Phytoplankton (microalgae) as an alternative of renewable energy sources

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Abstract. At present, primary energy resources for various industrial activities including transportation and electricity still rely on fossil sources, such as oil, gas, and coal. Fossil resources are increasingly depleted from time to time and cause the impact of erratic price increases, resources are increasingly limited and cause various environmental effects due to emissions of greenhouse gases produced due to burning these fossils. Various biological resources in Indonesia can be used as a source of raw materials for the production of renewable fuels as a substitute for fossils. Research on phytoplankton (microalgae) as an alternative to renewable energy sources has been carried out. This research was carried out in Teluk Kupang, which has been known as a habitat for Algae life and breeding. The purpose of this study is to obtain data on the existence and types of algae that can be utilized as an alternative renewable energy source. The research method used was observation and laboratory testing to determine the types of algae in Teluk Kupang. The results showed there were 34 types of microalgae, including species of Nitzschia, sp and Thallassiosira, sp which had cell compositions in the form of cellulose, glucan and oils (triglycerides) that had the potential to be extracted and utilized for the manufacture of renewable fuels and other valuable chemicals.

1. Introduction
The phenomenon of global warming and the depletion of energy sources of oil and gas at this time resulting in various studies to find potential alternative energy sources that can be developed from materials that can be renewed (renewable energy) which is also environmentally friendly. Rahmaniaih et al. [1] states that to meet the level of consumption of oil and encourage the development and utilization of renewable alternative energy in the form of biofuel, such as Biodiesel. Chisty [2] states that biodiesel can be produced from various types of plants. Currently, the most commonly used sources of biodiesel are jatropha, palm oil, coconut, corn, as a diesel mixture. However, it is feared that there will be disruptions to world food stability because the market demand for biodiesel will compete with market demands for food crops so that one effort in several countries to increase energy independence is to prepare potential alternative fuels that are come from microalgae [3].

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Research on microalgae, especially phytoplankton, as biodiesel base energy resources have been done. Microalgae is a unicellular species that can live in a colony or solitary and is a photosynthetic microorganism that has the ability to use sunlight and carbon dioxide to produce biomass and produces about 50% of oxygen in the atmosphere [4].

Microbial diversity on earth is very high, so with the high diversity, it is possible to get potential microalgae to produce biodiesel in large quantities. The content of fats and fatty acids in microalgae is a source of energy produced from the process of photosynthesis which is a hydrocarbon and is thought to produce energy that has not been extracted and fully utilized [5].

### Table 1. Composition of oil content of several microalgae species in the stationary and exponential phases

| Types of Fatty Acids | Nitzchia, sp | Thalassiosira, sp | Synechococcus, sp | Dictiosphaerium pulchellum | Stichococcus sp | Synechocystis, sp | Scenedesmus sp |
|----------------------|-------------|-------------------|------------------|---------------------------|----------------|-----------------|---------------|
| Ac.miristat (C14:0) | 3.15*       | 6.37*             | 26.09*           | 2.45*                     | 1.54*          | 28.24*          | 1.03*         |
| Ac.palmitat (C16:0) | 2.67        | 4.59              | 25.96            | 2.38                      | 2.12           | 13.34           | 1.12          |
| Ac.stearat (C18:0)  | 13.25       | 19.61             | 13.94            | 12.56                     | 17.61          | 5.89            | 5.76          |

Source: [6], Note: * = stationary phase

According to Pratoomyot et al. [6], the diversity of microalgae species will make the fatty acid content in microalgae also varied. His further research shows that in general there are differences in the content of fatty acids in microalgae during the exponential phase and the phase of the stage, as shown in Table 1.

One reason for the development of biodiesel from microalgae by developed countries in Europe is because the oil content in microalgae is quite high, but it is also related to the environment. The composition of fatty acids in microalgae is very varied causing the characteristics of biodiesel produced also varies.

This study aims to obtain data on the existence of types of microalgae that have been studied containing oil so that they can be used as an alternative renewable energy source in Teluk Kupang, Kupang City, Nusa Tenggara Timur.

### 2. Material and Method

This research is an observational study and laboratory testing to obtain information about the types of phytoplankton in the waters of Teluk Kupang, so it can be identified several types of microalgae that have been studied in advance which are proven to have oil content so that it can be developed for further research.

#### 2.1. Research Tools and Materials

The tools, materials, and uses used in this study are shown in Table 2. Determination of sampling locations using a purposive sampling method and sampling techniques using composite sampling and plankton net methods. Water and phytoplankton samples were then analyzed at the Biology Laboratory of the Faculty of Science and Engineering, Nusa Cendana University, Kupang.
Phytoplankton's identification to determine the types of microalgae refer to the book Identification of Plankton and references from Sachlan [7].

