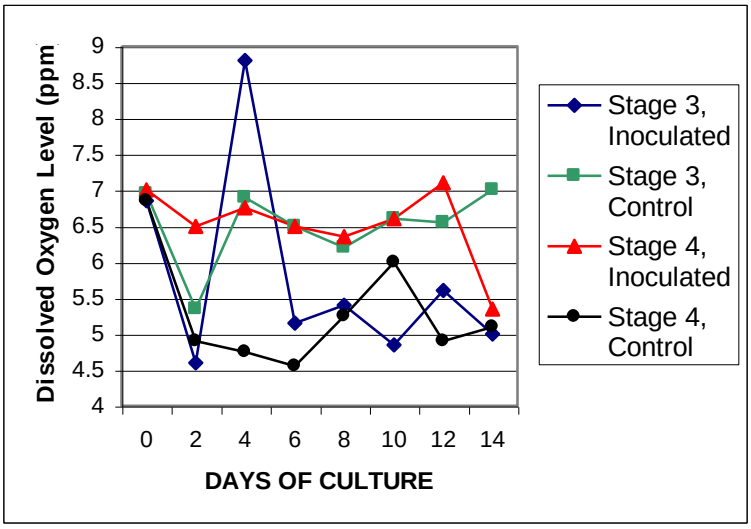


Fig. 12-B

Figs. 12-A & B. Dissolved oxygen of the rearing medium of four growth stages of prawns inoculated with **Accelobac AG**

Effect on Prawn Survival (Fig. 13)

Accelobac AG treatment was most effective in increasing the percent survival of prawns. This was observed in all growth stages although the effect was most pronounced for 50 day-old and 76 day-old (Stages II and III) prawns. High survival values of 96.7% and 86.67% were obtained for stages II and III respectively. For the control lots without bacterial inoculation, survival was lowest at stage III (40%) and stage III (43.3%).

Statistical analysis showed significant differences between inoculated and control lots for all the growth stages of prawns.

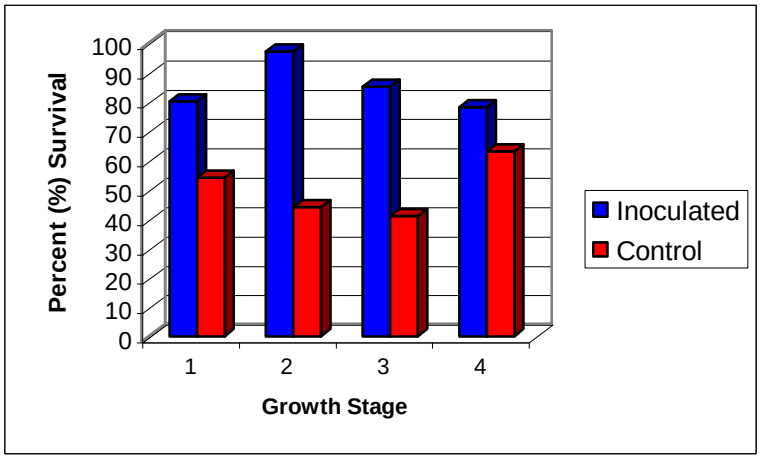


Fig. 13. Survival of four growth stages of prawns with **Accelobac AG** treatment in the rearing medium

Total solids: (Figs. 14 A & B)

The ability of the inoculated bacteria to decompose uneaten feeds and fecal matter in the growing media is shown by the total solids content in the water. The amount of feed increases

with age of prawns and this is reflected in the increasing total solids content in the growing medium with prawns at stages I to IV in the control lots.

At termination of the experiment, the growing media in the control lots were very turbid. Analysis of the total solids present showed that as much as 1.87 g/li or 1,870 ppm accumulated in the control. On the other hand, in all the **Accelobac AG** treated media, the water seemed almost transparent. This condition demonstrates the fact that despite the increasing amount of feeds given to the prawns, the inoculated bacteria were able to decompose the feeds efficiently. The total solids content of the water with bacterial inoculation did not go beyond 0.28 g/li or 280 ppm after two (2) weeks.

