J. Appl. Environ. Biol. Sci., **1(12)592-596, 2011**

© 2011, TextRoad Publication

ISSN: 2090-4215
Journal of Applied Environmental and Biological Sciences
www.textroad.com

The Effect of Consortia Bacteria on Accumulation Rate of Organic Matter in Tiger Shrimp, *Penaeus Monodon* Culture

Anik Martina Hariati[1]*, Sudirdjo[1], Aulanni'am[2], Soemarno[3] and Marsoedi[3]

[1]Aquaculture Department, Faculty of Fisheries and Marine Sciences, University of Brawijaya, Malang, 65145, Indonesia
[2] Laboratory of Biochemistry, Department of Chemistry, Faculty of Sciences, University of Brawijaya University, Malang, 65145, Indonesia
[3] Post Graduate Program, Department of Environmental & Natural Resources Management, University of Brawijaya, Malang, 65145, Indonesia

ABSTRACT

The present study was conducted to study consortia bacteria inoculate in the juvenile of *Penaeus monodon* culture. Two hundred juvenile were divided in five experimental groups each with four replicates. The experiment was conducted for 21 days. Experimental media were homogenized in all the aspects except for variation in the consortia bacteria concentration. The four treatments group of bacteria ranging from 0.5 to 2.0 ml/L and the control group (without bacteria). The consortia bacteria were found to have decreasing effects on accumulation of total organic matter, TAN and nitrite concentration, while nitrate was increasing accordingly. Growth and survival rate of shrimp which is consortia bacteria applied groups was no significantly different ($P>0.05$) from the control group.

Keywords: Consortia-bacteria, water quality, shrimp growth

INTRODUCTION

The year of 1985 can be said as the starting point where shrimp culture system in East Java changed, from traditional into intensive. The changed was marked by increase in PL's stocking density (from 2-5 to 40-60 individual/ m^2), adding artificial feed, antibiotic and oxygen input using paddle wheel. This turned the land rent for Tambak system into much higher value that made positive impact to regional and national economy [1] [2]. However, 7-10 years after the intensification, around the year of 1992, there was a clear symptom on collapse of shrimp-based aquaculture in the region. Hariati et al. [3] found that within two years 1992 to 1993, total shrimp yield in East Java decreased by 50%. The decrease in shrimp production was mainly due to the collapse of intensive culture, although traditional system faced the same pattern after the intensive.

From the findings, the cultured shrimp was infected by MBV, Mododon Baculo Virus. There were confirmed that both shrimps, under traditional and intensive culture system, have been infected by MBV. Following the collapse, there more viral and vibriosis identified from culture media and shrimp tissues [4].

In the second research of Hariati et al. [2], it was found that water visibility decreased as time of culture increases. This was strongly related to sharp increase in total organic matter accumulated in the top soil of the pond, toward the end of culture period. Other water quality parameters were not significantly different. Shrimp farmers suspected and assumed organic waste from the excess of pellet feed as the main source of the problems. All efforts were then focused to minimize the organic matter during the culture period while maintaining the stocking density at still high level (40 – 60 individual/m^2)

Increased accumulation of organic matter must be caused by the excess of artificial feed (pellet) provided into the culture system. Of the total amount of feeds applied to the pond aquaculture, only 6.7% by dry weight is converted into shrimp biomass, the rest is leached or otherwise not consumed, egested as feces, eliminated as metabolites [5]. This would increase the population of bacteria, from which some bacteria may induce stress and reduce health status of cultured shrimp. The cultured shrimp is getting higher risk to be infected by pathogenic bacteria and viral diseases [6][7]. Shrimp farmers in the region applied consortia to enhance (from normal) microbial degradation of organic matter and expected there will be minimum organic matter accumulated toward the end of culture period. Several new processes and operational strategies like combined nitrification–DE nitrification have emerged in order to remove high concentrations of nitrogen compound in wastewaters [8][9][10]. An experiment was designed to investigate what parameters affected by the addition of consortia bacteria into the culture system.

