Dynamic ratio correlation of N:P toward phytoplankton explosions in intensive systems of white shrimp pond

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Abstract. Phytoplanktons are a supporting microorganism within the activities of vannamei shrimp cultivation. Phytoplankton are useful to control the color and clarity of the water, increasing the dissolved oxygen (DO), controlling the growth of weeds in the bottom of the pond, and absorbing the abundance of organic compounds in the water such as ammonium, nitric and nitrate. This research aimed to find out the influence of the ratio dynamic of N:P toward the abundance of phytoplankton in an intensive system shrimp pond. The main parameters observed were ammonium, nitric, nitrate and phosphate, as well as the abundance of phytoplankton. The study was being held in the laboratory of PT Surya Windu Kartika Banyuwangi, and measurements were also taken of the supporting parameters of pH, clarity, and salinity. The results of the data analysis and study revealed that the dynamic composition of ammonium, nitric, nitrate and phosphate influences the dynamic composition of the phytoplankton in the water. The increasing nitric levels affect and increase the composition of Bacillariophyceae and decrease the abundant composition of Cyanophyceae. An increase in the level of ammonium in the water causes an increase in the abundant composition of Cyanophyceae and a decrease in the abundant composition of Bacillariophyceae, and dynamic PO4 influences the dynamic abundant composition of phytoplankton classified as Bacillariophyceae and Cyanophyceae.

1. Introduction
Phytoplanktons are a supporting microorganism in the context of the activities of Vannamei shrimp cultivation. Phytoplanktons are important as natural feed due to its characteristic as the foundation of the food chain in the aquatic system [1]. In the activities of intensive shrimp cultivation, phytoplankton are also important when it comes to controlling water color and turbidity, as well as increasing the level of dissolved oxygen (DO) in the water, restraining the growth of the moss in the bottom of the pond, and also absorbing harmful organic compounds like ammonia, nitrite and nitrate, which are necessary nutrients for growing phytoplankton [2].

The diversity and abundance of phytoplankton is useful as stability parameter regarding the nature of the water; a high diversity of phytoplankton species and the prevalence of a high number of each species means that the water quality is in a range that is suitable for the growth of cultivation organisms [2]. An excess density of plankton shows there to be a discrepancy in the aquatic
condition; an increase of plankton density in daylight can cause a high saturation level of oxygen up to 250% and this generate eboligas in gills; this causes death in shrimp. At night, there will be a lack of oxygen because of the high respiration process of the phytoplankton [3].

The problem formulated in this research was: ‘Does the influence of the ratio of dynamic N:P have an effect on the abundance of phytoplankton in an intensive system shrimp pond?’ The aim of this research was to discover the influence of the ratio of dynamic N:P on the abundance of phytoplankton in an intensive system in a shrimp pond.

The benefit of this research was that it has been able to provide information related to the influence of the N:P conversion ratio in the context of affecting the diversity and abundance of phytoplankton in one of the intensive system shrimp ponds. Therefore the potential of various species of phytoplankton can be predicted.

2. Methodology
2.1 Tools and materials
The tools used in this research were as follows: a DO meter, spectrophotometer, secchi disk, refractometer, pH pen, plankton net, bucket, ropes, hemocytometer, drop pipette, microscope, glass object, glass cover, sample bottle, burette, static, clamp, a 100 ml measurement glass, a 300 ml Erlenmeyer glass, a Erlenmeyer 500 ml glass, a volume pipette of 2 ml and a filler/bulb. The materials being used in this research were: a water sample from the intensive vannamei shrimp pond in PT. Surya Windu Kartika in Banyuwangi Regency, a PP indicator, 1% aquadest, ammonia test kit, the reagent of water hardness, nitrate, nitrate and phosphate.

