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by Cek Turnitin

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Environmentally friendly shrimp culture technology: the effectiveness of seaweed – *Gracilaria verrucosa* as filtration organism in maintaining physico-chemical culture water habitat

By

Moch. Nurhudah¹⁾, Jatna Supriatna²⁾, Patria Mufti²⁾, Michael Rimmer³⁾

¹⁾ Sekolah Tinggi Perikanan, Jakarta,

²⁾ University of Indonesia – UI, Department of Biology

³⁾ Senior Research Fellow – ACIAR Expert

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ABSTRACT

The aims of this research were to characterize the effectiveness of individual filtration organisms mainly sea-weed (*Gracilaria verrucosa*) and blood cockle (*Anadara granosa*) in the black tiger shrimp (*Penaeus monodon* Fab.) polyculture system to stabilize culture water habitat. The research was performed at the Development Centre of Marine, Brackish water and Shrimp Culture (DCMBSC) Karawang, West Java, in plastic-coated wooden tanks (0.9 x 0.6 x 1 m) with no water exchange for 53 days. The treatments were related to difference levels of weight and culture methods of *G. verrucosa* and stocking density of *A. granosa*. Culture methods included bottom method and off bottom method with the seaweed depth of about 40 cm and 0 cm from the water surface. For the bottom method, seaweed was sown at about 240 g and 640 g and for off bottom method with thallus bound of about 30 g and 80 g and hung 40 cm under water surface with interval of 25 cm. Stocking density of *A. granosa* consisted of 50, 100, and 150 inds.m⁻². Polyculture system shrimp and seaweed of 30 and 80 g each attachment hung 40 cm from water surface had ability to stabilize culture media and showed significantly different in shrimp growth from the other treatments except from shrimp polyculture with 240 g seaweed cultivated by bottom method. All the systems had suitable colony of micro-organism based on magnification of *Vibrio* of about 10⁵ and total bacteria 10⁶. Seaweed, *G. verrucosa* had ability to absorb phosphorus concentration. Polyculture system shrimp (*P. monodon*) with blood cockle (*A. granosa*) revealed that the lowest concentration of total organic matter in culture water was found in the treatment 50 individuals of blood cockle per square meter. Similar results were found for ammonia and nitrate, but not for nitrate. In term of reducing total organic matter and ammonia concentration in culture water, polyculture of tiger shrimp with *A. granosa* at 100 individuals.m⁻² gave the best performance. Moreover, the highest growth rate of *A. granosa* was obtained by polyculture of shrimp (*P. monodon*) with *A. granosa* at a stocking density of 50 inds.m⁻² followed by 100 and 150 inds.m⁻². After harvesting, shrimp and blood cockle tissue were found to have the highest concentration of carbon followed by nitrogen and phosphorus. *A. granosa* tissue showed the highest gain of phosphorus.

Key Words : Culture water quality; cultivation method; growth rate; stocking density.

INTRODUCTION

Brackish-water shrimp ponds and all other aquaculture environments are man-made ecosystems and represent subunits within their surrounding natural ecosystems. The major components of pond ecosystems are the cultured organisms and the aquatic environment, including all of its biological, chemical and physical characteristics and the nutrient (feed and fertiliser) inputs (Boyd, 1999). Shrimp farming can be separated into extensive, semi intensive and intensive culture systems (Midlen & Redding 2000; Direktorat Jenderal Perikanan Budidaya 2003; Pillay 2004). Extensive shrimp farming is the oldest technology and is still used by the majority of shrimp farmers in Indonesia. However, in the late 80's and early 90's, due to market forces, many black tiger shrimp farmers switched their culture practices to semi-intensive and intensive systems. Contrary to the extensive system, intensive black tiger shrimp aquaculture practices rely on high protein feed pellets to produce high rates of growth and to increase production. Intensive farming is becoming more dominant, increasing the potential impact of shrimp farming not just to the surrounding environment but also to pond system itself.

The main cause of environmental degradation in intensive culture systems is the level of input required to support high production outputs. During the culture period, a large proportion of the high protein commercial pellets are not assimilated by the shrimps (Primavera 1994) and settles down to the pond bottom as pollutant material. Approximately 10% of the feed is dissolved and 15–50% may remain uneaten (FAO 1991). The remaining 75% is ingested, but 50% is excreted as metabolic waste, producing large amounts of gaseous, dissolved and particulate waste (Lin *et al.* 1993). Subsequently, the pond effluent contains elevated concentrations of dissolved nutrients (primarily ammonia), plankton and other suspended solids (Ziemann *et al.* 1992). The dissolved nutrients and organic material in shrimp ponds stimulate rapid growth of bacteria, phytoplankton, and zooplankton (Lin *et al.* 1993). These accumulated materials may enhance eutrophication, hypereutrophication, and organic enrichment (FAO 1991; Pillay 2004) that can generate unsuitable pond water quality for black tiger shrimp, leading to disease outbreaks or even mass mortality. In addition, during the water

exchange and harvesting time where the water is completely drained out, untreated wastes or accumulated organic matter are usually discharged directly into the surrounding environment. Effluent water from shrimp ponds typically contains elevated concentrations of dissolved nutrients and suspended particulates compared to influent water (Jones *et al.* 2001) mainly in the form of inorganic nitrogen and phosphorus (Pillay 2004). Therefore, in order to maintain a suitable culture medium during the rearing period for the cultured organism and alleviate negative impacts to the surrounding environment, fish farmers need to implement a sustainable and environmentally friendly culture technology, such as polyculture.

Based on the definition of polyculture as discussed in the previous chapter, more than one cultured organism can be used within a culture system, particularly where the other organisms are used to play ecological roles as filtration organisms (Lazur & Britt 1997) to support the main cultured organisms. Several previous studies noted that macro algae or seaweed – *G. verrucosa* (Troell *et al.* 1999b; Troell *et al.* 2003) and blood cockle – *A. granosa* have ability to reduce high concentration of dissolved nutrients and suspended particulates (Jones *et al.* 2001) produced by aquaculture practices (FAO 2000; Muller-Feuga 2000; Troell *et al.* 2003).

Most of the studies previously undertaken on seaweed and bivalve as filtration organisms were not carried out using a polyculture system but in separated culture units separating the main cultured organism from the filtration organism(s), so these represent co-cultivated or integrated aquaculture system rather than polyculture. Generally, filtration organisms were cultivated in several types of ponds, such as sedimentation pond (Lazur and Britt 1997), biological treatment pond, reservoir pond (Baliao & Tookwinas 2002) or drainage canal (Gunarto 2003; Shimoda *et al.* 2005; Shimoda *et al.* 2006). Related to the type of water exchange, these studies were performed using flowthrough system (Jones *et al.* 1999; Jones *et al.* 2001; Baliao & Tookwinas 2002) or closed recirculation system (Gunarto 2003; Shimoda *et al.* 2005; Shimoda *et al.* 2006) or a combination of both (Baliao & Tookwinas 2002; Matos *et al.* 2006). Although the results of these studies showed good filtration ability in reducing concentration of

aquaculture waste, there is a need to undertake advanced research on the filtration ability of "single-filtration organism" in polyculture systems. Information on the filtration ability in more complex ecosystems would help develop more effective polyculture approaches for shrimp farmers. Therefore, this study was intended to characterize the roles of seaweed (*G. verrucosa*) and blood cockle (*A. granosa*) as single filtration organism in polyculture system with black tiger shrimp as the main cultured organism.

9 MATERIALS AND METHODS

The experiments were conducted at the Development Centre of Marine and Brackish water Culture (DCMBC) Karawang, West Java, in plastic-coated wooden tanks (0.9 x 0.6 x 1 m) with no water exchange throughout the 53-day experimental period (from September 10 to November 3, 2008). Samples of water quality parameters were analyzed at DCMBC Karawang, Fisheries University Jakarta (Sekolah Tinggi Perikanan Jakarta), and Tambak Pandu Karawang. The objectives of this research were to determine the ecological roles and the ability of sea-weed (*G. verrucosa*) and blood cockle (*A. granosa*) as single filtration organism to stabilize environment in black tiger shrimp (*P. monodon*) polyculture system.