MATERIALS AND METHODS

Consortia Bacteria preparation

A local commercial consortia bacterium was obtained from a distributor. Consortia were isolated and identified. This product is claimed by the manufacturer to be a blend of fermentation productions of *Pseudomonas* sp., *Nitrobacter* sp., *Nitrosomonas* sp., *Bacillus* sp. and *Aerobacter* sp. There were non-pathogenic bacteria were found in the consortia and *Bacillus* sp was the dominant bacteria in the product. Following the distribution instruction, the consortia was

***Correspondence Author:** Hariati, Aquaculture Department, Faculty of Fisheries and Marine Sciences, University of Brawijaya, Malang, 65145, Indonesia. Email: a_hariati@ub.ac.id

increased in number by addition of 25 ml to 1 litre of seawater containing mixed bran, soy meal, fishmeal and continuously aerated for 3 days incubated at 30°C. The mixed was freshly prepared and adjusted to a final concentration of approximately 10^{10} CFU/ ml.

Experimental set-up

The research was applied in 20 aquaria (each of 40 l). Dry soil (autoclave) was deployed on the bottom of each aquarium of 2 cm thick. Each aquarium was stocked with 10 shrimp juvenile with individual weight of 1.0 – 1.5 g. This assumed to be equal to 60 juveniles per square meter stocking density in the field, a typical density of intensive culture system in East Java. Four treatments of different consortia bacteria concentration were applied randomly into 20 aquaria (apart from the control, without treatment).

The experiment lasted for 21 days and no changing in water aquaria. The shrimp fed with standard locally-commercial pellet at 10% BW/day. The commercial diet composition was 42% protein, 5% fat, 25% carbohydrate, 12% moisture, 13% ash and 3% crude fiber. Each individual aquarium was covered with a plastic lid to prevent shrimp escape and to reduce water evaporation. Water in each aquarium was gently aerated via a single air-stone to keep DO above 5 mg/l. Water temperature, DO, pH and salinity were measured daily. Mortality was observed daily too. Water organic matter, nitrite, nitrate and TAN were measured weekly. Substrate organic matter was measured at initial and end phase of the experiment. Daily growth rate was adapted from weekly measurement of individual body weight. TAN, nitrites and nitrates were measured according to Standard Methods [11].

Statistical analysed

Percentage accumulation rate is defined as total value of parameter at the end of the experiment subtracted by the initial value; the result is divided by the initial value. This measure is aimed to describe on how control group accumulate organic matter compare with the ability of different treatment to remove the organic matter. Data were shown as means ± standard deviation (SD). All parameters were determined in four replicates. Completely Randomized Design (CRD) was performed by five treatments level and four replications each. Significant difference in the change of water quality parameters of TAN, nitrite, nitrate, organic matter among the treatments over time was tested by using a one-way analysis of variance (ANOVA). Duncan's New Multiple Range Test was used to isolate any significant differences detected in the ANOVA (SPSS ver. 16). All data were considered significant at $P<0.05$.

RESULTS AND DISCUSSION

During the culture period it can maintain the daily water quality. Daily temperature was kept constant at 26.5°C. Through aeration, it also can maintain daily oxygen around 5.4 mg/l. pH of average 7.9 and salinity around 28.0 g/l.

In the normal condition (control group), TAN will increase up to 70% in 21 days. Whereas, consortia bacteria application can remove TAN up to 37% that was found in the highest concentration of consortia (Table 1). Ammonia yielded of oxidized by bacteria nitrification especially become nitrite. This indicated that in the water which is used as a research media have contained the *Nitrosomonas* sp. The consortia also lead to removal of nitrite and transfer it into nitrate. When the bacteria were added into the water, they could decompose the excreta of shrimps, uneaten feed, and other organic materials to CO2, nitrate and phosphate [12]. Nitrification processes are influenced by a variety of environmental factors and toxic substrates, salt, organic carbon and need significant amount of oxygen [13][14]. However, aeration can always fill the oxygen deficiency in the system, even during nitrification – hence the oxygen all over the treatments remains constant. As the result of nitrification by bacteria, higher nitrate concentration was always found in all consortia treatment. As we always provide enough oxygen to the system (aeration) nitrification process is surely need.

Table 1. Statistical analyses of water quality parameters: Ammonia, Nitrite, Nitrate and Total bacteria (concentration and percentage).