2.2 Research methods
The research methodology used was a survey, which is a research method that aims to describe both existing and ongoing phenomena. The research was applied without manipulating or changing the free variables, in order to describe the real condition [4]. The research plan was done by the measurement of the water quality at 3 sample points to observe the water quality and 12 sample points to observe the abundance of phytoplankton. A sample is taken at the 12 sample points in the intensive shrimp pond of PT. Surya Windu Kartika. Samples to observe the water quality of DO, temperature, pH, salinity, water level, abundance and the diversity of the plankton were taken twice a day at 4 PM, depending on the availability of sunlight. The measurement of the water quality parameters of ammonium, ammonia, nitrite, nitrate, and phosphate were taken once a day at 7 AM. The sample plankton was taken using a plankton net, and were then observed under a microscope using the direct measurement method using a Sedgwick Rafter at 100 to 400 times enlargement, twice repeated. The water quality observation of ammonium, ammonia, nitrite, nitrate and phosphate was directly performed at 3 sample points using a sample bottle, and they were then observed using the spectrophotometer adjusted to the water. The research preparation stage was applied to prepare the necessary tools and materials, i.e. pH paper, secchi disk, plankton net, bailer, hand counter, hemocytometer, dropping pipette, microscope, object glass, cover glass, test kit, sample bottle, and lugol. The water samples, as the material to observe the diatom abundance, were taken at 3 stations, which were the pond plots with 4 points in each corner of the intensive system for data clarification. The samples were taken with filtered water using a plankton net, to calculate the density. The samples had to be directly brought to the laboratory to be observed and analyzed. The samples were observed at 100 and 400 times the enlargement under a binocular microscope with the direct calculation done using a hemocytometer. The samples needed to be directly observed to maintain the quality of the phytoplankton. The samples to observe the ammonium, nitrite, nitrate, and phosphate were 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.
3. Results and discussion

3.1 Results
The results of this research consist of the main data and supporting data. The main data is the numbers involved in the ratio of N:P and the abundance of phytoplankton. The supporting data was water quality, including the acidity level (pH) and clarity.

3.1.1 Grade of N: P ratio
The data for the ratio of N:P was obtained from the data of the total nitrogen in the ammonia, ammonium, nitrate, and nitrite compared to phosphate; this then became the grade of the ratio of the dynamic N:P. The data used was the result calibration done every 3 days. The results of the ratio of the dynamic N:P can be seen in the following graphic.

The measurement result of the N:P ratio movement in plot 1 showed a stable level of nitrate at DOC 33-48, and a high fluctuation at DOC 15 by 15 mg/L. It then decreased again the next day. The dynamic condition of ammonium occurred in plot 1 with an adequate fluctuation and there was a frequent level of contact between ammonium and nitrate at DOC 27-30, 30-34, 45-48 and 55-58. In plot 1, there was a high level of ammonium and nitrate at DOC 31, 49, 52, and 55. The phosphate level in plot 1 was generally stable and increased at DOC 46-61.

The measurement result of the nitrite and phosphate in plot 4 showed a highly dynamic situation at DOC 51 until the end of the observation session, with a nitrite increase occurring in DOC 51, 57 and 60 with the highest level of nitrate being up by 25 mg/L at DOC 60. The level of nitrite as an intermediate nutrient significantly increased at DOC 60 of 20 mg/L. The other nutrients in plot 4 didn’t occur significantly or in a dynamic condition. The nitrite level in plot 4 significantly increased to 20 mg/L, which is above the threshold of 1.0 mg/L. This can be dangerous to live organisms in the aquatic system [5].

The observation result of the dynamic situation of nitrate and phosphate in plot 7 showed that there was no significant dynamic situation from the initial observation until DOC 48. The increased and decreased levels of ammonia, nitrite, nitrate, and phosphate in DOC 24-48 were about 0.1 – 3.5 mg/L. The dynamic level of nitrate significantly increased at DOC 57 and 60 to be the highest at 60 ml/L, while the other nutrients such as ammonia, nitrite, nitrate, and phosphate did not significantly increase; they only went up about 5-15 mg/L in DOC 51-60.

3.1.2 Plankton abundance
We discovered phytoplankton in all of the observation stations, consisting of 19 genus from 4 classes. The 4 classes were Cyanophyceae (6 genus), Bacillariophyceae (9 genus), Chlorophyceae (3 genus) and Dinophyceae (1 genus). All of the discovered genus could be seen in Table 4. The abundance of phytoplankton in this research consists of the density level in relation to the total number of cells in the water and the percentage composition of phytoplankton in the water. The data was taken by observation every 3 days.

