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Total Lipid and Morphology Microalgae *Skeletonema costatum on* Nitrogen Nutrition Physiological Stress

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ABSTRACT

Biodiesel is an alkyl ester compounds produced by alcoholysis of triglycerides and methanol or ethanol with the aid of an alkaline catalyst. Indonesia is rich in marine resources, one of which is a microalgae. Microalgae can make alternative as a raw material for biodiesel. This study aimed to influence physiological stress nutrient nitrogen to total lipid content of microalgae *Skeletonema costatum*. The method used is the intermediate-scale culture 100 L. Harvesting microalgae as a candidate biodiesel is done is done at the end of the exponential phase. Parameters measured were total lipid content using the Soxhlet. Total lipids were analyzed using ANOVA test followed to Dunnet test Test. The results showed that nitrogen stress affect the lipid content of Skeletonema costatum. On media without nitrogen has the highest lipid content of 0, 0690%. Morphology microalgae Skeletonema costatum decreased form chloroplasts in all treatments compared to controls.

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INTRODUCTION

Indonesia has the potential biodiversity as biodiesel. One of the biological resource potential as biodiesel is microalgae. One group of algae that can be used as raw material for biodiesel is Skeletonema costatum. The algae has a fat content of 7.42%, a relatively rapid doubling time is 0.340 days, 1,625 days of harvest time and the relative growth rate of 3.2764 cells / h and can be cultivated mass [5].

In the cultivation of microalgae requires a wide range of both macronutrient and micronutrient elements [6]. The element nitrogen plays an important role in lipid metabolism [7]. Increasingly low nitrate concentrations, the total lipid content in the larger diatoms [8]. Nitrogen levels are too low causing protein split into acetyl-Co-A, so as to increase the levels of lipids in the cells [9]. Therefore, it is necessary to study the physiological effects of stress, nutrient nitrogen total lipid content of microalgae *Skeletonema costatum.*

The results showed that the nitrogen nutrient physiological stress affects on total lipid and morphology of microalgae Skletonema costatum. The highest total lipids achieved on the nitrogen nutrient stress conditions without the addition of nitrogen (0% N). The existence of physiological stress N nutrients have an effect on morphological condition of Microalga Skletonema costatum that is having lysis on its cell.

2. RESEARCH METHOD

A. Growth Curve *Skeletonema costatum*

The growth curve *Skeletonema costatum* diatom and cultured on medium density calculated for 6 hours. Cell density calculation using Hemacytometer. Then do the 100L scale intermediates in the treatment of 100% of KNO 3 (75 g / L fertilizer composition diatoms), 75%, 50%, 25% and without the addition of KNO3 fertilizer.

B. Analysis of Lipid Content

Total lipid content analysis was performed using Soxhlet method. Calculation of the total lipid content according to AOAC (Association of Analytical Communities) [10].

C. Analysis of Data

Analysis of total lipid levels were tested by analysis of variance (ANOVA) followed by Dunnet Test Test at the level of 95%.

3. RESULTS AND ANALYSIS

3.1 Total Lipid Microalgae Skletonema costatum In Nutrients Nitrogen Physiological Stress

Growth is the increase in the number or size of cells that are affected by environmental factors [11]. The growth of microalgae is also influenced by environmental factors such as the addition or reduction in of nutrients in media culture. Conditions of algae biodiesel that is potentially as high lipid content is also characteristic growth. The observed growth characteristics can determine the length of time harvesting [5]. Based on the results obtained penelitiaan has done Skeletonema costatum total lipid content shown in Table 1

Table 1. Percentage of Lipid Content of S. costatum On Nitrogen Stress

Concentration	Average
KN 100%	0.0240a
KN 75%	0.0195b
KN 50%	0.0190b
KN 25%	0.0190b
KN 0%	0.0692c

KN100% (Control) = Nitrogen concentration 100%, KN75%= Nitrogen concentration, KN50%= Nitrogen concentration 50%, KN25= Nitrogen concentration 25%, KN0%= Without the addition of Nitrogen