Stocking density of shrimp was 4 individuals.m⁻². Shrimp was adapted in the concrete tanks with the size 2 x 2.5 x 1m for 3 days and examined for WSSV (White spot syndrome virus) using polymerase chain reaction (PCR) technique before stocking. In term of seaweed, the treatments were related to differences of cultivation methods and the weight of seaweed. Seaweed cultivation methods included "bottom method" and "off bottom method" with two culture depth treatments i.e. 40 cm and 0 cm from the water surface. For the bottom method, seaweed treatments were sown and for the off bottom method, seaweed was tied with about 30 g and 80 g mass at each attachment and hung at intervals of 25 cm. Furthermore, the treatment of blood cockle consisted of 3 different levels of stocking density. In the experimental design, polyculture systems of shrimp with combinations of weight and cultured methods of seaweed and different levels of blood cockle stocking density produced several treatments as presented in Table 1-1 and Table 1-2, respectively. Control of the experiment (abbreviated by SB0)

was black tiger shrimp (*P. monodon*) monoculture system (without filtration organism). The placement for each treatment to the available tanks was completely randomized and all treatments had 3 replications.

Table 1-1

List of seaweed treatment

Treatment	Description	Total Weight (g)
SS30	seaweed was evenly sown	240
SS80	seaweed was evenly sown	640
SL430	Seaweed was tied with about 30 g at each attachment and hung with depth of 40 cm at intervals of 25 cm	240
SL480	Seaweed was tied with about 80 g at each attachment and hung with depth of 40 cm at intervals of 25 cm	640
SL030	Seaweed was tied with about 30 g at each attachment and hung on the water surface at intervals of 25 cm	240
SL080	Seaweed was tied with about 80 g at each attachment and hung on the water surface at intervals of 25 cm	640

There were no water exchange except the addition of water to replace loss due to seepage and evaporation. Experimental tanks were lined with 10 cm sand collected from the shore and then filled to 80 cm depth with sea water ± 28 ppt and no aeration during experimental period. Shrimp were fed with commercial feed 3 times a day at 07:00, 14:00 and 20:00 h with a feeding rate of 6% of total body weight.

Measurement of water quality parameters included dissolved oxygen, temperature, salinity, pH, ammonia, nitrite, nitrate, H_2S , phosphate, total dissolved solid (TDS), total suspended solid (TSS), and total organic matter (TOM). The first four parameters were measured every 3 days at dawn – 5:00 h and noon – 11:00 h and the other parameters were measured every two weeks. The growth parameters measured were weight of shrimp and blood cockle as well

as weight of seaweed; these were measured before stocking and at harvest. The measurement and taking water sample were performed at about 10 cm above bottom of experimental unit. Dissolved oxygen, temperature, salinity and pH were measured by multi-water quality parameters checker. The measurement equipment of salinity was Atago refractosalinometer. The rest of the parameters were observed by using Spectrophotometer Optima – SP300. The measurements all parameters followed Standard Methods of APHA (1979); Alerts & Santika (1987); Effendi (2003).

The results were analysed using one-way ANOVA and considered significant at an alpha level of 0.05 (Supranto 2004).

RESULTS

Shrimp-seaweed polyculture system

Physico-chemical aspects

Some of the main water quality parameters observed included dissolved oxygen (DO), pH, salinity, temperature, ammonia (NH₃), nitrite (NO₂), nitrate (NO₃), and total organic matter (TOM). Results showed that concentration of dissolved oxygen were relatively lower and more flat trend in the early morning than in the afternoon (**Figure 1-1**). One week after stocking, the highest DO concentration in the morning and at noon were observed in treatment SL430 of about 3.36 mg.l⁻¹ and 4.86 mg.l⁻¹ (**Appendix 1-1**), respectively. In general, all treatments showed similar pattern of stable tendency of DO concentration. However, just after day 30 of dawn observation and day 10 of noon observation, treatment SL430 and 480 revealed higher DO concentration than the other treatments (**Figure 1-1**).

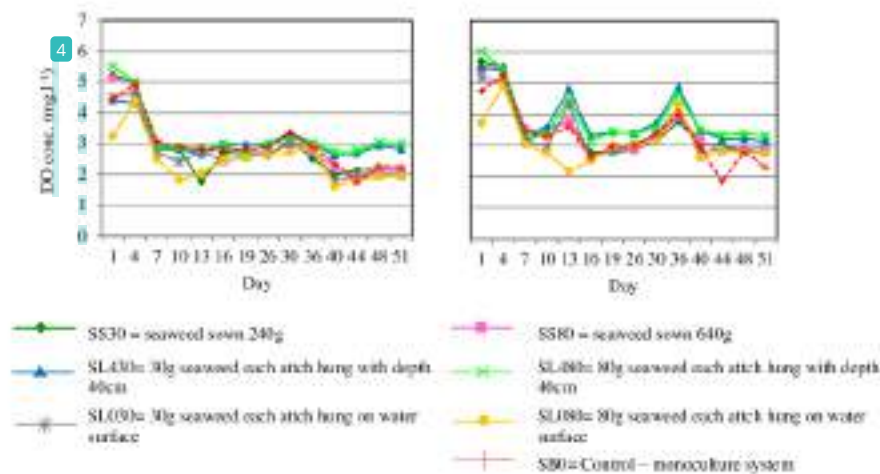


Figure 1-1

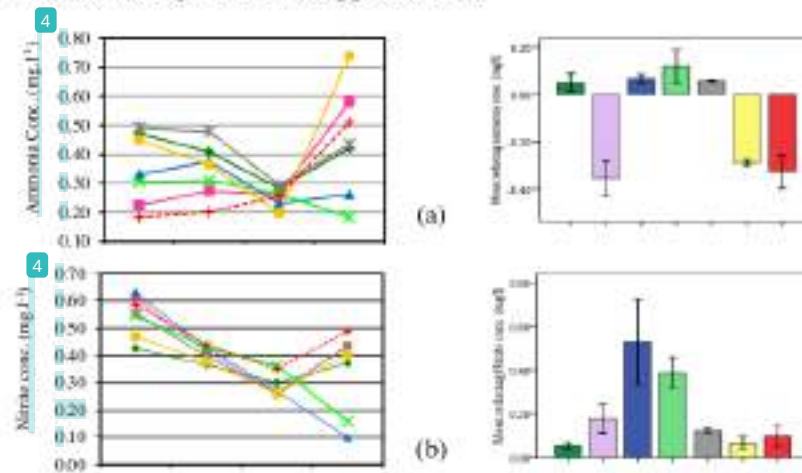
Dissolved oxygen concentration measured every 3 days in dawn (left) and noon (right) in polyculture black tiger shrimp with seaweed

All treatments showed a similar pattern of tendency of temperature, pH, and salinity. Temperature ranges were about 28 - 31.30°C in the early morning and 28.57 - 32.13°C in the afternoon (**Appendix 1-3**). Started from measurement at day 10, water temperature showed rising tendency. pH values did not exceed 8.5 and remained above 7.0 throughout the experimental period (**Appendix 1-2**). The lowest (7.07) & highest (8.44) pH values were recorded on day 4 at dawn and on day 36 at noon, respectively. During the experimental period, the fluctuation of water salinity was about 4 ppt (**Appendix 1-4**). However, the lowest and the highest salinity at dawn measurement was 2‰ higher than that of at noon.

Ammonia concentration tended to be low in all treatments just after day 17 of the experimental period and increase afterward, except treatment SL430 and SL480 (**Figure 1-2a**). However, according to the decreasing ammonia concentration, treatment SS30, SL430, SL480, and SL030 were significant different decreasing rate ($P < 0.05$) than treatment SS80, SL080, and SB0 (**Figure 1-2a** and **Appendix 1-5**). Nitrite concentration presented a similar pattern to the trend of ammonia (**Figure 1-2b**). The treatment SL430 and SL480 showed contrary trend and had significant higher ability ($P < 0.05$) of decreasing nitrite concentration than the other treatments (**Figure 1-2b** and **Appendix 1-6**). The

lowest concentration of last measurement and highest ability decreasing concentration were recorded at treatment SL430 of about 0.1005 mg.l^{-1} and $0.5289 \pm 0.194 \text{ mg.l}^{-1}$ and SL480 of about 0.1589 mg.l^{-1} and $0.3889 \pm 0.070 \text{ mg.l}^{-1}$, respectively (**Figure 1-2b** and **Appendix 1-6**). The rest parameters had significant higher concentration at last measurement and lower decreasing concentration of nitrite. Nitrate concentration of treatment SL030 & control (SBO) showed decreasing tendency and the other treatments presented opposite tendency (**Figure 1-2c**). Treatment SS80 and SL480 revealed higher decreasing concentration of nitrate than the others (**Figure 1-2c** and **Appendix 1-7**).

