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Dynamic ratio correlation of N:P on the abundance of Blue-green algae in an intensive system in a white shrimp (*Litopenaeus vannamei*) pond

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Abstract. Phytoplankton in an intensive system of white shrimp cultivation is useful as an indicator of the water quality. Blue-green algae are a group of phytoplankton that has an important role, although there are several diverse species. Blue-green algae are an abundant primary producer and can be found a lot both in freshwater and in the sea. Blue-green algae are also a potential bio-indicator to designate how good or bad the water condition is. This research aimed to determine the correlation of the ratio dynamic of N:P toward the abundance of Blue-green algae within the system of a Vaname (*Litopenaeus vannamei*) shrimp pond. The main parameters observed were ammonium, nitric, nitrate, phosphate and the abundance of blue-green algae, as well as the supporting parameters of pH and the clarity of the intensive system of the Vaname shrimp pond in Banyuwangi. Based on the data analysis and study of the dynamic ratio correlation of N:P toward the abundance of Blue-green algae, it can be concluded that ratio of N:P influences the level composition of phytoplankton in water cultivation. The difference in the levels of ammonium, nitric and phosphate influence the abundance of blue-green algae in the water. A high level of ammonium and low level of nitric causes a high level of abundance of Blue-green algae.

1. Introduction

Vaname shrimp can be cultivated using the intensive system method. One of the characteristics of intensive system cultivation is the high density involved. High density affects the feed needed, the space for movement, and the oxygen level, which will influence the quality of the caring media, growth and the viability of the Vaname shrimp [1].

An excess of feed will increase the organic waste in the water. Leftover feed contains the necessary nutrient for plants in order to survive and grow, including phytoplankton. The growth of phytoplankton in the water is influenced by the availability of nitrogen and phosphorus. Nitrogen and phosphorus are more needed than carbon, hydrogen and oxygen, because nitrogen and phosphorus can be utilized by phytoplankton in small quantities [2].

Nitrogen and phosphorus levels in nature are limited, so this affects the growth of the phytoplankton. The usage of organic and inorganic fertilizer in cultivation ponds is one way to develop water fertility. Fertile water is indicated by the abundance of phytoplankton [3]. The usage of fertilizer with a certain ratio of N:P can determine the abundance of phytoplankton in an aquatic system. A ratio value for N:P



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of 10:1 or less can bring up the domination of Blue-green algae. This potentially happens because Blue-green algae can survive in extreme conditions, including conditions where there is a low level of nitrate. The ideal ratio for N:P within a cultivation pond is 16:1 [4].

Blue-green algae often experience a population explosion (blooming algae). The phytoplankton explosion of population is caused by excess nutrients in the water. Too many nutrients in the water is known as eutrophication. Eutrophication is caused by natural processes or by contamination due to too many nutrients being dissolved in the water, or it can be from the waste stream that is being discarded into the water or contaminating the cultivation water source. Water with excess nutrients causes the phytoplankton to grow too fast or in an uncontrollable manner, including Blue-green algae [5].

Blue-green algae are also known to be able to produce toxins, which lead to disrupted productivity in the Vaname shrimp. Blue-green algae can also be a competitor with other plankton. Vaname shrimp cultivation ponds that contain a lot of harmful Blue-green algae will experience productivity disruption. This will lead to the potential death of the cultivation organisms. Blue-green algae often dominate water when the population blooms, which occurs commonly in harmful genus' [5].

The blooming population of Blue green algae is mostly avoided by vaname shrimp cultivators. Therefore this research into the "Correlation of Dynamic Ratio N:P Towards the Abundance of Blue-green algae in The Intensive System of white Shrimp (*Litopenaeus vannamei*)" needed to be done in order to discover the influence of the dynamic ratio of N:P toward the dynamic of Blue-green algae in relation to the nature of white shrimp pond cultivation.

2. Methodology

2.1 Tools and materials

The tools used in this research included pH paper, a secchi disk, a plankton net, bailer, hand counter, hemocytometer, drop pipette, microscope, object glass, cover glass, test kit, and sample bottle. The materials used in the research included sample water from the intensive system pond and lugol related to the deactivated plankton movement.