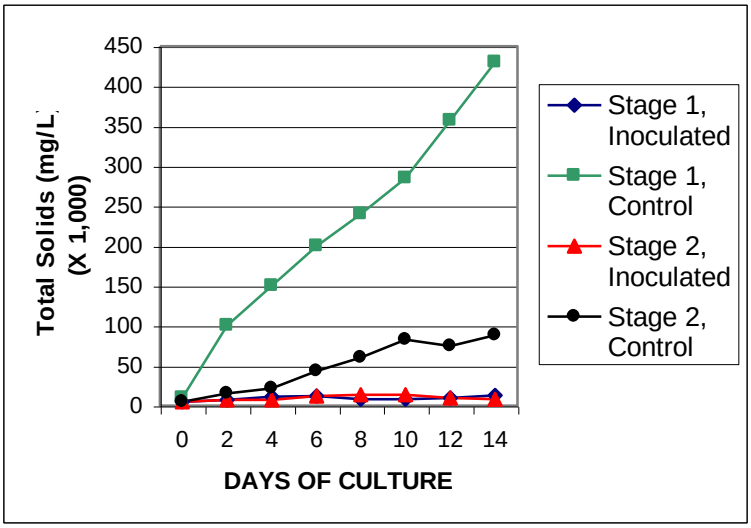


Fig. 14-A

Based on the amount of **Accelobac AG** inoculated in the media which is 5 g/cu.m., the decomposition rate was computed at 117.21 mg/li/day for Stage IV on 96 day-old prawns.

Significant differences between control and treated lots occurred on the 4th day for all the growth stages of prawns based on Duncan's Multiple Range Test.

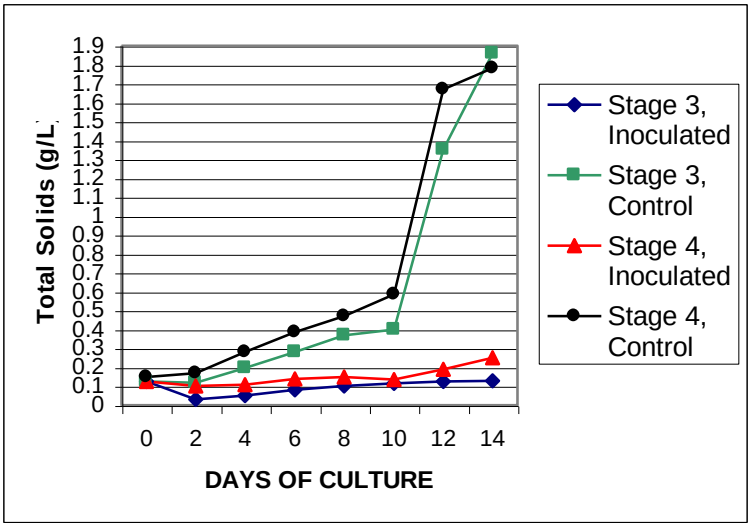


Fig. 14-B

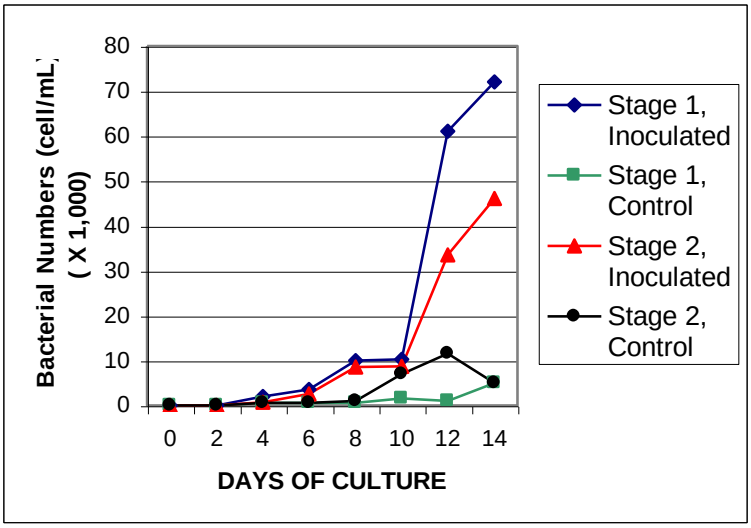
Fig. 14-A & B Total Solids in the rearing medium of four growth stages of prawns treated with **Accelobac AG**