Contrary to the previous parameters, hydrogen sulphide revealed increasing at all treatments (**Figure 1-3a**). Reducing hydrogen sulphide concentration of treatment SS30 and SL430 was not significant different from SS80 but significant different from the other treatments ($P < 0.05$) (**Figure 1-3a** and **Appendix 1-8**). Treatment SB0 revealed significant increase in hydrogen sulphide concentration (**Appendix 1-8**). During the experiment, TOM concentration presented decreasing tendency (**Figure 1-3b**). At the end of the experiment, treatment SL480 revealed significant different from the other treatments (**Appendix 1-9**). Treatment SL480 showed significantly higher ($20.0470 \pm 0.359 \text{ mg.l}^{-1}$) ability in reducing TOM concentration than that of the other treatments (**Figure 1-3** and **Appendix 1-9**).



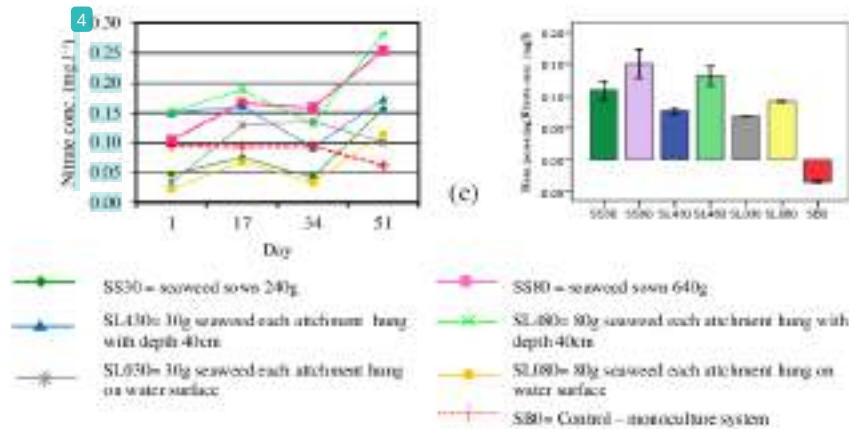
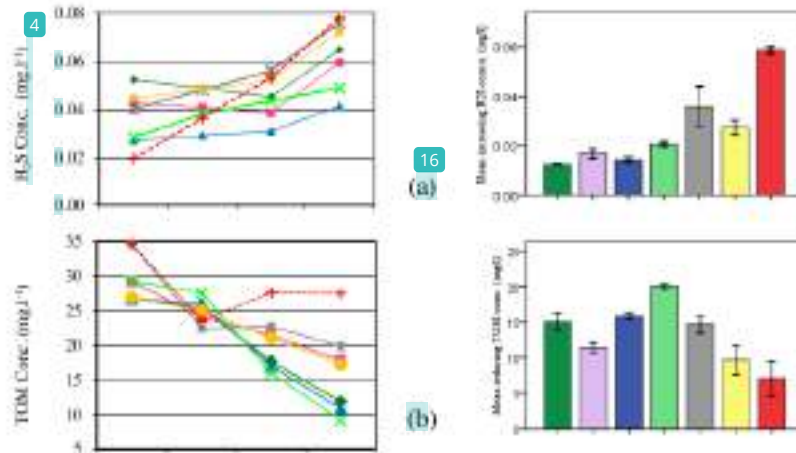


Figure 1-2

Measurement results (left graphs) of and reucing concentration (right graphs) for ammonia (a), nitrite (b), and nitrate concentration (c) in polyculture system of black tiger shrimp-seaweed and black tiger shrimp monoculture system

Generally, PO₄ concentration of most treatments tended to increase at the beginning of experiment period and decrease afterward except treatment SL480. Treatment SL480 showed relatively flat trend of PO₄ concentration from first till last measurement (Figure 1-3c). Treatment SL030 was not significant different from treatments SS30 and SB0 as control but significantly sound lower than that of the rest parameters (Figure 1-3c and Appendix 1-10).



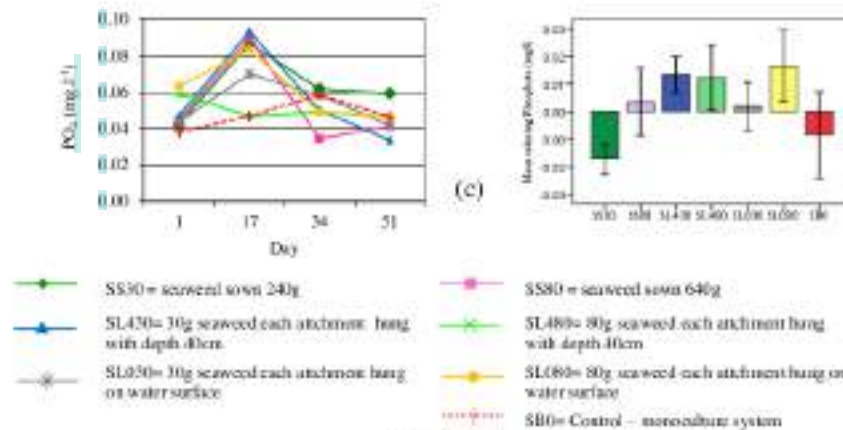
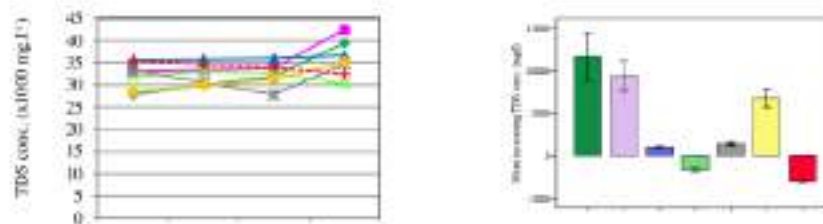


Figure 1-3

Measurement results (left graphs) of and reducing concentration (right graphs) for hydrogen sulphide (a), total organic matter (b), and phosphate concentration (c) in polyculture system of black tiger shrimp-seaweed and black tiger shrimp monoculture system

During the experiment, TDS concentration of treatment SL480 tended to decrease significantly (**Figure 1-4 - upper**). At the end of the experiment, treatment SL480 and SB0 as control demonstrated significantly higher ability in reducing TDS concentration ($P < 0.05$) than the other treatments (**Figure 1-4 - upper** and **Appendix 1-11**). Meanwhile, treatment SS30 and SS80 had significantly lower ability in maintaining concentration of TDS than the other treatments. All treatments except SB0 as control presented increase TSS concentration at the end of experiments (**Figure 1-4 - lower**). Treatment SB0 as control by employing monoculture system showed significantly higher reducing TSS concentration ($P < 0.05$) than the other treatments (**Appendix 1-12**).