2.2 Research methods

The method used in this research was a survey. We prepared the necessary tools and materials, i.e. the pH paper, secchi disk, plankton net, bailer, hand counter, hemocytometer, drop pipette, microscope, object glass, cover glass, test kit, sample bottle and lugol. The water samples were taken at 3 stations, which consisted of 4 points at the pond plots in each corner of the intensive system to improve the data clarification. The samples were taken using a plankton net, and we then calculated the density. The samples had to be directly brought to the laboratory in order to be observed and analyzed. The samples were observed in 100 and 400 times of enlargement under a binocular microscope with direct calculation using hemocytometer. The samples needed to be directly observed in order to maintain the quality of the phytoplankton. The samples observed were ammonium, nitrite, nitrate and phosphate taken in the morning at 5 AM, while the water samples to observe the plankton and water quality were taken in the afternoon at 4 PM.

2.3 Calculation and observation of diatom

The diatom observation consisted of identifying abundance in the pond. The observation was applied in 3 plots of the pond, and taken at 4 points in the corners for each plot. The samples were taken using a plankton net with a mesh size of 20 microns. A plankton net with a mesh size of 20 microns is able to filter the phytoplankton class of diatom, as well as enabling the water to come out through the micro holes of the plankton net.

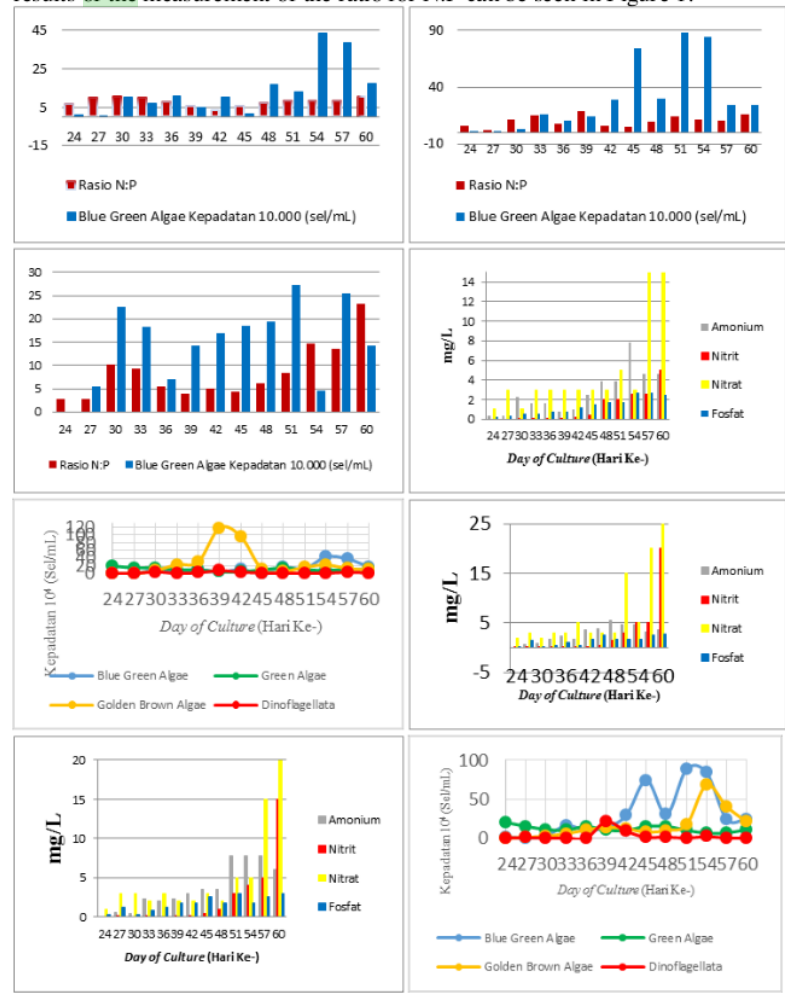
The method used in the identification and observation of the phytoplankton was a direct calculation using a hemocytometer. It was applied by taking a 1 ml water sample from the sample bottle, and then covering it with a glass cover. The observation was done by identifying the phytoplankton and calculating its density in the hemocytometer. The sum of the phytoplankton was calculated using the method suitable for the particular size of plankton. The samples had to be directly calculated in order

to maintain the quality of the observed phytoplankton, to make them easier to identify and to improve the accuracy of the density calculation when observing them under the microscope.

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3. Results and discussion

3.1 Results

The results of this research used the data on the ratio of N:P taken from the accumulation data of ammonium, nitrite, nitrate to be N and the phosphor in order to getting the ratio of N:P itself. The results of the measurement of the ratio for N:P can be seen in Figure 1.