Treatment	NH4-N*		NO2-N*		NO3-N*	
ml/L	mg/L	%	mg/L	%	mg/L	%
0.0	0.05 ^{a}	70 ^{a}	0.20 ^{a}	64 ^{a}	0.54 ^{a}	-75 ^{a}
0.5	0.04 ^{a}	-10 ^{b}	0.12 ^{b}	-55 ^{b}	0.84 ^{b}	34 ^{b}
1.0	0.04 ^{a}	-17 ^{b}	0.12 ^{b}	-43 ^{b}	1.44 ^{c}	123 ^{c}
1.5	0.04 ^{a}	-37 ^{b}	0.10 ^{c}	-65 ^{b}	1.63 ^{c}	163 ^{c}
2.0	0.03 ^{a}	-37 ^{b}	1.09 ^{c}	-66 ^{b}	2.38 ^{d}	286 ^{d}

*Values followed by the same notation there was no significant different among treatment

The graphs showed TAN and nitrite accumulation in control group and consequently increase in nitrate concentration when consortia added into the culture system (Figure 1). This means that consortia bacteria could reduce TAN and nitrite and oxidized it into nitrate, N compound that not toxic for shrimp.

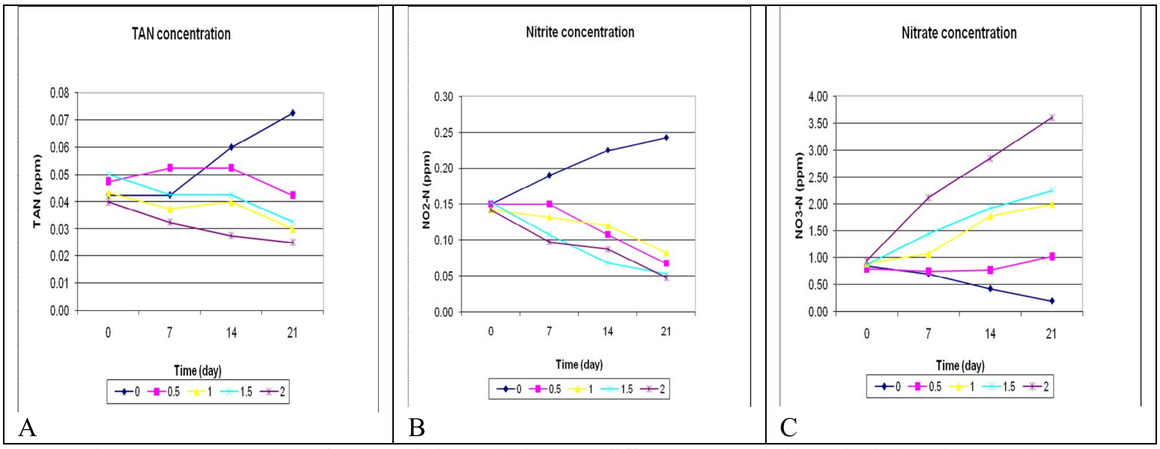

Figure 1 Concentration of TAN, Nitrite and Nitrate at different consortia bacteria during the experiment

Consortia bacteria seem to have an ability to remove organic matter in the water and bottom substrate. The data analyses showed significantly different ($P < 0.05$) among treatments as printed at Table 2. At the first week of culture period organic matter in the water almost the same, except for treatment of 1.5 ml/L. After day 7, the organic matter for control group was increasing, showing increased accumulation. Meanwhile, all treatments showed tendencies to remove organic matter around 67% compared to control group (Figure 2). The same pattern also occurred for organic matter removal from the substrate depressed until 35%. There is no clear explanation on higher removal rate for treatment of 1.5 ml/l of consortia. The gram-positive *Bacillus* spp. are generally more efficient in converting organic matter back to CO_2 than are gram-negative bacteria, which would convert a greater percentage of organic carbon to bacterial biomass or slime [15]. Another reason due to the activity of the bacteria which is inoculated [16].

The C organic would be used as energy source for bacteria growth, *Pseudomonas* sp using 60-80 C-organic source to survival. During the experiment period the C/N ratio decreasing from 49 to 22.72 equal to 54%. This value is quite high compared to Hariati et al. [4]. The variation of C/N ratio in the soil substrate was not only caused by decomposition process, but also quality sources of organic matter forms.

Table 2 Statistical analyses of Organic matter in the water, soil substrate and C/N ratio (concentration and percentage).