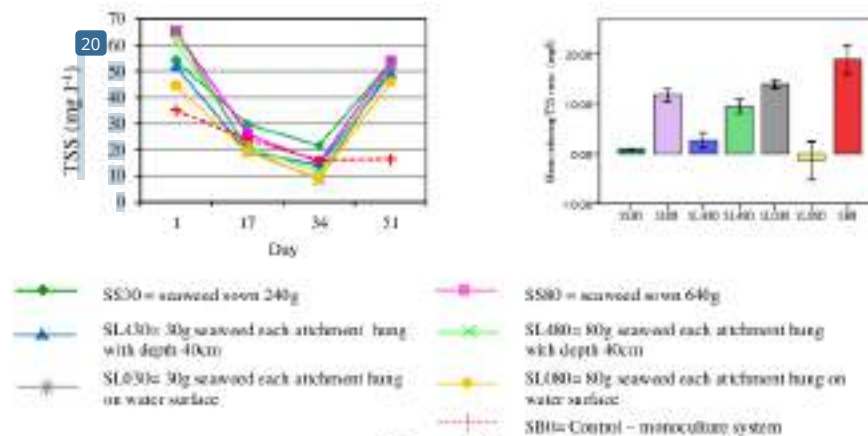


Figure 1-4

Measurement results of (left graphs) and maintaining capacity (right graphs) for Total Dissolved Solid (TDS) (upper) and Total Suspended Solid (lower) in polyculture system of black tiger shrimp-seaweed and black tiger shrimp monoculture system

Biological aspects

The main indicators of success in aquaculture practices are the mortality and growth rate of shrimp as the main cultured organism. During this experiment, all individuals of shrimp in one replication of treatment SS30, SS80, SL080 and 1 individual shrimp in one replicate each of treatment SL480 and SL30 died (**Appendix 1-14**).

The average harvested weight in all treatments varied from 11.85 g to 19.98 g per individual shrimp. Related to the shrimp growth, the average daily growth rate was not significant different amongst treatment S30, SL430 and SL480 and significant different from treatments SS80, SL30, SL80, and SB0 ($P < 0.05$) (**Figure 1-5; Appendix 1-13; Appendix 1-14; Appendix 1-15; Appendix 1-16**).

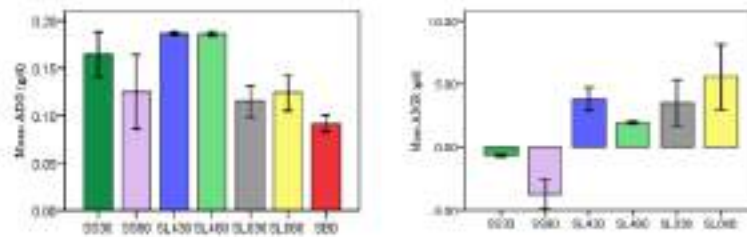


Figure 1-5

Average daily growth rate of shrimp (left) and seaweed (right) in polyculture system

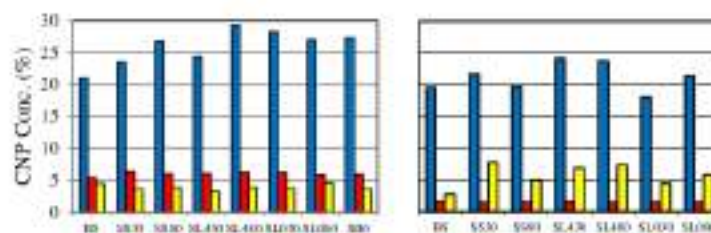
The seaweed used in this experiment had a wide range of biomass at harvest. The polyculture black tiger shrimp with bottom method of both 240 g (treatment SS30) and 640 g (treatment SS80) had lower average harvested biomass of about 203.55 g and 440.80 g than that of stocking, respectively (**Appendix 1-16**). The heaviest weight gain was performed by treatment SL080 (polyculture with 80 g seaweed of each attachment) with the average daily growth rate of about 5.5527 ± 2.561 g. However, its average daily growth rate had not significant different compare to treatment SL430 and SL030S.

Furthermore, C, N, and P content in shrimp and seaweed tissue were also measured. C content in shrimp and seaweed tissue was higher than N and P content (**Figure 1-6**). Statistically, concentration of C in shrimp tissue of treatment SL480 was not significant different from treatment SL030 but significant different concentration from the other treatments ($P < 0.05$). Nitrogen concentration of treatments SS30 was not significant different from treatment SL480 and SL030. However, the treatment SL480 and SL030 were found no significant different from the rest treatments. The Phosphorous concentration of treatment SL080 was significant different from the other treatments ($P < 0.05$).

In seaweed tissue (**Figure 1-6**), SL430 and SL480 had significant highest concentration of C but there was no significant difference between treatments SL480, SS30, SS80 and SL080. The lowest concentration was found in treatment SS80 and SL030. Concentration of N was not significantly different amongst the treatments. Concentration of P of treatment SS30 were not significant different

from treatment SL480 but were significantly different from the other treatments. The lowest concentration of P was attributed by treatment SS80 and SL030.

During experiment, total number of *Vibrio* in treatments SS80, SL430 and SB0 showed a relatively flat trend and bit decrease at the end of experiment. However, *Vibrio* numbers in the other treatments showed a tendency to increase at the beginning at the experiment and decrease later (Figure 1-7). A different trend was observed for total bacterial. There were two common trend of total bacterial. Firstly, the trend of treatments SL080, SL430, SL480, and SB0 was similar to that of total *Vibrio* (Figure 1-7). Secondly, a tendency in reduction in numbers through the course of the experiment was seen in treatments SS30, SS80, and SL030. As presented in Appendix 1-17, at the end of experiment total *Vibrio* and bacteria had similar levels of about 10^2 (total *Vibrio*) and $10^5 - 10^6$ (bacteria), respectively. However, at the end of the experiment treatments SS80 and SL480 had higher total number of *Vibrio* than the other treatments of about $8.13 \times 10^2 \pm 90.19$ CFU/ml and $8.80 \times 10^2 \pm 131.15$ CFU/ml, respectively (Appendix 1-18). Number of total *Vibrio* in treatments SS30, SL030, and SB0 were significantly higher than in SL430, SL080 and significantly lower than SS80 and SL480. The number of total bacteria in treatment SS80 was significantly higher than SS30, SL430, SL030, SL080, and SB0. SL030 showed the highest total bacteria count amongst the treatments. SL080 was not significant different from SB0 (control) and showed the lowest number of total bacteria amongst the others.



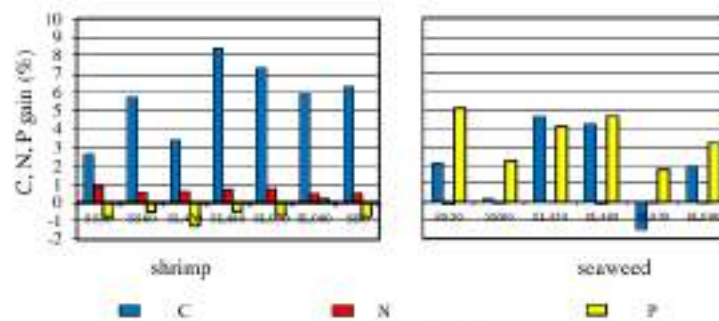


Figure 1-6

C, N, P content and gained in shrimp (left graphs) and seaweed tissue (right graphs) in polyculture system

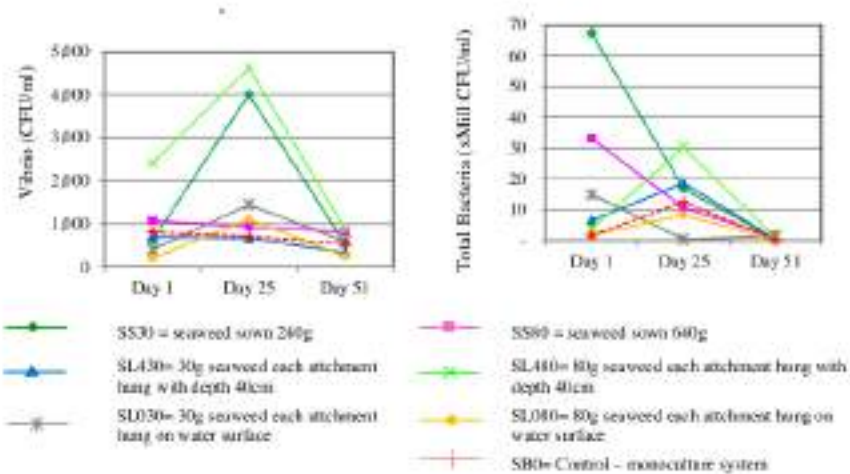


Figure 1-7

Total *Vibrio* (left) and bacteria (right) during experiment in polyculture black tiger shrimp with seaweed

DISCUSSION

One of the components of the shrimp pond culture ecosystem is water as the culture medium which interacts ecologically with other components. During the rearing period, water quality of shrimp pond tends to decrease due to added production inputs. Using of filtration organisms, such as seaweed (*G. verrucosa*) and blood cockle (*A. granosa*), in a polyculture system seems to be able to reduce

the environmental impacts of culture activities. Seaweed as a phototrophic organism will absorb micro- and macro- dissolved nutrients from the environment. Meanwhile, blood cockle as a filter-feeder will ingest dissolved and suspended particulates from the environment. Therefore, both those filtration organisms naturally seem to be able to reduce organic and inorganic substances in the culture medium. One of the ecological consequences is to stabilize shrimp pond culture habitat.

