

Antioxidant Phytoplankton community at intensive cultivation system of *whiteleg shrimp*, *Litopenaeus vannamei* in Probolinggo, East Java

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Abstract

Phytoplankton is a source of natural feed for shrimp cultivation in the pond. Phytoplankton productivity increases by the increasing nutrient content in the pond. Feed and metabolic waste is the sources of nutrient for phytoplankton growth. This study aimed to evaluate productivity and identify phytoplankton at intensive whiteleg shrimp, *Litopenaeus vannamei* cultivation system. This study was conducted at three intensive whiteleg shrimp located in Probolinggo, East Java. Samples were collected on the early and the late of culture period (day 17 and 87 after stocking). Four environmental parameters including total of ammonia nitrogen (TAN), nitrate, orthophosphate, total of suspended solid (TSS) and chlorophyll-a were measured. Identification and density of phytoplankton were also performed of each pond. The result showed that productivity of three ponds was 22,893.83 kg/ha to 23,600.61 kg/ha with an average size 12.74 g to 14.35 g. During culture period, the concentration of TAN, TSS and chlorophyll-a tended to increase. Meanwhile, the average of both nitrate and orthophosphate tended to decline. Several phytoplankton identified in this study were in the genus of *Oocystis*, *Chlorella*, *Nannochloropsis*, *Chaetoceros*, *Stephanodiscus*, *Nitzschia*, *Coscinodiscus*, *Cyclotella* and *Ulothrix*. Phytoplankton of the group Chlorophyta is predominance for pond 1 and 2, while pond 3 was dominated by phytoplankton in the group of Diatom/Baccillariophyta.

This study indicated that the present of phytoplankton in

the pond provide natural feed and good environmental condition for shrimp cultivation.

1. INTRODUCTION

Shrimp culture industry in Indonesia has been started at east java province in 1980 (Taw, 2005), and became one of the most important of aquaculture activities until today (Fakhri, et al., 2013). In 2002, whiteleg shrimp *Litopenaeus vannamei* was introduced and has been cultured around Indonesia, particularly in east java. In 2014, Indonesia government target shrimp production approximately 699.000 tons through intensification program (Andri, 2010). Intensive shrimp culture is characterized by the use of qualified and quantified feed and also high stocking density in one pond (Piedrahita, 2003). In intensive shrimp culture, water exchange is the general method to reduce organic material accumulation both from uneaten feed and metabolic waste to improve water quality (Boyd, 2003). However, minimum or zero water exchange strategies have been practiced since 2001 in Indonesia (Taw, 2005).

Minimum or zero water exchange strategies in intensive shrimp culture frequently associated with biofloc technology. Biofloc is defined as macroaggregate consist of microalgae, feces, death organism, bacteria, protist and invertebrate (Taw, 2014). The basic of biofloc technology is the application of nitrogen cycle in fish or shrimp culture in stagnant water by stimulating microbial growth assimilated nitrogen waste converted as natural feed for fish or shrimp (Ekasari, et al., 2015). Galvez, et al. (2015) demonstrated that in biofloc integrated system plankton communities changed with the decreasing of number of Cyanobacteria and increasing of number of Heterokontophyta and Chlorophyta. On the other hand, the number of protozoa decreased while Rotifera and Cladocera increased. The increasing of plankton of communities might stimulate growth of shrimp in biofloc system.

Similar pattern was also demonstrated in the previous study that the pond contained high total suspended solid, TSS (1.413,6 mg/L) and without water exchange had low feed conversion ratio and higher productivity than that of the pond containing lower TSS (411 mg/L) (Fakhri, et al., 2015). The result demonstrated by Fakhri, et al. (2015) also showed that pond without water exchange has better survival rate and more environmental friendly. However, natural food productivity in the previous study is not evaluated yet. The aim of this study was to evaluate phytoplankton communities in intensive shrimp culture in Probolinggo, East Java. Also, we evaluated water qualities including chlorophyll concentration, total of ammonia nitrogen (TAN), TSS, orthophosphate and nitrate

2. MATERIALS AND METHODS

Shrimp pond management

This study was conducted by collecting samples from three different intensive shrimp culture ponds in Probolinggo, East Java. On those three ponds, shrimp has been cultivated for 104 days. Each pond had 0.09 ha in width and 1.3 m in depth. Water exchange level was applied approximately 10 % of the total pond volume per day by replacing apart of volume of the water and filled again at initial volume. At the first month of shrimp culture was not applied water exchange. Moreover, freshwater addition was also practiced to maintain stability of salinity at level 20 ppt. Accumulation of organic material at pond bottom was thrown away routinely by using siphon technic or through outlet as well.