The observation results of the phytoplankton abundance in plot 1 were generally dominated by phytoplankton of the Bacillariophyceae class. The increased dynamic situation of Bacillariophyceae started at DOC 30 and gradually increased until it achieved the highest density at DOC 39. It then decreased with no significant dynamic situation present until the end of the observation session. Significantly increased phytoplankton also occurring in the class of Cyanophyceae, which increased significantly at DOC 54. The phytoplankton of Chlorophyceae and Dinoflagellate didn't have a significant condition, but their existence in plot 1 was always consistent. This shows the stability condition of the water in plot 1. There was a significant dynamic situation at the end of the observation session at DOC 54.

The observation results of the phytoplankton abundance in plot 4 showed there to be highly dynamic conditions between the phytoplankton of class Cyanophyceae. The phytoplankton of class Cyanophyceae started to have a dynamic condition at DOC 42 and achieved the highest density at DOC 51; this shows the dynamic condition of Cyanophyceae in decreased aquatic condition. In plot 4,
the dynamic condition of the phytoplankton of class Bacillariophyceae was generally stable, with the highest density at day 54. In general, the highest abundance of phytoplankton in plot 4 occurred at DOC 54, with the domination of class Cyanophyceae and Bacillariophyceae. The observation results of the phytoplankton abundance in plot 7 showed that the highest density was of the phytoplankton of class Bacillariophyceae at DOC 39. Besides that, the phytoplankton of class Bacillariophyceae significantly fluctuated in condition from DOC 33 to 39, with the fluctuations increasing and decreasing until the end of the observation session. The phytoplankton class of Blue-green algae did not occur in a dynamic condition and were more stable compared to Bacillariophyceae; the highest growth of Cyanophyceae occurred at DOC. In plot 7, the phytoplankton of class dinoflagellate highly increased at DOC 54.
Figure 1. (1) N: P ratio of plot 1 during research, (2) Ammonium, Nitrite, Nitrate and Phosphate Plots 1 during the study, (3) N: P ratio of plot 4 during research, (4) Graphs of Ammonium, Nitrite, Nitrate and Phosphate Plot 4 During Research, (5) N: P Ratio 7 during Research, (6) Ammonium, Nitrite, Nitrate and Phosphate Plot 7 During Research, (7) Phytoplankton Plot 1 During Research, (8) Phytoplankton Plot 4 During Research, (9) Phytoplankton Plot 7 During Research

3.2 Discussion

The diatom nutrients in the aquatic system are important for the existing life of microorganisms, in particular in aquatic cultivation where the existence of microorganisms like phytoplankton and bacteria is important in relation to the success of cultivation activities. The most influencing nutrients in the lives of phytoplankton are divided into macro elements and microelements, which is determined based on the needs of the phytoplankton towards the elements. Among the various nutrients, there are 2 important nutrients that play the role of being a divider factor in the lives of phytoplankton i.e. N and P [6].

N and P are important nutrients in the viability of phytoplankton, both in water in general and in cultivation water. The two nutrients play a role as the media of energy transfer in the cells and also as an essential element in the formulation of the phytoplankton cells [7]. The influence of N and P is not only limited to the ratio dynamic N:P in the water, but it is also a dynamic form of each element, like ammonium and nitrate in water and the dynamic condition of phosphate and nitrite.

Nitrate and ammonia are the most dominant form of the N elements that influence the dynamic condition of the phytoplankton in the water. This is because nitrate is formed of N, which is directly absorbed by the phytoplankton. Ammonia is the initial material in the nitrification process in cultivation water, and it stimulates the growth of certain phytoplankton. According to previous studies [6], the increase in the content of nitrate in the aquatic system is able to increase the domination of Bacillariophyceae and decrease the domination of Cyanophyceae. Meanwhile, an increase of the ammonia in water is potentially decreasing the population of Bacillariophyceae and increasing the density of Cyanophyceae.

Based on the research, the observation data from 3 observation stations (plot 1 DOC 33 and 51; plot 4 DOC 27, 33, 51 and 57; and plot 7 DOC 27, 36 and 51) showed there to have been an enhancement of the nitrate level in the cultivation water affected by the increase in the domination of Bacillariophyceae and a decrease in the phytoplankton class of Cyanophyceae. Besides that, the decrease in the nitrate level caused a decrease in the Bacillariophyceae domination in the water. It aligns with the statement in a previous study by [6] on the enhancement level of NO3, which is able to increase the abundance of Bacillariophyceae in an aquatic system.