Shrimp-seaweed polyculture system

In this study, employing *G. verrucosa* with different levels of stocking weight and cultivation methods improved water quality. The concentration of dissolved oxygen concentration increased with the method of seaweed cultivation. Black tiger shrimp Polyculture with *G. verrucosa* hung 40 cm under water surface showed higher dissolved oxygen concentration than the other treatments. It probably due to diffusion and solubility rate of oxygen in water resulted from photosynthesis (Boyd 1992), the concentration in dawn was lower than in the afternoon and the position of seaweed in water column. After day 30 of dawn observation of experimental period, DO concentration in the shrimp polyculture with seaweed cultivated by bottom and on the surface water long-line methods decreased much sharper than in shrimp polyculture with seaweed cultivated by hung 40 cm from water surface. The position of seaweed cultivation to the water surface influence sun light penetration, photosynthesis rate and subsequently related to the adding up of dissolved oxygen to the culture water. Bottom method culture will cause lower intensity of sun light and photosynthesis rate of seaweed. On the other hand, on the water surface position, seaweed will exposure to the highest sun light intensity but oxygen resulted from photosynthesis process might easily be released out to the air due to the low water solubility rate. Therefore, during the experiment, polyculture (shrimp – *G. verrucosa*) with seaweed hung 40 cm under water surface provided more than the minimum requirement of DO concentration according to the requirement for black tiger shrimp culture of more than 3 mg.l⁻¹ (Boyd 1992; Boyd and Green 2002; Darmono 1993). Suitable dissolved oxygen concentration of shrimp culture water

is one of main important ⁷ water quality parameters to support growth and survival of shrimp and the other ecological processes within culture ecosystem (Boyd 1990; Hawerton 2001).

Interesting results were observed in all polyculture system black tiger shrimp (*P. monodon*) with *G. verrucosa* in term of ammonia, nitrite, nitrate, hydrogen sulphide, total organic matter and total dissolved solid concentration of which tended to decrease with culture period. Aquaculture practices lead to have organic matter rich sediment accumulated on the pond bottom and degraded water quality from which may cause eutrophication, hypereutrophication, and low DO concentration (Donovan 2001; Pillay 2004; FAO 2006). Decreasing concentration of several water quality parameters were comparable to the study reported by Jones and Preston (1999). Seaweed as a phototropic organism cultivated along with shrimp in the same culture unit might have the ability to lift up DO concentration of culture water habitat and ensure ecological process included production, consumption, and decomposition (Zonneveld *et al.* 1991; Wetzel 1993). It might reduce nutrient loading resulted by shrimp rearing activities (Troell 1999). Dissolved oxygen concentration trend appeared to have relationship with the concentration trend ⁶ of ammonia, nitrite, nitrate, total organic matter, and phosphate. The decomposition process of organic substances was supported by availability of dissolved oxygen concentration (Boyd 1990; Knud-Hansen 1998; Effendi 2003). Since, ammonia concentration in aquaculture ecosystem tends to increase with culture period resulted from fish excretion (Hargreaves & Tucker 2004). In this study, *G. verrucosa* in polyculture with shrimp might have potential for playing an important ecological role in order to maintain suitable water culture media for shrimp, related to the DO, ammonia, nitrite, nitrate, hydrogen sulphide, total organic matter and total dissolved solids concentration.

Moreover, the range of those water quality parameters was within the range of black tiger shrimp requirement (Boyd 1990). The highest ammonia, nitrite, and total organic matter concentration was found at the monoculture system. This may have resulted from the lower ability of the system to oxidize and mineralize accumulated organic matter (Knud-Hansen 1998). Contrary to

results mentioned previously, total suspended solid was not affected by the inclusion of *G. verrucosa*.

Black tiger shrimp (*Penaeus monodon*) as the main culture organism can be used as a biological indicator of achievement of aquaculture practices. The best growth rate in these experiments was observed from the shrimp polyculture system with 30 and 80 g seaweed hung 40 cm under the water surface and seaweed cultivated by bottom method with total weight of 240 g. However, these growth rates were lower than those found by Lumare (1993). Another result of biological factors investigations were bacterial colony and C, N, P content in the shrimp and seaweed tissue. All different culture systems did not affect the colony of *Vibrio* and total bacteria. It is contrary to the study performed by Jones and Petterson (1991) and Troll *et al.* (1999). In general, shrimp absorbed nitrogen from the environment and released phosphorus to the system. In contrast, seaweed could absorb phosphorus from the system. It seems to be useful in maintaining P concentration within the system. Because, in aquaculture practices, phosphorus input through feed rang from 68.8 to 90.6% of the total (Thakur & Lin 2003). Besides that, phosphorus has tendency to be demobilize to the system (Boyd 1990).

CONCLUSION AND RECOMMENDATION

Conclusion

Based on the result of study concluded as follows:

1. Black tiger shrimp (*P. monodon*) polyculture with seaweed (*G. verrucosa*) was tied with about 30 g at each attachment and hung with depth of 40 cm from water surface had ability to stabilize culture media by maintaining several water quality parameters.
2. The best growth rate was achieved by polyculture system of black tiger shrimp (*P. monodon*) with 30 and 80 g seaweed (*G. verrucosa*) at each attachment and hung with depth of 40 cm from water surface and 240 g seaweed cultivated by bottom method.
3. All the systems had suitable collony of micro-organism based on magnification of *Vibrio* of about 10^2 and total bacteria of 10^6 .

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Appendix 1-1

Results of dissolved oxygen (mg.l^{-1}) measurement in dawn in polyculture black tiger shrimp seaweed

Treatment	Day													
	1	4	7	10	13	16	19	26	30	36	40	44	48	51
Dawn														
SS30	5.23	5.00	2.82	2.81	1.76	2.72	2.60	2.99	3.14	2.51	1.99	2.13	2.17	2.15
SS80	5.10	4.97	3.00	2.85	2.81	2.87	2.78	2.68	2.98	2.77	2.22	1.77	2.14	2.11
SL430	4.42	4.32	2.93	2.81	2.67	2.99	2.90	3.00	3.36	3.01	2.61	2.68	2.95	2.83
SL480	5.52	5.01	2.85	2.86	2.78	3.00	2.68	3.01	3.20	3.01	2.74	2.76	3.05	3.01
SL030	4.52	4.64	2.69	2.42	2.72	2.45	2.56	2.68	3.00	2.91	1.79	1.99	2.01	1.98
SL080	3.25	4.37	2.50	1.80	2.06	2.46	2.55	2.64	2.71	2.91	1.61	1.79	1.94	1.94
SB0	4.47	4.92	3.09	2.90	2.81	2.76	2.82	2.87	3.34	2.91	2.36	1.80	2.27	2.25
Noon														
SS30	5.60	5.52	3.11	3.53	3.76	2.70	2.77	2.87	3.15	3.78	3.07	3.01	2.86	2.95
SS80	5.44	5.41	3.53	3.46	3.76	2.60	2.92	2.85	3.31	3.92	3.17	2.80	2.92	2.92
SL430	5.52	5.43	3.40	3.57	4.81	3.31	3.43	3.39	3.73	4.86	3.47	3.21	3.23	3.13
SL480	6.01	5.44	3.38	3.36	4.48	3.16	3.43	3.38	3.59	4.63	3.50	3.33	3.38	3.31
SL030	5.18	5.08	3.16	2.97	4.31	2.58	2.84	3.05	3.36	4.30	2.75	2.92	2.80	2.77
SL080	3.72	4.92	3.04	2.74	2.17	2.80	2.90	3.05	3.11	4.20	2.61	2.82	2.78	2.74
SB0	4.74	5.26	3.44	3.26	3.50	2.60	2.98	2.90	3.38	4.00	2.94	1.83	2.80	2.29