Bacterial Count (Figs. 15 A & B)

The bacterial population in the rearing media increased with time. Higher bacterial counts were obtained in the treated lots which is expected because of bacterial inoculation. The bacterial population was lower in the control lots. Moreover, the type of bacteria between the control and treated lots are assumed to be different.

The Duncan's Multiple Range Test showed that differences in the bacterial count between treated and control lots in all growth stages of prawns are highly significant ($p<0.05$).

Fig. 15-A



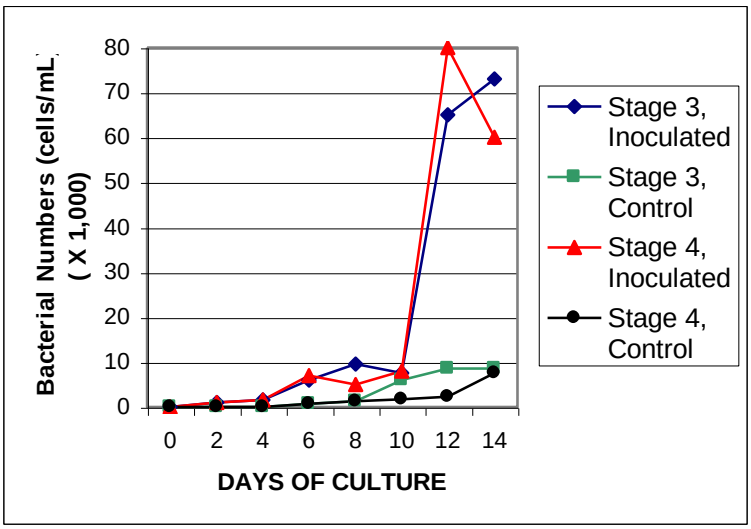


Fig. 15-B

Fig. 15 A & B Bacterial population in the water treated with **Accelobac AG** for four growth stages of prawns (P. monodon)

Third Experiment: Determination of the optimum dosage application of **Accelobac AG**

Materials and Methods

Eighty pieces each of prawns belonging to two (2) stages of growth A = Stage I (61 day-old), B = Stage II (100 day-old), obtained from Nasugbu, Batangas were transported to the experimental site. Upon arrival, the transported prawns were placed in 100-liter capacity tanks containing 40 liters of pond water with a salinity of 26 ppt.

The set up was provided with moderate aeration. The experimental animals were conditioned to its new environment for 24 hours. They were then acclimated to a water salinity of 18 ppt prior to the conduct of the experiment.

Eight pieces each of sugpo representing the two stages of growth indicated above were stocked in 18 units of 100-liter capacity tanks containing 30 liters of water at 18 ppt or an effective stocking density of 33.6/sq.m.

Three dosage levels of **Accelobac AG** were inoculated to each tank as follows:

Treatment	Dosage g/cu.m.	g/30 li
I	0.5	0.015
II	1.0	0.030
III	1.5	0.045

The required amount of **Accelobac AG** was applied by preparing a stock solution of 1 gram of culture in 1 liter of water. Aliquot amounts were added in each tank i.e. 15 ml, 30 ml, and 45 ml each for treatments I, II and III respectively.

Prawns were fed with commercially prepared diet (SMI-President Feeds) used in earlier experiments given at 12% BW. Feed ration for the day was divided into six equal parts and fed to the prawns at every 4 hours interval.

The physico-chemical parameters of the rearing water were closely monitored as in the first two experiments. These consisted of pH, D.O., nitrite, ammonia and total solids (unconsumed feeds and fecal matter). Samples were taken initially and every two days thereafter using standard methods.