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Figure 1. 1) Comparison of the N:P ratio with Blue-green algae density in plot 1; 2) Comparison of the N:P ratio with Blue-green algae density in plot 4; 3) Comparison of the N:P ratio with the density of the Blue-green algae in plot 7; 4) the dynamics of ammonium, nitrite, nitrate, and phosphate (Plot 1); 5) the dynamics of ammonium, nitrite, nitrate, and phosphate (Plot 4); 6) the dynamics of ammonium, nitrite, nitrate, and phosphate (Plot 7); 7) total phytoplankton dynamics (Plot 1); 8) Charts of the total phytoplankton dynamics (Plots 4) and 9) total phytoplankton dynamics (Plot 7).

For the results of the N:P ratio in plot 1 on day 24 of the Vaname shrimp cultivation, there was an increase in day 27, and then less of an increase on day 30. Furthermore, the ratio of N:P was gradually decreasing until day 42. The ratio and dynamic of N:P in plot 1 increased until day 60 of the cultivation. The lowest ratio for N:P in plot 1 was on day 42, while the highest value was on day 60.

The results of the N:P ratio in plot 4 had more fluctuating values. From day 24 to day 27 of cultivation, the ratio decreased and then increased until day 33. N:P decreased at day 36 and then increased until day 39. The decrease occurred again at day 45, and then increased on day 51. The N:P ratio decreased on day 57 and increased on day 60. The dynamics of the ratio of N:P in plot 4 were the most unstable compared to plots 1 and 7. The lowest ratio for N:P in plot 4 was on day 27, while the highest ratio was on day 39.

The results of the ratio in plot 7 decreased on day 27 and increased on day 30. The ratio of N:P decreased until day 39, and then increased until day 45. N:P increased until day 54 and then decreased at day 57, then increased at day 60. The lowest ratio for N:P in plot 7 was on day 27, while the highest value was on day 60.

The lowest ammonium level in plot 1 was on days 24 and 27, while the highest ammonium level was on day 54. The ammonium level increased on day 54 and then decreased on day 57. The nitrite level remained stable between the 24th to 45th day, then increased on day 48. It then remained stable until day 57. The nitrite level doubly increased on day 60th. The lowest nitrite level was on day 24 and 30, while highest level was on days 57 and 30. Nitrate underwent a dynamic condition from day 24 to day 33, and then remained stable until day 4^h, increased on day 51 and significantly increased on days 57 and 60^l. Meanwhile, phosphate remained stable. The level of phosphate did not decrease until day 57 but then decreased on day 60. The highest level was on day 57.

The lowest ammonium level in plot 4 was on day 24, which then increased until day 36 and then decreased on day 39. The level of ammonium achieved the highest level on day 48, and then decreased until day 57. It then slightly increased on day 60. The level of nitrite remained stable until day 45, and then increased until day 54. The lower level of nitrite was on day 45, and the highest level was on day 60. Meanwhile, nitrate was in a dynamic condition from day 24 to day 48, and then significantly increased on day 51 and decreased on day 54. The nitrate level increased again on day 57, and achieved the highest level on day 60. The lowest level of phosphate was on day 24. The phosphate was in a dynamic condition until day 48, and then remained stable until day 54. The phosphate level increased until day 60. The highest level of phosphate was on day 60.

The lowest ammonium level in plot 7 was on day 24. The level of ammonium underwent a dynamic condition until day 45, and then became stable on day 48. It then increased until the highest level on day 51 and then became stable until day 57. The level of ammonium decreased on day 60. Meanwhile, the nitrite achieved the lowest level on days 24, 30 and 36. The nitrite level tended to remain stable until day 45, and then increased until day 60. The highest level of nitrite was on day 60 and lowest level was on day 24. Moreover, the nitrate didn't undergo a substantial dynamic condition until day 57. The nitrate level rapidly increased and achieved the highest level on day 60. Meanwhile, the lowest level of phosphate was on day 24^l. The phosphate level didn't undergo any substantial dynamic situations. The highest levels of phosphate were on days 51 and 60.

The lowest density of Blue-green algae in plot 1 was on day 27. The Blue-green algae decreased from day 24 to day 27 and then increased on day 30. The Blue-green algae decreased again on day 33 and then increased on day 3⁹. Once more, the Blue green algae decreased until day 51 and then significantly increased until its highest density on day 54. Then the Blue-green algae decreased until day 60.