Note:

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hung with depth 40cm

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control – monoculture system

Appendix 1-2

Results of pH measurement in dawn in polyculture black tiger shrimp seaweed

Treatment	Day													
	1	4	7	10	13	16	19	26	30	36	40	44	48	51
Dawn														
SS30	7.92	7.13	7.67	7.60	7.91	8.07	8.00	8.06	8.21	8.15	8.08	8.02	8.13	8.13
SS80	7.91	7.07	7.40	7.53	7.95	8.04	8.00	8.38	8.30	8.12	8.23	8.20	7.99	8.08
SL430	7.91	7.13	7.60	7.73	8.28	8.09	7.99	8.35	8.26	8.12	8.14	8.16	8.26	8.33
SL480	7.91	7.07	7.47	7.47	7.86	7.88	8.00	8.41	8.35	7.98	8.01	8.01	8.30	8.21
SL030	7.91	7.13	7.53	7.60	7.96	7.97	7.97	8.29	8.26	8.20	8.22	8.19	8.18	8.15
SL080	7.84	7.13	7.73	7.60	7.81	7.88	8.01	8.43	8.27	8.24	8.16	8.15	8.17	8.24
SB0	7.88	7.07	7.67	7.53	8.15	8.20	8.00	8.10	8.08	8.32	8.20	8.15	8.12	8.14
Noon														
SS30	7.68	7.66	7.73	8.01	7.98	8.01	8.05	8.38	8.15	8.11	8.14	8.17	8.21	8.23
SS80	7.75	7.67	7.73	7.93	8.00	8.16	8.06	8.41	8.17	8.17	8.10	7.93	8.09	8.22
SL430	7.88	7.68	7.93	8.07	8.06	7.91	8.04	8.41	8.20	8.36	8.04	8.21	8.33	8.43
SL480	7.64	7.51	7.53	7.83	7.92	8.11	8.04	8.41	8.18	8.44	8.11	8.15	8.33	8.31
SL030	8.21	7.64	7.80	7.98	8.11	8.02	8.01	8.33	8.16	8.28	8.08	8.12	8.28	8.38
SL080	7.59	7.58	8.07	7.87	7.82	7.88	8.06	8.43	8.23	8.30	8.16	7.99	8.27	8.34
SB0	7.97	7.81	7.67	8.08	8.14	7.96	8.04	8.20	8.01	8.15	8.20	8.26	8.22	8.28

Note:

SS30 = seaweed sown 240g

SL430 = 30g seaweed each attachment hang with depth 40cm

SL030 = 30g seaweed each attachment hang on water surface

SS80 = seaweed sown 640g

SL480 = 80g seaweed each attachment hang with depth 40cm

SL080 = 80g seaweed each attachment hang on water surface

SB0 = Control - monoculture system

Appendix 1-3

Results of temperature (°C) measurement in dawn in polyculture black tiger shrimp seaweed

Treatment	Day													
	1	4	7	10	13	16	19	26	30	36	40	44	48	51
DAWN														
SS30	28.97	28.73	28.00	29.67	29.97	30.77	30.67	29.83	28.93	30.37	30.47	29.92	29.47	29.43
SS80	28.90	28.27	28.00	29.67	30.00	30.17	30.25	30.33	29.33	31.07	31.68	30.73	30.10	29.83
SL40	29.07	28.83	28.00	30.32	29.83	30.00	30.17	30.33	29.30	30.90	31.10	30.47	30.00	29.73
SL80	29.07	29.17	28.00	29.67	30.07	30.93	30.40	30.33	29.37	30.71	30.83	30.43	29.71	29.63
SL030	29.00	28.73	28.00	30.13	30.03	30.57	30.60	30.30	29.43	30.63	30.93	30.20	29.07	29.87
SL080	29.40	28.63	28.00	30.32	30.00	30.03	30.23	30.43	29.40	31.03	31.23	30.47	30.10	29.97
SB0	29.07	28.00	28.00	29.67	29.00	30.37	30.32	30.07	29.37	31.10	31.30	30.13	30.11	30.13
NOON														
SS30	29.40	28.53	30.00	28.93	30.37	30.70	30.88	31.37	30.53	32.13	32.00	30.23	31.10	31.23
SS80	29.17	28.57	30.00	29.10	30.17	30.37	30.75	31.40	30.47	31.80	31.88	30.33	31.07	31.07
SL40	29.40	28.53	30.00	28.43	30.13	30.67	30.47	31.17	30.33	31.90	31.97	30.30	30.87	30.87
SL80	29.37	28.70	30.00	28.92	30.40	30.50	30.73	31.43	30.53	31.90	31.77	30.30	30.80	30.87
SL030	29.33	28.57	30.00	29.12	30.13	30.37	30.97	31.30	30.53	31.83	31.87	30.37	30.97	31.07
SL080	29.77	28.80	30.00	28.90	30.23	30.23	30.63	31.40	30.43	31.77	32.10	30.33	31.07	31.13
SB0	29.43	28.70	30.00	29.13	30.53	30.47	30.68	31.10	30.47	31.60	32.13	30.43	31.10	31.40

Note:

SS30 = seaweed crown 340g

SS80 = seaweed crown 640g

SL40= 30g seaweed each attachment hang with depth 40cm

SL80= 80g seaweed each attachment hang with depth 40cm

SL030= 30g seaweed each attachment hang on water surface

SL080= 80g seaweed each attachment hang on water surface

SB0 = Control - monoculture system

Appendix 1-4

Results of Salinity (‰) measurement in dawn in polyculture black tiger shrimp seaweed

Treatment	Day													
	1	4	7	10	13	16	19	26	30	36	40	44	48	51
DAWN														
SS30	31.33	33.00	31.33	29.67	29.67	30.00	30.00	32.00	27.67	34.33	33.00	33.00	29.33	30.00
SS80	31.33	33.33	32.33	29.67	31.00	30.33	30.00	34.00	28.67	36.33	34.50	32.67	30.00	30.00
SL430	31.33	33.00	34.00	30.33	31.00	30.00	30.00	34.67	29.00	35.33	32.67	32.00	30.00	30.33
SL480	32.00	35.33	32.00	29.67	30.67	30.33	30.00	31.67	28.00	35.00	33.83	35.00	29.67	30.33
SL030	31.00	33.00	33.00	30.33	30.33	30.00	30.00	33.67	28.67	35.33	33.00	32.67	30.00	30.00
SL080	30.67	33.67	35.00	30.67	30.00	30.00	30.00	34.33	29.00	35.33	32.67	32.67	30.00	30.00
SB0	32.00	33.67	33.00	29.67	31.00	31.00	30.33	34.00	28.00	35.33	32.67	32.67	29.67	30.00
NOON														
SS30	26.00	25.33	28.00	30.33	27.67	31.33	30.00	29.67	28.33	33.33	33.50	30.00	29.33	30.00
SS80	26.00	25.33	29.00	30.33	29.33	32.00	29.33	29.33	29.00	32.67	33.83	31.00	29.33	29.33
SL430	26.00	25.67	29.00	30.67	29.00	33.00	29.67	30.33	29.33	33.33	32.33	30.33	29.00	29.67
SL480	26.00	25.00	28.67	30.33	30.00	32.00	30.00	29.33	28.00	32.00	34.17	29.00	28.67	30.00
SL030	25.67	25.33	28.67	30.67	27.00	32.33	30.00	30.00	29.33	32.00	33.33	31.00	29.00	29.00
SL080	25.33	26.00	29.67	31.00	28.33	33.33	29.33	29.00	28.67	33.00	32.33	29.00	29.00	29.67
SB0	25.67	24.67	28.67	30.33	29.67	32.00	30.00	29.67	28.67	32.33	33.00	29.00	28.67	29.33

Note:

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hang with depth 40cm

SL030= 30g seaweed each attachment hang on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hang with depth 40cm