No water change was done for the entire duration of the experiment which lasted for 14 days.

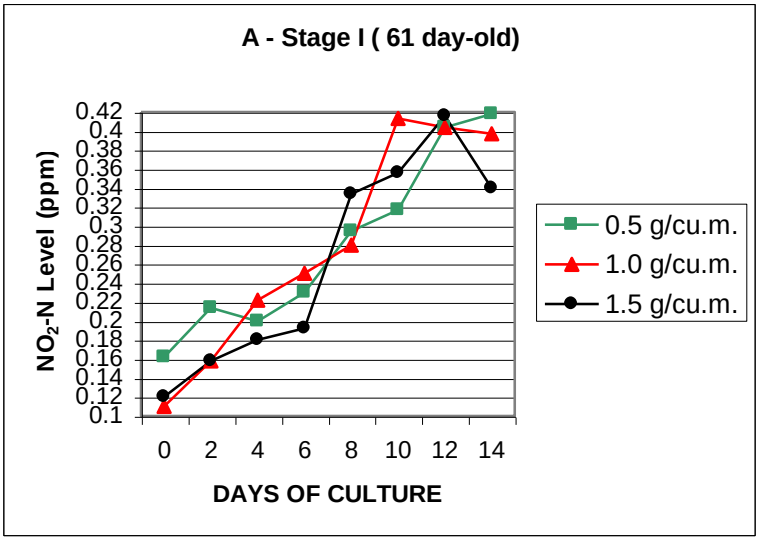
Initial and final length and weight of the test animals were taken. The survival of the test animals in each of the treatments were taken at the termination of the experiment.

Results and Discussion

Effect on the Nitrite level (Figs. 16 A & B)

Varying dosages of **Accelobac AG** (0.5, 1.0, 1.5 g/cu.m.) inoculated into the growing medium of prawns did not produce differences in the nitrite level. In all the treatments the values were maintained well within optimum limits which were set at 0.1 ppm – 0.5 ppm for nitrite.

Among the three inoculum levels applied, no marked differences in the nitrite levels were observed during the conduct of the experiment which lasted up to 14 days. This means that 0.5 g/cu.m. which is the lowest rate of application of **Accelobac AG** is sufficient to reduce the nitrite content of the water to a safe level. This may be explained by the fact that bacteria multiply and grow as long as there is available substrate in the environment.



In both the 61 day-old prawns and 100 day-old prawns, the three dosages of **Accelobac AG** were equally effective in controlling the nitrite levels within optimum limits. This demonstrates the efficiency of the bacteria inoculated in the growing medium which act on higher amounts of uneaten feeds and fecal matter as the prawns grew older.

Fig. 16-A

Statistical analysis showed no significant differences among the three dosages of **Accelobac AG for two growth stages of prawns.**

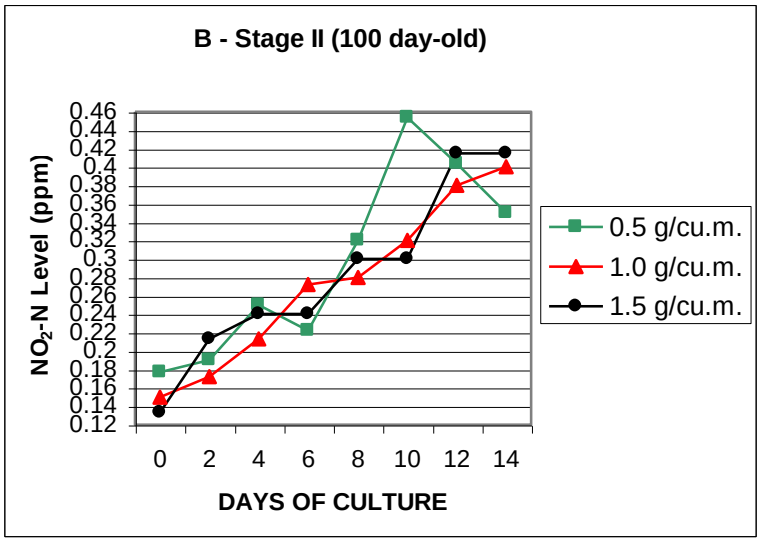


Fig. 16-B

Figs. 16 – A & B. Nitrite level of the water inoculated with varying dosages of **Accelobac AG** for two growth stages of prawns