Commercial feed was supplemented for those three ponds during production cycle. Nutritional composition of the commercial feed was 10 % moisture, 30-32 % protein, 5 % lipid, and 4 % crude fiber. Lime was

administered at a dose 7 mg/L to those three ponds to maintain pH stability at the level 7.7-8.0 during production cycle. Molase was also supplemented at the level 3 mg/L for each pond to increase decomposer bacterial growth. Furthermore, probiotic administration (0.5 mg/kg, *Bacillus subtilis*) and vitamin (vitamin C and B, 2 g/kg of feed of each) were practiced as well

Sample collections

Pond water samples were collected two times during study that are in the early and late of culture periods (day 17 and 87). The water samples were collected at 12.00 AM and 02.00 PM for those three ponds. The water samples were collected at 30 m in depth under the surface water for two spot of each pond (Biao, et al., 2009). Water samples collections were held in mineral bottle with 1.5 L in volume. Then, the water samples put into cool box and transferred to the laboratory for further examination including chlorophyll content, total of ammonia nitrogen (TAN), nitrate, orthophosphate, and total of suspended solid (TSS).

Phytoplankton identifications and enumerations

Phytoplankton identification was conducted according to Schrader, et al. (2011) with slight modification. Approximately 50 mL of pond water sample was added with 1% lugol for preservation until microscopic observation. Of each 50 mL of samples, identification and enumeration of phytoplankton were performed by observing 1 mL sample in Sedgewick-Rafter counting chamber under 400x microscope magnification. Natural unit (single cell and colonies) was counted at five field of microscopic observation. Phytoplankton identification was performed according to Prescott (1962) and data base from website: <http://www.algaebase.org/>. Phytoplankton density was reported as individual/liter. Phytoplankton density

counting was calculated using equation according to APHA (1989) as a follow:

$$N = O_i/O_p \times V_r/V_o \times 1/V_s \times n/p$$

where:

N = Number of individual per liter samples

O_i = Width of sedgewick rafter cover (mm²)
1000 mm²

O_p = Width of observation under microscope (mm²) 1 mm²

V_r = Volume of water samples (mL) 10 mL

V_o = Volume of observed water samples (mL)
1 mL

V_s = Volume of filtered water samples (L) 6 L

n = Number of plankton observed under microscope

p = Number of field observed under microscope

Water quality parameters

Chlorophyll content, total of ammonia nitrogen (TAN), nitrate, and orthophosphate were examined using spectrophotometric method for each samples. For TSS, examination was performed using gravimetric method at 100 mL of samples.

Production performance

We also evaluated production performance of those three ponds including survival rate (SR), specific growth rate (SGR) and feed conversion ratio (FCR), total harvest, harvest size per shrimp and productivity per width of pond (hectar). Production performance including SR, SGR and FCR were evaluated according to Brito, et al. (2015).

Data analysis

Phytoplankton density was reported as individual per liter (individu/L). Water quality including TAN, nitrate, orthophosphate and chlorophyll content were presented as milligram per liter (mg/L). Furthermore, SR and SGR were reported as percent (%) and percent body weight per day (%BW/day), respectively.

3. RESULTS

Twelve species of phytoplankton were able to be identified in this study (Table 1). The identified phytoplankton belong to the group of Chlorophyta, Bacillariophyta / Diatom, and Cyanophyta. The dominant phytoplankton differs in those three ponds (Figure 1). Chlorophyta was predominantly phytoplankton in pond 1 and 2, while bacillaryophyta / diatom was the most abundance phytoplankton in pond 3.

Table 1. Identified phytoplankton in intensive whiteleg shrimp *Litopenaeus vannamei* pond.

Groups	Species
Chlorophyta	<i>Oocystis solitaria</i>
	<i>Oocystis naegelii</i>
	<i>Chlorella</i> sp.
	<i>Nannochloropsis</i> sp.
Bacillaryophyta (Diatom)	<i>Chaetoceros densus</i>
	<i>Chaetoceros decipiens</i>
	<i>Stephanodiscus</i> sp.
	<i>Nitzschia</i> sp.
	<i>Coscinodiscus radiatus</i>
	<i>Coscinodiscus asteromphalus</i>
	<i>Cyclotella</i> sp.
Cyanophyta	<i>Ulothrix</i> sp.