### Table 2. Equipment, materials and use of research

| Name of Equipment and Material | Use                                      |
|--------------------------------|------------------------------------------|
| Thermometer Hg                 | Measure temperature                      |
| Sample Bottle                  | Store water samples                      |
| pH meter                       | Measuring the pH of water                |
| Label                          | Sample marker                            |
| Plastic bags                   | Storing sample bottles                   |
| Plankton net                   | Take and filter plankton samples         |
| Balance                        | Weigh the sample                         |
| Filter paper                   | Filter sample                            |
| GPS (Global Positioning System)| Determine the coordinates of the sampling location |
| Ice Box                        | Save sample                              |
| Binocular Microscope           | Identification Aids                      |
| Magnifier                      | View and identify samples                |
| Identification Book            | Identifying Plankton                     |
| Seawater                       | Observation object                       |
| Phytoplankton                  | Observation object                       |
| Lugol 4%                       | Preserve the sample                      |
| Acetone 90%, H_2SO_4, HgCl_2   | Material analysis                        |

2.2 Research procedures

This study used an observational method at two research stations with eight sampling points for microalgae collection in Teluk Kupang. The sampling points are shown in Table 3.

### Table 3. Sampling points

| No | Location | Station | Coordinate       |
|----|----------|---------|------------------|
|    |          |         | S               |
| 1  | Manikin  | I A     | 10° 07'339"     |
| 2  | Manikin  | I B     | 10° 07'474"     |
| 3  | Manikin  | I C     | 10° 07'567"     |
| 4  | Manikin  | I D     | 10° 07'646"     |
| 5  | Alak     | II A    | 10° 10'450"     |
| 6  | Alak     | II B    | 10° 10'463"     |
| 7  | Alak     | II C    | 10° 10'477"     |
| 8  | Alak     | II D    | 10° 10'492"     |

Phytoplankton sampling using a 20mm plankton net [8] by filtering as much as 10 liters of water at each sampling point, netted phytoplankton samples are collected in a container and poured into a 250 ml sample bottle. Samples are then given 4 drops Lugol 4% and samples are taken to the laboratory for identification.
3. Results and Discussion

Land use along the Teluk Kupang coast is residential areas, ports or water transportation, fishing and aquaculture, hotels, restaurants, markets, shop houses, and agricultural land. These activities can affect the quality of coastal waters, where each coastal activity produces both organic and inorganic waste that can enter the bay waters which can be carried by rainwater or from irrigation and carried by river water or discharged directly by residents into the seas.

Table 4. Types of microalgae species at the research location

| No | Station | Total Species | Number of Species | Species Type |
|----|---------|---------------|------------------|--------------|
| 1  | IA      | 8             | 27               | Triceratium americanum(7), Rhizosolenia alata(4), Fragilaria, (4) Rhizosolenia acicularis(1), Navicula(1), Lioloma pacificum(1) Asteromphalus (1), Biddulphia(8) |
| 2  | IB      | 6             | 16               | Chylindrotheaclosterium(1), Fragilaria(3), Coscinodiscus(1), Thallassiosira(1), Rhizosolenia alata(6), Guinardia(4) |
| 3  | IC      | 8             | 18               | Stephanophysis nipponica (2), Rhizosolenia setigera(3), Rhizosolenia sp(4) Rhizosolenia acicularis(2), Nitzschia sp(1), Lioloma pacificum(3), Flagilaria (2), Bacillaria(1) |
| 4  | ID      | 2             | 3                | Pinnularia (1), Leptocylindrus danicus (2) |
| 5  | IIA     | 9             | 26               | Triceratium (1), Streptooheca thamensis (1), Stephanophysis palmeriana (3), Rhizosolenia sp(12), Rhizosolenia acicularis (1), Peridium (1), Nitzschia sp(1), Coscinodiscua (4), Navicula (2) |
| 6  | IIB     | 7             | 24               | Sagita enflata (5), Rhizosolenia styliformis(6), Rhizosolenia antennata (1), Rhizosolenia alata(2), Rhizosolenia acicularis (1), Pinnularia (4), Nitzschia seriata (5) |
| 7  | IIC     | 6             | 15               | Rhizosolenia alata (5), Navicula (1), Leptocylindrus danicus (1), Eucampia (2), Coscinodiscus (2), Leptocylindrus minimus (4) |
| 8  | IID     | 6             | 13               | Rhizosolenia alata(5), Eucampia (2), Flagilaria(1), Navicula (1), Nitzschia seriata (2), Pinnularia (2) |

Of all activities on the coast of Teluk Kupang, nitrate and phosphate are the most predominant predicted nutrients that can enter the waters where there is an agricultural activity directly adjacent to the coastal waters and applying chemical fertilizers namely Urea as a source of nitrate nutrients, SP36 as phosphate nutrients and KCl as an element of Potassium. The remnants of chemical fertilizers that are wasted from agricultural activities can enter the water and can increase water fertility, but excessive amounts can result in enrichment or eutrophication which triggers growth and population of phytoplankton (microalgae).