Based on the results of the data analysis of the correlation of the decreased dynamic condition and dynamic abundance of phytoplankton, there are 4 conditions that lead to phytoplankton dynamic change in the water. This includes crashing between NO3 and NH3 causing a dynamic change in the Bacillariophyceae asin plot 1 at DOC 30 and 54; plot 4 at DOC 30; and plot 7 at DOC 33 and 39. The enhancement of PO4 influences the dynamic condition between the nitrate level and the phytoplankton like in plot 1 at DOC 54 and in plot 7 at DOC 39. This is because the enhancement of the PO4 levels in the water is affected by the decrease in the ratio of N:P as supported by the Bacillariophyceae growth. The significant enhancement of the NO2 levels influences the dynamic of the nitrate in relation to the dynamic condition of the phytoplankton in the water, as in plot 4 on DOC 54 and 16 and plot 7 on DOC 57 and 60. This happened because the NO2 at a level of 1.0 ppm was toxic toward aquatic organisms, including phytoplankton [5].

Ammonia is an organic compound that influences the dynamic condition of the phytoplankton in water, in particular, cultivation water. This is because the existence of ammonia in the water, in general, is increasing along with the increase in the age of the cultivation. According to the previous studies [6], the enhancement of the ammonia level in the water is able to decrease Bacillariophyceae
and increase the domination of Cyanophyceae. This is suitable for the observation results in plot 1 at DOC 30, 42, 48 and 54, plot 4 at DOC 30, 33, 42 and 45 and plot 1 DOC 39, 42, 48 and 51. This is where the ammonia level increased and was followed by the domination and enhancement of Blue-green algae. The decreased level of ammonia in the water also showed there to be a decrease in the domination of phytoplankton class of Cyanophyceae. It is suitable as seen in the observation data in plot 1 at DOC 33, 39 and 57, plot 4 at DOC 57 and plot 7 at DOC 36 and 60.

There are several conditions that lead to an exception in the correlation of the ammonium and phytoplankton dynamic condition, such as an intensive fluctuating condition of the nitrate level in the water. This influences the domination of Cyanophyceae in the water. It is possible that this happens because high levels of nitrate may prompt Cyanophycean growth, like in plot 4 on DOC 27 and 60. Meanwhile, in conditions where there are lower nitrate levels or there is a crash of nutrients between ammonium and nitrate, this can push Cyanophycean growth. The PO₄ dynamic condition in the water is also important in relation to phytoplankton domination in the water as in plot 4 DOC 39 and 48. Cyanophyceae domination is more affected by the availability of PO₄ than ammonia in that aquatic system. As in the previous study [8], water condition with PO₄ levels higher than 1.10 ppm can cause the phytoplankton domination of Cyanophyceae.

A condition where the water nutrients are not consumed by phytoplankton can be seen in plot 1 DOC 39, 57 and 60, plot 4 DOC 39, 57 and 60 and plot 7 DOC 60. In this condition, nitrate as the dividing factor is abundant but the phytoplankton, in general, is decreased or increased. The phytoplankton class of dinoflagellate is a bioindicator of discrepancies in aquatic systems. Therefore, the nutrient concentration in water is not fully absorbed by the phytoplankton. Generally, nutrients cannot be absorbed because of several factors, such as a low level of DO, a high pH in the water, a high level of nitrite, a high level of ammonia and also significant changes in the water temperature [8].

4. Conclusion
Based on the results of the data analysis and the discussion related to the correlation of the ratio of N:P towards the abundance of phytoplankton, it can be concluded that the value of the ratio of N:P influences the composition of the phytoplankton class in the water of Vannamei shrimp cultivation. The dynamic composition of ammonia, nitrite, nitrate, and phosphate influences the dynamic composition of phytoplankton in aquatic systems. The enhancement of the nitrate level increases Bacillariophyceae and decreases the abundant composition of Cyanophyceae. The enhancement level of ammonia in the water can increase the abundant composition of Cyanophyceae and decrease the abundant composition of Bacillariophyceae. The dynamic condition of PO₄ influences the dynamic composition of the phytoplankton classes of Bacillariophyceae and Cyanophyceae.

5. References
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