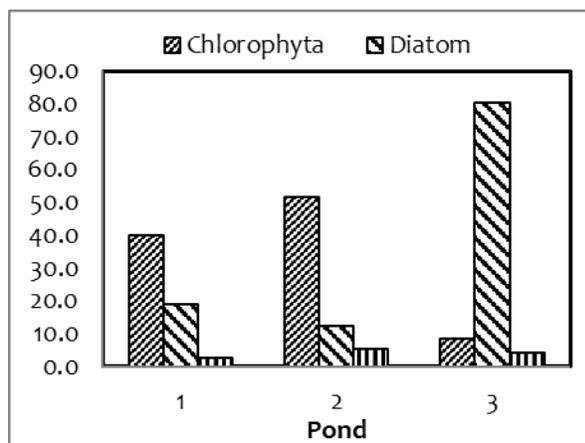


Figure 1. Phytoplankton density (Chlorophyta, Diatom and Cyanophyta) on day 87 after

stocking in each pond of intensive whiteleg shrimp *L. vannamei* pond.

The results of water quality measurement showed different in those three ponds (Figure 2 to 6). The highest total of ammonia nitrogen (TAN), orthophosphate, total suspended solid (TSS) and chlorophyll content were obtained in pond 2 while the highest nitrate was found in pond 1. TAN, TSS and chlorophyll content at day 87 were higher compared to day 17 culture periods. On the other hand, nitrate and orthophosphate on day 87 were lower than day 17.

Shrimp production of those three ponds during cultivation periods were presented in Table 2. The highest shrimp productivity was achieved in pond 1 (23.600,61 kg/ha by the size 14,35 g/shrimp). Pond 2 and 3 had productivity approximately 23.391,21 kg/ha and 22.893,83 kg/ha by the size 12,99 g/shrimp and 12,74 g/shrimp, respectively. Similar result was also obtained for FCR. Pond 1 had lower (1.11) FCR compared pond 2 and 3 (1.12 and 1.23, respectively). Moreover, the highest SR achieved in pond 2 (87,41 %) while pond 1 and 3 were approximately 79,79 % and 81,40 %, respectively.

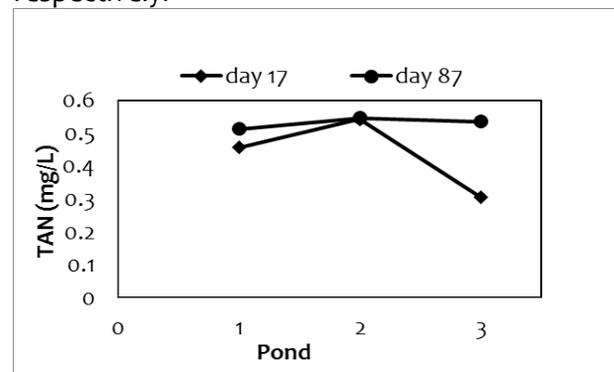
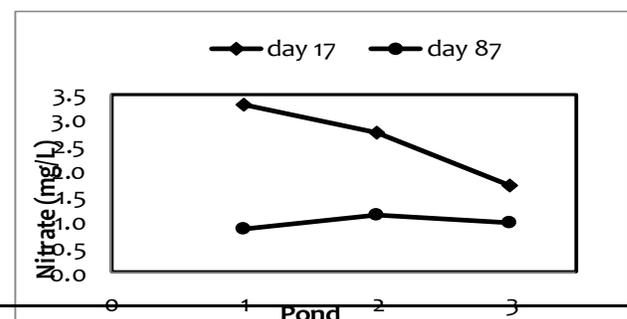


Figure 2. The difference total of ammonia nitrogen in each pond at different time.(Figure 2 until 6 have not been modified as recommended)



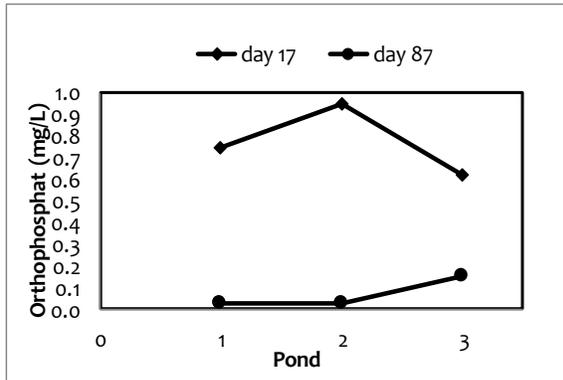


Figure 3. The difference nitrate in each pond at different time

Figure 4. The difference orthophosphate in each pond at different time.