Effect on the Ammonia level (Figs. 17 A & B)

The ammonia level in all the treatments remained very low and within tolerable limits set for prawn. The same trend was observed, that is, there were no differences in the ammonia level of the water inoculated with varying dosages of bacteria. The rearing water in both Stage I (61 day-old) and Stage II (100 day-old) prawns showed comparable values throughout the experimental period.

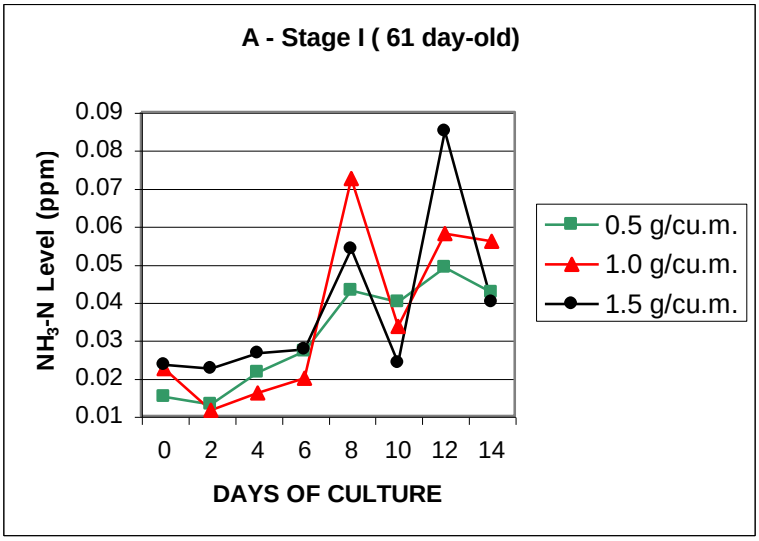


Fig. 17-A

Fig. 17-B

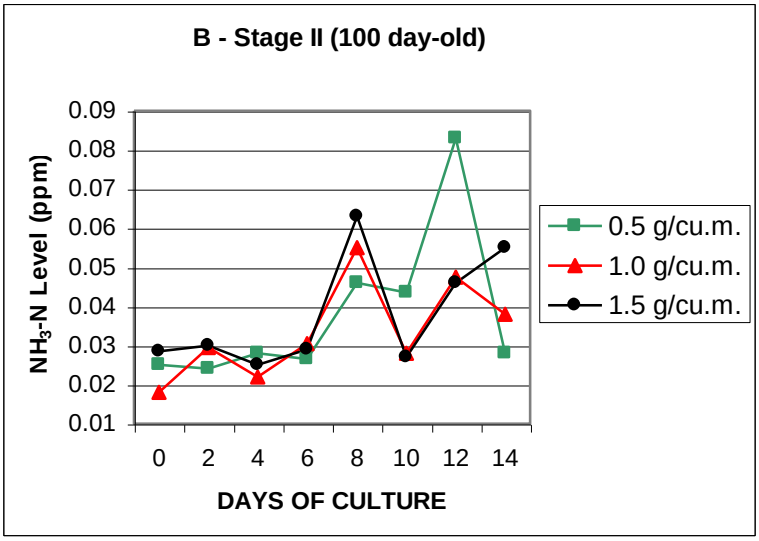


Fig. 17 – A & B Ammonia level of the water inoculated with varying dosages of **Accelobac AG** for two growth stages of prawns.

Effect on Dissolved Oxygen (Figs. 18 A & B)

The varying levels of bacterial inoculation produced comparable values of dissolved oxygen. The D.O. levels in the various treatments were all greater than 3.5 ppm, the optimum limit set for prawn farming.