Microalgae are microscopic plants that belong to the class of algae and live in colonies and single-celled in all waters, both freshwater and seawater. Based on observations found 34 species of microalgae at eight observation points with a total number of species of 142 microalgae species including 4 classes are Bacillariophyceae, Dinofyceae, Chlorophyceae, and Cyanophyceae. Microalgae observations are shown in Table 4.

From the results of 34 species of microalgae found at the site, there are two types of microalgae namely Nitzschia sp and Thallassiosira sp that have been previously studied have cell walls containing cellulose, glucan, and oil that have the potential to be extracted and used for the manufacture of renewable fuels and valuable chemicals. Nitzschia sp and Thallassiosira sp are microalgae that belong to the Bacillariophyceae class [9] and have an important role in aquatic ecosystems as primary producers. Environmental factors that influence the growth of microalgae include light, salinity,
temperature, water pH, osmose pressure which may spur or inhibit growth [10]. According to Pratoomyot et al. [6], the diversity of microalgae species will make the content, fatty acids in microalgae also vary. Further research shows that in general, there are differences in the content of fatty acids in microalgae during the exponential and stationary phases (Table 1). In Botes [11], Nitzschia sp and Thalassiosira sp can be classified as follows (Table 5).

| Table 5. Classification of Nitzschia sp and Thalassiosira sp |
|---------------------------------------------------------------|
|                  | Nitzschia sp       | Thalassiosira sp |
| Division         | Bacillariophyta    | Chrysophyta      |
| Class            | Bacillariophyceae  | Bacillariophyceae|
| Ordo             | Bacillariales      | Centrales        |
| Family           | Bacillariaceae     | Coscinodiscineae |
| Genus            | Nitzschia          | Thalassiosiraceae|
| Species          | Nitzschia, sp      | Thalassiosira sp |

Microalgae is a source of biomass containing high beneficial components such as protein, carbohydrates, fatty acids, including triglycerides, which can be converted into biodiesel. Several types of microalgae have potential as a source of oil. Microalgae oil content varies depending on the type of microalgae [12], it can be seen in Table 6 that the percentage of oil in Nitzschia sp is quite high at 45-47%, as well as the Thalassiosira sp percentage of 30%. Fat in microalgae consists of glycerol, saturated fatty acids or unsaturated fatty acids.

| Table 6. Oil content in some microalgae |
|----------------------------------------|
| Microalgae                            | Oil content (%) |
| Botryococcus braunii                   | 25-75           |
| Chlorella sp                           | 28-32           |
| Cryptothecodinium cohnii               | 20              |
| Cylindrotheca sp                       | 16-37           |
| Dunaliella primolecta                  | 23              |
| Monallanthus salina                   | 20              |
| Nannochloropsis sp                     | 20-35           |
| Neochloropsis oleabundans             | 35-54           |
| Nitzschia sp                           | 45-47           |
| Phaeodactylum tricornutum              | 20-30           |
| Schizochytrium sp                      | 50-77           |
| Thalassiosira sp                       | 30              |

(Source: [12])

The use of microalgae as raw material for biofuels has a higher profit compared with food crops. The advantages are phytoplankton growth that is faster than food crops, which is time to grow twice in 3.5 hours [2] microalgae can live in freshwater and seawater, and do not compete with food crops and also relatively more production costs low [13]. In addition, microalgae also produce at least 30 times more than land plants. Therefore, the cultivation of microalgae as a raw material for biofuels can save space and have high efficiency and effectiveness which is quite promising in the future.

Through several processes such as biophotolysis and fermentation, microalgae can produce hydrogen. Hydrogen is very easy to convert to heat, electricity, and fuel. The fat accumulation that occurs in the body of microalgae tends to increase when it is in a concentration and stressed environment [14]. So with the condition of Indonesia which is experiencing an energy crisis, microalgae can be an alternative development potential to produce promising biofuel fuels.
4. Conclusion
The climate in Indonesia is a tropical climate with sun exposure throughout the year, which is very suitable for the development and life of microalgae. The condition of the waters of the Kupang Bay, where there is a supply of nutrients from agricultural activities and domestic waste in the coastal areas, can be nutrients that increase water fertility. *Nitzschia sp* and *Thallassiosira sp* species that have the potential to become sources of energy or biodiesel raw materials have been found in Teluk Kupang, so they can be recommended for further research on microalgae culture to be extracted into biodiesel.

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