SL080= 80g seaweed each attachment hang on water surface

SB0 = Control - monoculture system

Appendix 1-5

Result of statistical analysis of mean value±SD of reducing ammonia at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	NH ₃ Concentration (mg l ⁻¹)		Δ NH ₃
	T0	T1	
SS30	0.4715	0.4208	0.0507±0.0375a
SS80	0.2245	0.5795	-0.3551±0.0748b
SL430	0.3293	0.2620	0.0673±0.188a
SL480	0.3048	0.1837	0.1211±0.0686a
SL030	0.4925	0.4338	0.0588±0.0326a
SL080	0.4495	0.7383	-0.2888±0.0108b
SB0 (Control)	0.1841	0.5108	-0.3268±0.0717b

Notes:

ab Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hang with depth 40cm

SL030= 30g seaweed each attachment hang on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hang with depth 40cm

SL080= 80g seaweed each attachment hang on water surface

SB0 = Control - monoculture system

Appendix 1-6

Result of statistical analysis of mean value \pm SD of reducing nitrite at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	NO ₂ Concentration (mg l ⁻¹)		ANO ₂
	T0	T1	
SS30	0.4259	0.3723	0.0535 \pm 0.013a
SS80	0.6106	0.4331	0.1774 \pm 0.065a
SL430	0.6293	0.1005	0.5289 \pm 0.194b
SL480	0.5479	0.1589	0.3889 \pm 0.070b
SL030	0.5554	0.4350	0.1205 \pm 0.010a
SL080	0.4691	0.4054	0.0637 \pm 0.028a
SB0 (Control)	0.5864	0.4880	0.0984 \pm 0.051a

Notes:

ab Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-7

Result of statistical analysis of mean value \pm SD of reducing nitrate at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	NO ₃ Concentration (mg l ⁻¹)		ANO ₃
	T0	T1	
SS30	0.0478	0.1569	0.1091 \pm 0.146a
SS80	0.1043	0.2547	0.1504 \pm 0.022b
SL430	0.1500	0.1732	0.0232 \pm 0.005cd
SL480	0.1514	0.2827	0.1313 \pm 0.016b
SL030	0.0334	0.1014	0.0680 \pm 0.0005c
SL080	0.0227	0.1142	0.0915 \pm 0.001ad
SB0 (Control)	0.0955	0.0617	-0.0338 \pm 0.0015e

Notes:

abcde Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-8

Result of statistical analysis of mean value \pm SD of reducing hydrogen sulphide at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	H ₂ S Concentration (mgJ ⁻¹)		ΔH ₂ S
	T0	T1	
SS30	0.0523	0.0647	0.0124±0.0004a
SS80	0.0425	0.0593	0.0168±0.0020ab
SL430	0.0270	0.0413	0.0143±0.0011a
SL480	0.0282	0.0488	0.0206±0.0010b
SL030	0.0399	0.0757	0.0358±0.0084c
SL080	0.0447	0.0720	0.0273±0.0030d
SB0 (Control)	0.0194	0.0781	0.0586±0.0014c

Notes:

abcd: Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-9

Result of statistical analysis of mean value±SD of reducing total organic matter at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	TOM Concentration (mgJ ⁻¹)		ΔTOM
	T0	T1	
SS30	26.8987	11.8630	15.0357±1.184a
SS80	29.2657	17.8573	11.4083±0.717b
SL430	26.7127	10.8643	15.8483±0.481a
SL480	29.2460	9.1990	20.0470±0.359c
SL030	34.5847	19.8553	14.7293±1.310a
SL080	26.9260	17.2503	9.6757±2.027b
SB0 (Control)	34.6027	27.5127	7.0900±2.449d

Notes:

abcd: Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-10

Result of statistical analysis of mean value±SD of reducing phosphate at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	PO ₄ Concentration (mgJ ⁻¹)	ΔPO ₄
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	T0	T1	
SS30	0.0423	0.0593	-0.0170±0.0057a
SS80	0.0445	0.0411	0.0034±0.0122b
SL430	0.0470	0.0336	0.0134±0.0064b
SL480	0.0588	0.0465	0.0123±0.0119b
SL030	0.0437	0.0417	0.0019±0.0086ab
SL080	0.0632	0.0466	0.0166±0.0128b
SB0 (Control)	0.0381	0.0465	-0.0083±0.0156ab

Notes:

ab Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-11

Result of statistical analysis of mean value±SD of reducing total dissolved solid at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	TDS Concentration (mgJ ⁻¹)		ΔTDS
	T0	T1	
SS30	27,838.67	39,466.67	11,628.00±2,825.10a
SS80	33,094.00	42,466.67	9,372.67 ±1,770.84a
SL430	35,623.33	36,633.33	1,010.00 ±92.71b
SL480	31,898.00	30,233.33	-1,664.67±213.85c
SL030	33,402.33	34,766.67	1,364.33±244.36b
SL080	28,414.33	35,200.00	6,785.67±1085.28d
SB0 (Control)	35,544.00	32,500.00	-3,044.00±55.51c

Notes:

abcd Means with different letters at the same column are significantly different at P<0.05

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-12

Result of statistical analysis of mean value±SD of reducing total suspended solid at first and last measurement in polyculture black tiger shrimp and seaweed

Treatment	TSS Concentration (mgJ ⁻¹)		ΔTSS
	T0	T1	

SS30	54.20	53.33	0.87 ±0.06a
SS80	65.34	53.67	11.67 ±1.25bc
SL430	51.93	49.33	2.60±1.51a
SL480	61.40	52.00	9.40±1.39c
SL030	65.80	52.00	13.80±0.79b
SL080	44.60	46.00	-1.40±3.81a
SB0 (Control)	35.20	16.38	18.82±2.89d

Notes:

abcd Means with different letters at the same column are significantly different at $P < 0.05$

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hung with depth 40cm

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-13

Average weight at stocking-harvesting time and absolute and daily growth rate of black tiger shrimp

Treatment	R	Average Weight (g)		Growth rate	
		Stocking	Harvesting	ABS. (g)	Daily (g.d ⁻¹)
SS30	1	8.8700	0.0000	0.0000	0.0000
	2	6.8650	14.1750	7.3100	0.1379
	3	8.6100	18.7250	10.1150	0.1908
	Mean	8.1150	10.9667	5.8083	0.1096
SS80	1	9.0600	15.9100	6.8500	0.1292
	2	8.3650	14.7450	6.3800	0.1204
	3	5.8750	0.0000	0.0000	0.0000
	Mean	7.7667	10.2183	4.4100	0.0832
SL430	1	5.5900	15.5700	9.9800	0.1883
	2	9.0200	18.8450	9.8250	0.1854
	3	6.9350	16.8300	9.8950	0.1867
	Mean	7.1817	17.0817	9.9000	0.1868
SL480	1	11.3400	21.2400	9.9000	0.1868
	2	11.2250	20.9800	9.7550	0.1841
	3	7.7300	17.7100	9.9800	0.1883
	Mean	10.0983	19.9767	9.8783	0.1864
SL030	1	8.1500	14.2100	6.0600	0.1143
	2	8.3250	14.4800	6.1550	0.1161
	3	11.4550	17.5000	6.0450	0.1141
	Mean	9.3100	15.3967	6.0867	0.1148
SL080	1	9.5400	15.9000	6.3600	0.1200
	2	10.1650	0.0000	0.0000	0.0000
	3	7.5450	14.3200	6.7750	0.1278
	Mean	9.0833	10.0733	4.3783	0.0826
SB0	1	6.0050	10.1350	4.1300	0.0779
	2	9.9600	0.0000	0.0000	0.0000
	3	10.3800	13.5700	3.1900	0.0602
	Mean	8.7817	7.9017	2.4400	0.0460

Note:

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hung with depth 40cm

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control - monoculture system

Appendix 1-14

Shrimp mortality rate in polyculture black tiger shrimp and seaweed

Treatment	Replication		
	1	2	3
SS30	100	0	0
SS80	0	0	100
SL430	0	0	0
SL480	0	0	0
SL030	0	0	0
SL080	0	100	0
SB0 (Control)	0	100	0

Note:

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hung with depth 40cm

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control – monoculture system

Appendix 1-15

Result of ONEWAY ANOVA Growth of black tiger shrimp by treatment in polyculture system shrimp-seaweed