Based on the above results, bacterial inoculation may be reduced to 0.5 g/cu.m. without adversely affecting the D.O. level in the water. Moreover, bioaugmentation of **Accelobac AG** at the start of the culture period prevented the accumulation of organic matter in the medium. This being the case, there was no drastic reduction in the D.O. level since decomposition occured gradually with time.

Statistical analysis showed no significant differences among the three dosages of **Accelobac AG** inoculated in the rearing medium for both 61 day-old and 100 day-old prawns.

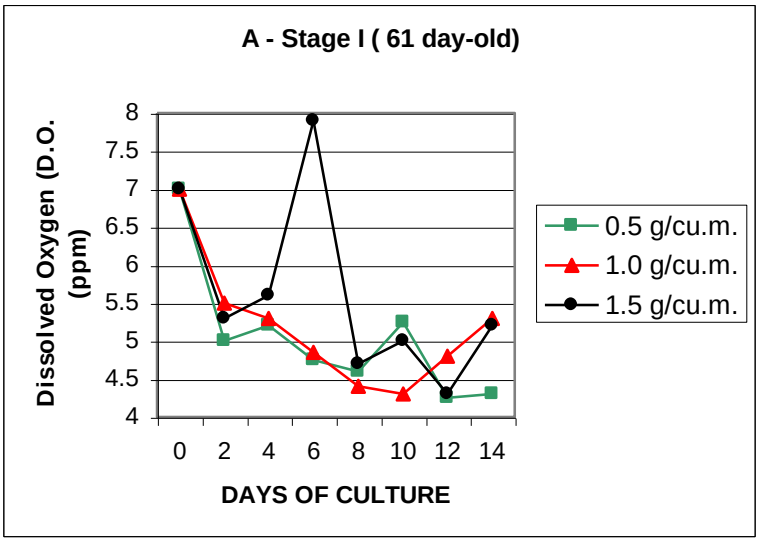


Fig. 18-A

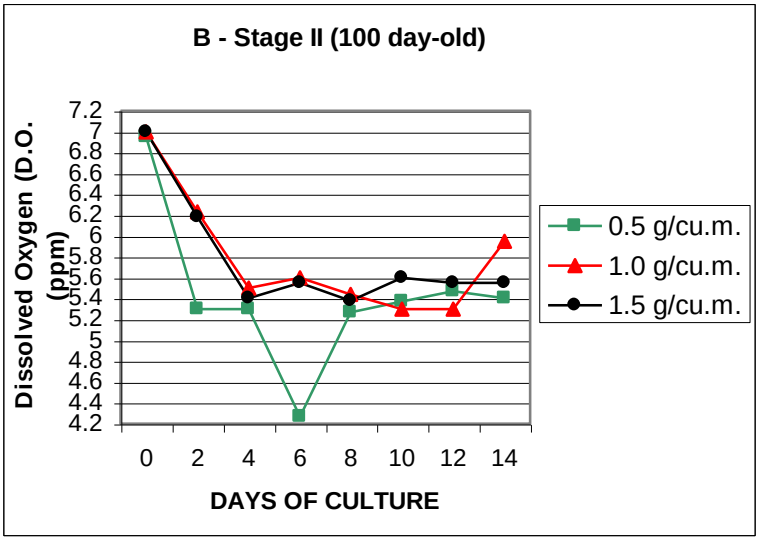
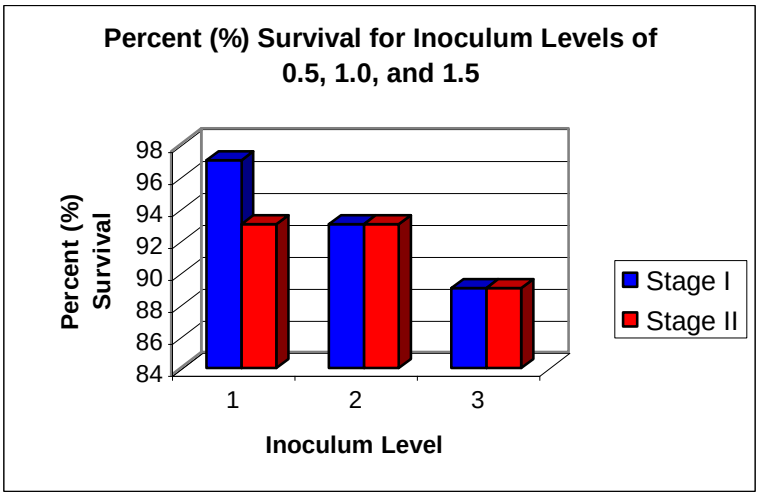