Based on Table 1 it can be seen that the microalgae S. costatum at the nitrogen limited conditions tend to accumulate carbon in the form of lipid metabolism [16]. Nitrogen limited The state of led to the concentration of nitrate reductase decreased. There was also a decrease in the concentration of nitrite reductase that have an impact on the increased concentration of glutamate synthase GSIII / NAD (P) H-GOGAT, resulting in the formation of excessive glutamate [17]. Nitrogen assimilation in the conversion of nitrate to ammonium catalyzed by nitrate reductase and nitrite reductase that work sequentially. In the conditions fulfilled ammonium nitrogen is catalyzed by glutamate synthase to produce glutamine. Glutamine will be assembled by α-ketoglutarate to form other amino acids such as aspartate. Meanwhile, ammonium catalyzed by glutamate dehydrogenase in nitrogen limited conditions, glutamate into the TCA cycle is converted to α-ketoglutarate. Then α-ketoglutarate is converted to malate and malic be converted into pyruvate. Furthermore, pyruvate is converted to acetyl co-A. Forms of microalgae defense system in a state gripped by forming an organic lipid. Acetyl co-A in this case as a precursor to the formation of organic lipids such as fatty acid [18]. [19] Adding organic lipids that serve as energy reserves as well as defense barrier of interference outside the cell. Related to defense barrier in the media stress S. costatum 75% to 25% do defense system by accelerating exponential phase when compared to control media. While the media without nitrogen S. costatum tends to heighten biomass. Therefore, cells of S. costatum use nitrogen in their cells for cell growth and development that would likely reduce the total lipid content.

3.1 Morphology Microalgae Skletonema costatum In Nutrients Nitrogen Physiological Stress

In particular nutritional stress conditions greatly affect the nutrients nitrogen morphological conditions Skletonema costatum microalgae. Nitrogen acts as a nutrient for vegetative growth of microalgae such as the formation clorophlast. This can be seen in figure 1.

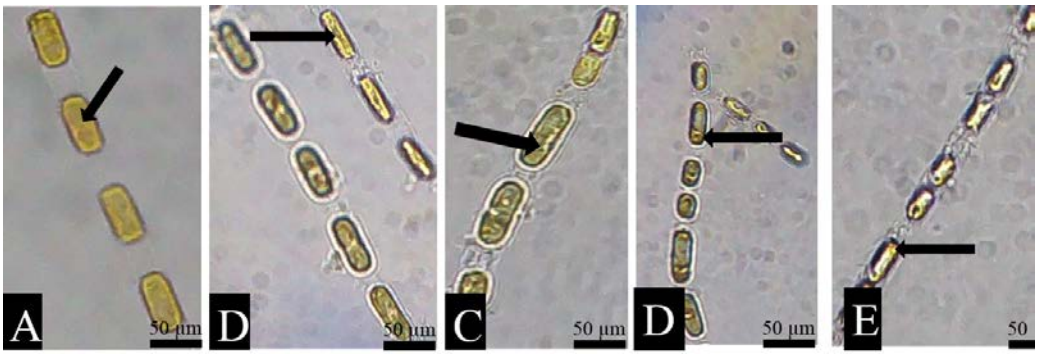

Figure 1. Observations of Cells *S. costatum* In Media Stressing Nitrogen: ↑ = Cloroplas; A: Control; B: 75% Nitrogen; C: 50% Nitrogen; D: 25% Nitrogen; E: Without nitrogen

Based on Figure 1 in all media stress chloroplasts nitrogen has a smaller space when compared to control media. The higher the stress accompanied with decreasing nitrogen chloroplasts in the cell space. Limitations of nitrogen can lead to a reduction of biomass, reduction in photosynthetic pigment composition and activity [14].

4. CONCLUSION

Physiological stress nutrient nitrogen significantly affect the total lipid content of *S. costatum*. In the media 75% to 25% decrease in total lipid content of the lipid with 0.0190%. While the media without nitrogen increased lipid content in the total lipids 0.0692%. Morphology microalgae Skeletonema costatum decreased form chloroplasts in all treatments compared to controls.

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