Treatment	OM-water		OM-soil		C/N ratio	
ml/L	mg/L	%	mg/L	%	mg/L	%
0.0	55.32 a	69 a	8.55 a	25 a	19.83 a	-60 a
0.5	48.30 b	25 bc	7.87 ab	7 ab	17.93 a	-63 a
1.0	47.56 b	14 cd	7.55 b	-1 b	27.64 b	-44 b
1.5	39.52 c	-3 d	6.94 b	-17 b	20.03 a	-59 a
2.0	49.54 ab	43 b	7.64 ab	1 ab	28.19 b	-42 b

Values followed by the same notation was no significant different

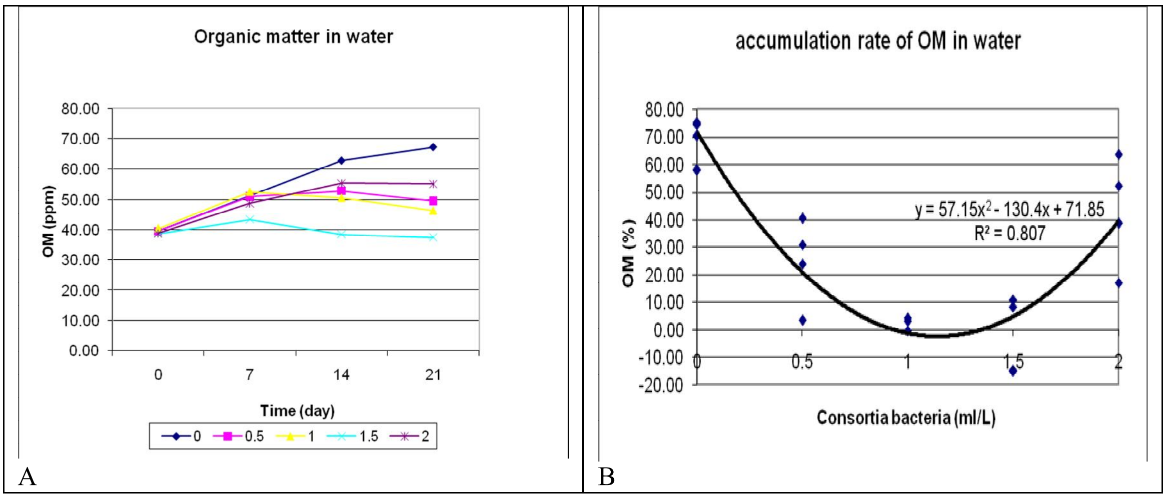

Figure 2 Concentration (A) and the relationship between consortia bacteria and water organic matter accumulation rate (B)

Mortality was not found in any of the experimental groups, indicating no adverse effect of bacteria on survival (Table 3). In this study found no significant different to survival rate (SR), specific growth rate (SGR) and FCR between different treatment. The accumulation of organic matter between control group and different treatments may not reach a threshold to affect shrimp mortality. There was no significant different among different treatment on growth measured as SGR. This means, higher organic matter accumulation in the control group did not lead to stress of shrimp, and shrimp can grow normal. The nutritional requirements to convert in growth can change according to variations in water quality factors, such as salinity, temperature, and dissolved oxygen [17]. In this research water quality parameter in all the treatment was maintained at a normal condition for shrimp growth. The experiment period might be too short (21 days) to explain the impact of organic matter on shrimp growth (culture period in the field was around 120 days)

Table 3 Statistical analyses of Growth (SGR), Survival rate (SR) and Feed conversion ratio (FCR).

Treatment	SGR %bw/d	SR %	FCR (g/g)
0.0	4.80 [a]	100	1.27 [a]
0.5	4.88 [a]	100	1.23 [a]
1.0	4.96 [a]	100	1.20 [a]
1.5	5.03 [a]	100	1.17 [a]
2.0	4.96 [a]	100	1.20 [a]

To prevent organic pollution of the aquaculture media, the mount of organic matter loaded on the aquaculture media must balance the amount of organic matter decomposed there. However, the amount of organic matter discharged from fish farms seems to exceed by far the potential for decomposition of natural ecosystems.