Fig. 18-B

Figs. 18 – A & B. Dissolved Oxygen level of the water inoculated with varying dosages of **Accelobac AG** for two growth stages of prawns

Effect on Prawn Growth and Survival (Fig. 19)

High survival of prawn (2 growth stages) in a closed semi-controlled culture system without water change was observed in all the treatments. Survival values ranged from 87.5% to 95.8%. This may be attributed to the maintenance of good water quality throughout the duration of the experiment.



Duncan's Multiple Range Test did not show significant differences among the three varied dosage applications.

Fig. 19 Percent Survival of prawns growing in media inoculated with three dosages of **Accelobac AG**

Total Solids (Figs. 20 A & B)

The total solids content of the rearing water inoculated with three levels of bacteria were comparable for specific stages of growth of

prawns. Note that the total solids content of the 100-day old prawn is 2 to 3 times more than in the younger (61 day-old) prawn. This demonstrates further the efficiency of **Accelobac AG** given at a low dosage of 0.5g/cu.m.

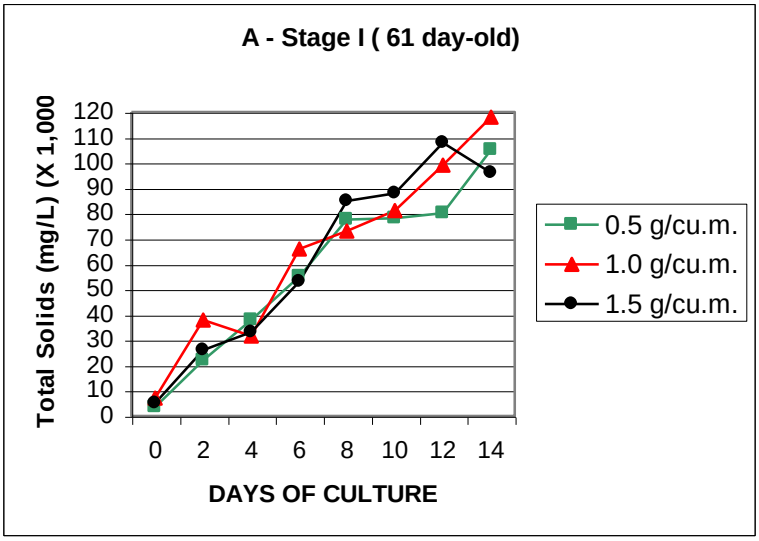


Fig. 20-A

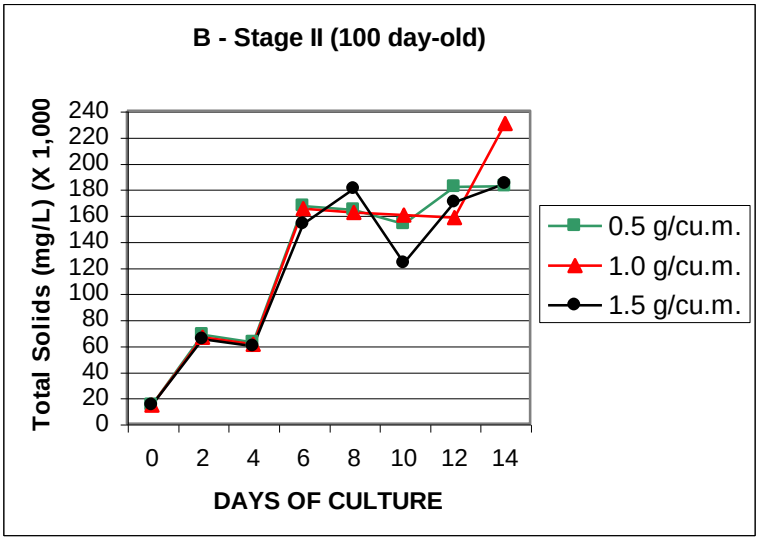


Fig. 20-B

Figs. 20 – A & B Total solids content of the water inoculated with three dosages of **Accelobac AG** for two growth stages of prawns