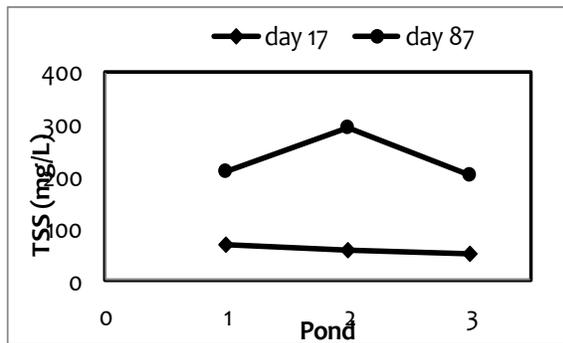


Figure 5. The difference total of suspended solid (TSS) in each pond at different time.

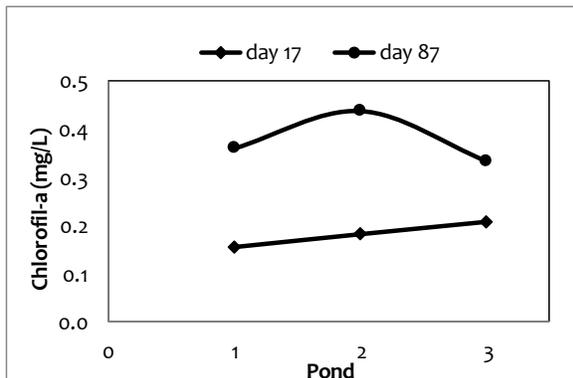


Figure 6. The difference chlorophyll content in each pond at different time.

Table 2. Productivity of those three ponds of intensive whiteleg shrimp *L. vannamei*

Parameters	Pond 1	Pond 2	Pond 3
Total harvest (kg)	2.336,46	2.315,73	2.115,39
Harvest size (g/fish)	14,35	12,99	12,74
Productivities (kg/ha)	23.600,61	23.391,21	22.893,83
Survival rate (%)	79,79	87,41	81,40
Feed conversion ratio	1,11	1,12	1,23
Specific growth rate (% BW/day)	6,64	6,55	6,54

4. DISCUSSION

Environmental factor such as nutrient level was able to affect the abundance of phytoplankton during a shrimp cultivation periods (Shaari, et al., 2011). In this study, total of ammonia nitrogen (TAN), total of suspended solid (TSS), and chlorophyll content were higher at the early period of shrimp cultivation compared to the late period one. Similar results were also obtained in the previous study that TAN and TSS concentration in the shrimp pond increased throughout a shrimp cultivation periods (Fakhri, et al., 2015; Shaari, et al., 2011; Cardozo, et al., 2011). The increasing nutrient level in the shrimp pond may be due to the increasing shrimp biomass and feed supplemented to the pond (Biao, et al., 2009). Throughout cultivation periods, a low nitrate and orthophosphate were also obtained in the previous study (Shaari, et al., 2011; Cardozo, et al., 2011). The decreasing nitrate and orthophosphate concentration in the pond during a shrimp cultivation indicating

that phytoplankton was able to utilize nutrient in the shrimp pond. The source of nutrient in intensive shrimp pond for phytoplankton growth could be from metabolic waste and uneaten feed (Keawtawee, et al., 2012)

The increasing nutrient entering to the pond affect the phytoplankton composition and its density (Case, et al., 2008). The increasing primary productivity (chlorophyll content) during culture periods demonstrated that there were a sufficient nutrient for phytoplankton growth in the pond (Chainark and Boyd, 2010). Chlorophyta predominantly present in the pond 1 and 2 were also obtained in the previous study (Cremen, et al., 2007). Chlorophyta domination in pond 1 and 2 caused pond water color turn to green. Meanwhile, pond 3 dominated by diatom group was also similar to the study conducted by Hadi, et al. (2016). Diatom domination in pond 3 caused pond water color turn to brown. Predominant phytoplankton in a shrimp pond may be affected by water quality such as nitrogen and phosphorus ratio (Cremen, et al., 2007). According to the results of this study, pond productivity probably not only depend on stocking density, feed administration and water quality but also depend on community and biomass of phytoplankton (Keawtawee, et al., 2012). Pond productivity dominated by diatom had lower production compared to pond dominated by chlorophyta. This study indicated that pond productivity was affected by phytoplankton productivity (Keawtawee, et al., 2012).

5. CONCLUSION

Phytoplankton present in each pond was different. Chlorophyta was the predominant phytoplankton in pond 1 and 2 while pond 3 was dominated by diatom. Water quality including TAN, TSS and chlorophyll content tended to increase while nitrate and orthophosphate tended to decrease. This study indicated that the present of phytoplankton supported the availability of natural feed and improved water quality for shrimp culture.

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