CONCLUSION

Consortia, that claimed to contain 5 different bacteria, seems to have significant effect on TAN and nitrite removal & oxidise it into nitrate, a non-toxic compound of N. Consortia also showed ability to degrade organic matter caused by excess of feed

High organic matter, TAN and nitrite in the control group did not affect survival and growth. Accumulation of OM did not reach a threshold value that causes stress to shrimp leading lower growth or mortality. The experiment might be too short (21 days compared to 120 days in the field) to see the effect of OM accumulation.

REFERENCES

1. Hariati, A.M., D.G.R. Wiadnya, A. Prajitno, M. Sukkel, J.H. Boon, and M.C.J. Verdegem, 1995. Recent development of shrimp, *Penaeus monodon* (Fabricus) and *Penaeus merguensis* (de Man), culture in East Java. *Aquaculture research* 26: 819-829 pp
2. Hariati, A.M., D.G.R. Wiadnya, M.W.T. Tanck, J.H. Boon and M.C.J. Verdegem, 1996a. *Penaeus monodon* (Fabricius) production related to water quality in East Java, Indonesia. *Aquaculture Research*, 27: 255-260.
3. Hariati, A.M., D.G.R. Wiadnya, M. Sukkel, J.H. Boon and M.C.J. Verdegem, 1996b. Pond production of *Penaeus monodon* in relation to stocking density, survival rate and mean weight at harvest in East Java, Indonesia. *Aquaculture Research*, 27: 277-282
4. Hariati, A.M., D.G.R. Wiadnya, R.K. Rini, J.H. Boon and M.C.J. Verdegem. (1998). *Penaeus monodon* (Fabricius) and *Penaeus merguiensis* (de Man) biculture in East Java, Indonesia. *Aquaculture Research*, 29: 1-8.
5. Primavera, J.H., 1993. A Critical review of shrimp pond culture in the Philippines. *Reviews in Fisheries Sciences* 1: 151-201.
6. Hopkins, S.J., P.A.Sandifer and C.L. Browdy, 1994. Sludge management in intensive pond culture of shrimp: Effect of management regime on water quality, sludge characteristics, nitrogen extinction and shrimp production. *Aquaculture Engineering*, 13: 11-30
7. Jetten, M.S.M., S.J. Horn and M.C.M van Loosdrecht, 1997. Towards a more sustainable municipal wastewater treatment system. *Water Sci. Technol*. 35 (9): 171–180.
8. Fux, C., M. Boehler, P. Huber, I. Brunner, and H. Siegrist, 2002. Biological treatment of ammonium-rich wastewater by partial nitritation and subsequent anaerobic ammonium oxidation (anammox) in a pilot plant. *J. Biotechnol*. 9: 295–306
9. Aslan, S., L. Miller, M. Dahab, 2009. Ammonium oxidation via nitrite accumulation under limited oxygen concentration in sequencing batch reactors. *Bioresource Technol*. 100, 659–664.
10. APHA, 1995. *Standard Methods for the Examination of Water and Wastewater. 19th ed.* American Public Health Association, Washington, DC.

11. Li zhoujia. Zhang Qing. and Yang Huaquan. 1997 The affect of the probiotics to the shrimp ponds. *Aquaculture of China (in Chinese)*. 5:30-31
12. Boyd, C.E., 1989. Water Quality in Management and Aviation in Shrimp Farming. *Fisheries and Allied quacultures Departemental Series No. 2 Alabama Agricultural Experiment Station.* Auburn University. Alabama.
13. Rene, E.R., S.J. Kim and H.S. Park, 2008. Effect of COD/N ration and salinity on the performance of sequencing batch reactors. *Bioresource Technol.* 99, 839–846.
14. Verschuere, L., G. Rombaut, P. Sorgeloos, and W. Verstraete, 2000. Probiotic Bacteria as Biological Control Agents in Aquaculture. *Microbiol Mol Biol* Rev. 64(4): 655–671
15. Boyd, C.E., 1989. Water Quality in Management and Aviation in Shrimp Farming. *Fisheries and Allied quacultures Departemental Series No. 2 Alabama Agricultural Experiment Station.* Auburn University. Alabama
16. Boyd, C.E., 1989. Water Quality in Management and Aviation in Shrimp Farming. *Fisheries and Allied quacultures Departemental Series No. 2 Alabama Agricultural Experiment Station.* Auburn University. Alabama