Bacterial Population (Figs. 21 A & B)

The bacterial population did not differ among the different levels of inoculation. Since bacterial growth is dependent on the amount of substrate in the medium, the amount of substrate (waste materials) available can only support bacterial growth to this level.

Between the two growth stages of the prawns, similar bacterial counts were also observed. Apparently, the minimum amount of bacteria applied (0.5 g/cu.m.) has the capacity to efficiently decompose even the higher substrate level produced by Stage II (100 day-old) prawns.

Statistical analysis did not show significant differences among the various treatment dosages.

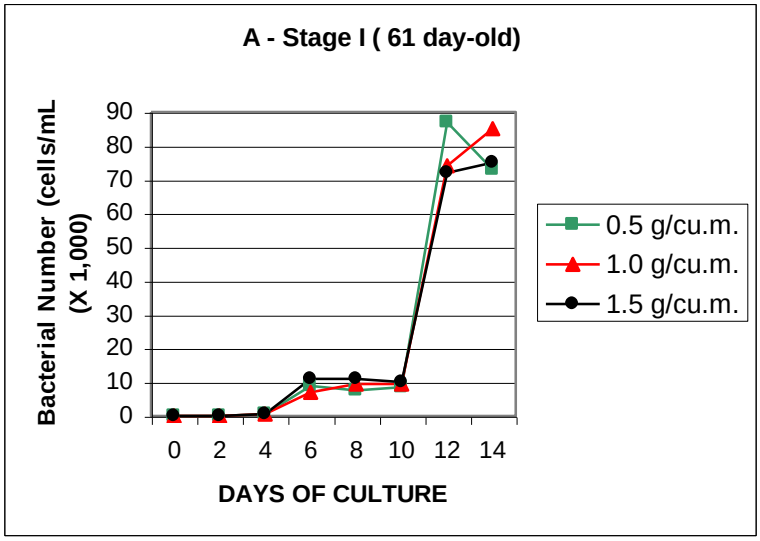


Fig. 21-A

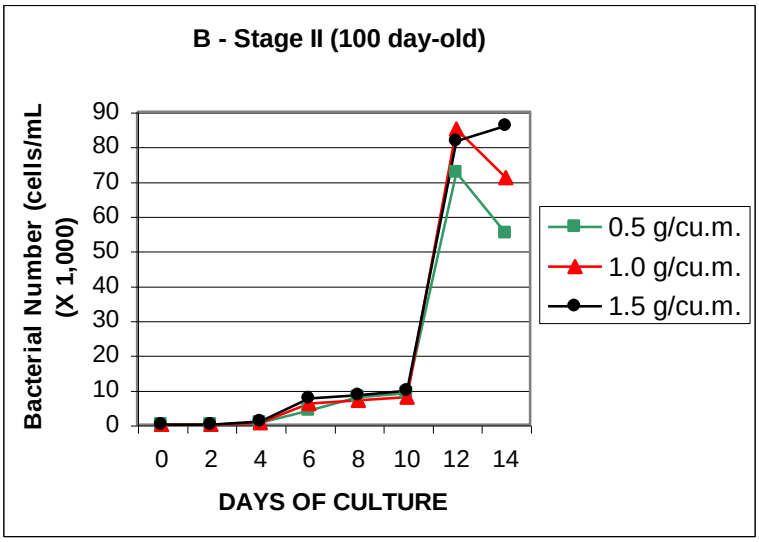


Fig. 21-B

Figs. 21 – A & B. Bacterial population of the water medium inoculated with three dosages of **Accelobac AG** for two growth stages of prawns