The lowest density of Blue-green algae in plot 4 was on day 27. The Blue-green algae decreased on day 36 and then increased until day 45. The Blue-green algae decreased again on day 48th, and then increased until its highest density on day 51. Once more, the Blue-green algae decreased until day 57 and then slightly increased on day 60.

The Blue-green algae in plot seven couldn't be discovered on day 24. The Blue-green algae started to develop from day 27 through to day 33. The Blue-green algae decreased on day 36 and increased on day 39. From day 39 until day 51, the Blue-green algae was growing. The Blue-green algae then decreased on day 54 and increased again on day 57. Afterward, it decreased again on day 60. The highest density of Blue-green algae was on day 5¹.

3.2 Discussion

The Diatom ratio of N:P in the intensive system in plot 1 was about 3.7 – 10.4. This number can be categorized as low regarding the activities of fish cultivation, because the highest number for N:P was 10. This phenomena can be viewed according to the statement that [6] mentioned, where the threshold on the contents of the N:P ratio in adequate cultivation water was 16:1. The ratio for N:P in plot 4 was about 2.3 – 18.3, which met the standard for cultivation. Meanwhile plot 7 showed a ratio of about 2.6 – 23.2, in which the upper point surpasses the standard of cultivation but remains normal because the N:P ratio of 20:1 will support the growth of diatoms. All of the plots showed that the ratio was dynamic, however plots 4 and 7 are the most ideal conditions according to [4]. This is because the most ideal ratio for N:P in aquatic cultivation is 16:1. The level of N:P is influenced by organic compounds. Organic compounds in the water come from the microorganisms' bodies, plants, and the result of the organisms' metabolism processes [7]. The organic compounds in water are decomposed by bacteria into N and P, and they can then be utilized by the organisms. The enhancement of the nitrogen level in the water can also be influenced by the derivation of total plankton density [8].

The increase in the ammonium level in plot 1 occurred on days 30, 42, 45, 48 and 54. Meanwhile, the level in plot 4 increased on day 27, 30, 33, 36, 42, 45, 48 and 60. The plot 7 increases occurred on day 27, 33, 39, 42, 45 and 51. Significant increases in plot 1 were on day 54 to 3.9 mg/L, in plot 4 on day 42 to 1.9 mg/L and in plot 7 on day 51 to 4.3 mg/L. The deprivation of the ammonium in plot 1 occurred on day 33, 39 and 57; in plot 4 on day 39, 51 and 57 and in plot 7 on day 30, 36 and 60. A significant deprivation in plot 1 occurred on day 57 down to 3.2 mg/L, in plot 4 on day 57 down to 1.4 mg/L and in plot 7 on day 60 down to 1.8 mg/L.

According to [9], the enhancement and deprivation of ammonium is caused by the factor of the bacteria's nitrification process. The enhancement of ammonium is caused when the ammonium can't be oxidized into nitrite. This possibly happens because the Nitrosomonas bacteria are not properly working to recast the ammonium. Ammonium deprivation is caused by the Nitrosomonas working properly to recast the ammonium. Dead organisms that cause an accumulation of organic compounds is also the cause of high-level ammonium in the water.

Nitrite enhancement in plot 1 occurred on day 30, 33, 42, 45, 48, 54 and 60. Enhancement in plot 4 occurred on day 27, 42, 45, 48, 51, 54 and 60. The enhancement in plot 7 occurred on day 27, 33, 39, 42, 45, 48, 51, 54, 57 and 60. A significant deprivation in plot 1 occurred on day 60 down to 2.5 mg/L; in plot 4 on day 60 down to 15 mg/L and in plot 7 on day 60¹ down to 10 mg/L. The nitrite deprivation in plot 1 occurred on day 33, 39 and 57; in plot 4 on day 39, 51 and 57th; and in plot 7 on day 30, 36 and 60. Significant deprivation occurred in plot 4 on day 30 down to be 1.4 mg/L and in plot 7 on day 30 and 36 down to 1.8 mg/L.

The enhancement of nitrate in plot 1 occurred on day 27, 33, 51 and 57. Meanwhile, in plot 4, the increases occurred on day 27, 33, 39, 51, 57 and 60. Furthermore, in plot 7, the increases occurred on day 27, 36, 45, 51, 57 and 60. A significant increase in plot 1 occurred on day 57 up to 12 mg/L; in plot 4 on day 51 and on 60 to 12 mg/L and 15 mg/L respectively and in plot 7, the increase occurred on day 60 to 45 mg/L. The deprivation of nitrate in plot 1 occurred on day 30 and 54. In plot 4, the decreases occurred on day 30, 42 and 54. In plot 7, the deprivation occurred on day 33, 39 and 48. The significant deprivation of nitrate only occurred in plot 4 on day 54 down to 10 mg/L.