	Sum of Squares	DF	Mean Square	F	Sig.
Between Groups	0.024	6	0.004007	13.7398	0.000153
Within Groups	0.003	11	0.000292		
Total	0.027	17			

Multiple Comparisons

Treatment (I)	Treatment (J)	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
SS30	SS80	.0395500*	.0170779	.041	.001962	.077138
	SL430	-.0224000	.0155899	.179	-.056713	.011913
	SL480	-.0220000	.0155899	.186	-.056313	.012313
	SL030	.0495333*	.0155899	.009	.015220	.083846
	SL080	.0404500*	.0170779	.037	.002862	.078038
	SBO	.0729333*	.0155899	.001	.038620	.107246
SS80	SS30	-.0395500*	.0170779	.041	-.077138	-.001962
	SL430	-.0619500*	.0155899	.002	-.096263	-.027637
	SL480	-.0615500*	.0155899	.002	-.095863	-.027237
	SL030	.0099833	.0155899	.535	-.024330	.044296
	SL080	.0090000	.0170779	.559	-.036688	.038488
	SBO	.0333833	.0155899	.055	-.000930	.067696
SL430	SS30	.0224000	.0155899	.179	-.011913	.056713
	SS80	.0619500*	.0155899	.002	.027637	.096263
	SL480	.0040000	.0139440	.978	-.030291	.031091
	SL030	.0719333*	.0139440	.000	.041243	.102624
	SL080	.0628500*	.0155899	.002	.028537	.097163
	SBO	.0953333*	.0139440	.000	.064643	.126024
SL480	SS30	.0220000	.0155899	.186	-.012313	.056313
	SS80	.0615500*	.0155899	.002	.027237	.095863
	SL430	-.0004000	.0139440	.978	-.031091	.030291
	SL030	.0715333*	.0139440	.000	.040843	.102224
	SL080	.0624500*	.0155899	.002	.028137	.096763
	SBO	.0949333*	.0139440	.000	.064243	.125624
SL030	SS30	-.0495333*	.0155899	.009	-.083846	-.015220
	SS80	-.0099833	.0155899	.535	-.044296	.024330
	SL430	-.0719333*	.0139440	.000	-.102624	-.041243
	SL480	-.0715333*	.0139440	.000	-.102224	-.040843
	SL080	-.0090833	.0155899	.572	-.043396	.025230
	SBO	.0234000	.0139440	.121	-.007291	.054091
SL080	SS30	-.0404500*	.0170779	.037	-.078038	-.002862
	SS80	-.0090000	.0170779	.559	-.036688	.038488
	SL430	-.0628500*	.0155899	.002	-.097163	-.028537
	SL480	-.0624500*	.0155899	.002	-.096763	-.028137
	SL030	.0090833	.0155899	.572	-.025230	.043396
	SBO	.0324833	.0155899	.061	-.001830	.066796
SBO	SS30	-.0729333*	.0155899	.001	-.107246	-.038620
	SS80	-.0333833	.0155899	.055	-.067696	.000930
	SL430	-.0953333*	.0139440	.000	-.126024	-.064643
	SL480	-.0949333*	.0139440	.000	-.125624	-.064243
	SL030	-.0234000	.0139440	.121	-.054091	.007291
	SL080	-.0324833	.0155899	.061	-.066796	.001830

*. The mean difference is significant at the 0.05 level.

Appendix 1-16

Result of statistical analysis of mean value \pm SD of daily growth rate of 53 rearing period of black tiger shrimp and seaweed in polyculture system

Treatment	Shrimp			Seaweed		
	W0 (g)	W1 (g)	DGR (g.d ⁻¹)	W0 (g)	W1 (g)	DGR (g.d ⁻¹)
SS30	8.12	16.45	0.1644 \pm 0.0240a	240	203.55	-0.6877 \pm 0.104a
SS80	7.77	15.33	0.1248 \pm 0.0391b	640	440.80	-3.7584 \pm 1.153b
SL430	7.18	17.08	0.1868 \pm 0.0015a	240	443.16	3.8331 \pm 0.923c
SL480	10.10	19.98	0.1864 \pm 0.0021a	640	742.56	1.9352 \pm 0.107cd
SL030	9.31	15.40	0.1148 \pm 0.0170b	240	423.19	3.4565 \pm 1.832cd
SL080	9.08	15.11	0.1239 \pm 0.0189b	640	934.29	5.5527 \pm 2.561c
SB0 (Control)	8.78	11.85	0.0691 \pm 0.0086b			

Note:

abcd Means with different letters at the same column are significantly different at $p < 0.05$

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430= 30g seaweed each attachment hung with depth 40cm

SL480= 80g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control – monoculture system

Appendix 1-17

Total colony of *Vibrio* and total bacteria in polyculture black tiger shrimp and seaweed

Treatment	<i>Vibrio</i> (CFU/ml)			Total Bacteria (CFU/ml)		
	Day 1	Day 25	Day 51	Day 1	Day 25	Day 51
SS30	6.30 x 10 ²	4 x 10 ¹	5.35 x 10 ²	6.72 x 10 ⁷	1.69 x 10 ⁷	4 x 10 ⁵
SS80	1.07 x 10 ³	9.33 x 10 ²	8.13 x 10 ²	3.31 x 10 ⁷	1.06 x 10 ⁷	1.003 x 10 ⁶
SL430	6.95 x 10 ²	6.7 x 10 ²	3.36 x 10 ²	6.49 x 10 ⁶	1.87 x 10 ⁷	3.7 x 10 ⁵
SL480	2.41 x 10 ³	4.62 x 10 ³	8.80 x 10 ²	3.8 x 10 ⁹	3.03 x 10 ⁷	9.15 x 10 ⁹
SL030	4.03 x 10 ²	1.44 x 10 ³	5.93 x 10 ²	1.49 x 10 ⁷	3.7 x 10 ⁷	1.6 x 10 ⁶
SL080	2.17 x 10 ²	1.11 x 10 ³	2.5 x 10 ²	1.84 x 10 ⁶	8.26 x 10 ⁶	2.33 x 10 ⁵
SB0	8.07 x 10 ²	7.13 x 10 ²	5.4 x 10 ²	1.56 x 10 ⁶	1.25 x 10 ⁷	2.7 x 10 ⁵

Note:

SS30 = seaweed sown 240g

SS80 = seaweed sown 640g

SL430=	30g seaweed each attachment hung with depth 40cm	SL480=	80g seaweed each attachment hung with depth 40cm
SL030=	30g seaweed each attachment hung on water surface	SL080=	80g seaweed each attachment hung on water surface
		SB0 =	Control – monoculture system

Appendix 1-18

Result of statistical analysis of mean value \pm SD of *Vibrio* and total bacterial colloni at last measurement in polyculture black tiger shrimp and seaweed

Treatment	<i>Vibrio</i> (CFU/ml)	Total Bacteria (CFU/ml)
SS30	$5.33 \times 10^2 \pm 20.82a$	$4 \times 10^5 \pm 48,000.00a$
SS80	$8.13 \times 10^2 \pm 90.19b$	$1.003 \times 10^6 \pm 37,527.77b$
SL430	$3.36 \times 10^2 \pm 15.28c$	$3.7 \times 10^5 \pm 75,498.34a$
SL480	$8.80 \times 10^2 \pm 131.15b$	$9.15 \times 10^5 \pm 80,000.00b$
SL030	$5.93 \times 10^2 \pm 60.28a$	$1.6 \times 10^6 \pm 50,000.00c$
SL080	$2.5 \times 10^2 \pm 45.00c$	$2.33 \times 10^5 \pm 15,275.25d$
SB0	$5.4 \times 10^2 \pm 5.00a$	$2.7 \times 10^5 \pm 36,055.51d$

Notes:

abcd Means with different letters at the same column are significantly different at $p < 0.05$

SS30 = seaweed sown 240g

SL430= 30g seaweed each attachment hung with depth 40cm

SL030= 30g seaweed each attachment hung on water surface

SS80 = seaweed sown 640g

SL480= 80g seaweed each attachment hung with depth 40cm

SL080= 80g seaweed each attachment hung on water surface

SB0 = Control – monoculture system

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