Nitrate is the final product of the biochemistry oxidization process in the water. Nitrate concentrations in an aquatic system are controlled in the nitrification process. Nitrate comes from fertilizer residual, leftover feed and free nitrogen binding from the air by microorganisms, as well

where streams enter the sea. The nitrate formation will be smooth when the bacteria work to recast the nitrite [10].

The enhancement of phosphate in plot 1 occurred on day 27, 30, 36, 42, 45, 48 and 54. Meanwhile, in plot 4, it occurred on day 27, 33, 36, 42, 45, 57 and 60. Then in plot 7, it occurred on day 27, 33, 36, 39, 45, 51, 57¹ and 60¹. The significant enhancement in plot 1 occurred on day 54 to 1 mg/L; in plot 4 on day 42 to 1.35 mg/L and in plot 7 on day 51 to 1.75 mg/L. The phosphate deprivation in plot 1 occurred on day 60; in plot 4 on day 30, 39 and 48¹ and in plot 7 on day 30, 48 and 54. Significant deprivation occurred in plot 4 on day 30^h to 1.2 mg/L and in plot 7 on day 57 to 1.75 mg/L.

Phosphor is in the dissolved inorganic form (orthophosphate), dissolved organic and phosphate particles. [11] mentioned that normally, phytoplanktons are able to directly assimilate dissolved inorganic material, and sometimes use dissolved organic phosphor. The phosphor is applied in energy transfer in the phytoplankton's cells from ADP to ATP.

The identification of the Blue-green algae during the research resulted in discovering *Anabaena* sp., *Chroococcus* sp., *Gomphosphaeria* sp., *Microcystis* sp., *Oscillatoria* sp., and *Spirulina* sp. The quantity of identifying the phytoplankton based on genus resulted in one class of Cyanophyceae.

The density enhancement of Blue-green algae in plot 1 occurred on day 30, 36, 42, 48 and 54. The most significant increase in plot 1 occurred on day 54 up to 30.5×10^4 cells/ml. In plot 4, the increase in density occurred on day 30, 33, 39, 42, 45, 51 and 60, with the most significant increase on day 51^h up to 58.25×10^4 cells/ml. Meanwhile, in plot 7 the density increase occurred on day 27, 30, 39, 42, 45, 48, 51 and 57. The most significant increase in plot 7 occurred on day 57 up to 21×10^4 cells/ml.

The decrease in the Blue-green algae density in plot 1 occurred on day 27, 33, 39¹, 45, 51, 57 and 60. The most significant decrease in plot 1 occurred on day 60 down to 21×10^4 cells/ml. Meanwhile, plot 4 underwent a density decrease on day 27, 36, 48, 54 and 57, with the most significant decrease occurring on day 57 down to 60×10^4 cells/ml. Furthermore, in plot 7, the density decrease occurred on day 33, 36, 54 and 60. The most significant decrease in plot 7 occurred on day 54 down to 22.75×10^4 cells/ml.

[12] mentioned that the enhancement of nitrate can bring up the domination of diatoms because diatoms need nitrate for cell division. Blue-green algae are more suitable to be in a condition where there is a high level of ammonium and a low level of nitrate. Nitrate is the substance that is used by the diatoms, so if the nitrate is abundant, then the diatoms will be abundant as well. The condition of the limited level of nitrate means that there will be less domination of diatoms because there is less nitrate to be utilized. [5] mentioned that Blue green algae have an important role as a producer of nitrogen in water; Blue green algae are able to survive in conditions where there is a low level of nitrate.

4. Conclusion

Based on the results of the data analysis and the discussion of the dynamic relationship of the N: P ratio on the abundance of green blue algae, it can be concluded that the value of the N: P ratio affects the composition of the phytoplankton classes in the aquaculture water. The N:P ratio affects the abundance of phytoplankton, but the levels and dynamics of ammonium, nitrite, nitrate and phosphate specifically influence the abundance of Blue green algae. High ammonium conditions and low nitrate can cause an abundance of Blue green algae to increase. Water quality parameters are a supporting factor that also affect the life of Blue Green Algae.

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