Cultivation of Caulerpa Taxifolia as Feedstock of Bioenergy

Tri Poespowati (Corresponding author)
Chemical Engineering Dept., The Institute of National Technology
Jl. Bend. Sigura-gura 2 Malang – Indonesiam (65145)
Tel: +62 341 551431 E-mail: poespowati@yahoo.com.au

Jimmy
Chemical Engineering Dept., The Institute of National Technology
Jl. Bend. Sigura-gura 2 Malang – Indonesiam (65145)
Tel: +62 341 551431 E-mail: j_roring@yahoo.com

Sidik Noertjahjono
Electrical Engineering Dept., The Institute of National Technology
Jl. Bend. Sigura-gura 2 Malang – Indonesiam (65145)
Tel: +62 341 551431 E-mail: sidik@fti.itn.ac.id

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Abstract

Research has been conducted on cultivating Caulerpa types macroalgae inside 60x60x30 cm^3 indoor aquarium. Caulerpa that has potential as a feedstock in the manufacture of bioethanol has been used merely as a counterweight plants in ornamental saltwater fish farming and as feed material in the salt water pond. Strain Caulerpa is attached to the rock substrate. Laminar conditions and alight are created by seawater circulation pump and installation of warm white 20W fluorescent lamp. This condition is presented every day with a height of 10 cm from water surface. Caulerpa nutrients derived from nitrate and phosphate released by clown fish, types of saltwater fish. Reduced sea water due to evaporation is recovered by adding fresh water to the extent of its original height. Caulerpa is weighed before and after cultivation for 2 months. In 2 months' time, the significant development of Caulerpa is visually noticed by its growth of fronds, rhizome and rhizoids. It is signed by the brightness of the green colour, with very strong roots attached to the substrate. Variations in the number of fish and the type of substrate performed during cultivation. The weigh results showed a significant difference before and after cultivation with an average percentage growth of 361.86%.

Keywords: Macroalgae, caulerpa, cultivation, clown fish

1. Introduction

IEO (International Energy Outlook) 2010 states that the world's energy consumption increased by 49% or 1.4% per year from 495 quadrillion BTU in 2007 to 739 quadrillion in 2035. So we need to think about the development of technology to utilize renewable energy sources derived from biomass such as macro algae (IEO, 2010).

In the 1990's, Caulerpa found in the Mediterranean has been causing problems. Its tremendous growth endangered other habitats (Meusnier 2001, Lars 2001, Box 2009, Ceccherelli 2002, Klein 2008). There are approximately 73 species of Caulerpa in the world, some of which are planted in the aquarium (Vides, 2002). In Southern California, Taxifolia is one of Caulerpa species that is widely grown in aquarium with live rock substrate (Frisch, 2002). As an archipelago, Indonesia has a long coastline. It is very suitable as a place for algae to naturally grow. There is at least about 550 species that taken their habitat in Indonesian coast. Some types of algae have been cultivated including the Euchema Cottonii and Gracelaria. In Indonesia, Caulerpa is often found in the sea coast of Java as wild crops that are used as food (Trono, 1988). Meanwhile, Caulerpa Taxofolia or aquarium Caulerpa has not yet being cultivated in Indonesia. Mostly this type of Caulerpa is being used as a Nitrate absorbing plant for saltwater aquarium.

Algae are the organisms that are resulted from photosynthesis process. They consist of two major categories, which are microalgae and macro algae. They grow in an aquatic environment, and containing carbohydrates that has potential as liquid fuel. Macro algae or seaweed is a type of multicellular plant that grows in saltwater. It can grow rapidly until it reaches 60 meters in length (Demirbas, 2010). Macro algae is classified into three groups based on their pigments. They are Brown Algae (Phaeophyceae), Red Algae (Rhodophyceae), and Green Algae (Chlorophyceae). Caulerpa is including in green macro algae family. Algae have the highest oil content compared to the other oil-contained biomass such as soybean, corn, palm, and castor. Oil content (lipid) from algae can reach 50-70%. And oil produced by algae could reach 1200 - 2200 gal/acre/year assuming a stable cultivation environment per year. When the oil is converted into biodiesel, it is much more efficient than biodiesel from soybeans (Woertz, 2007).

From the 6 different Caulerpa species that have been studied, highest carbohydrate content (30%) is produced by Caulerpa Prolifera occurring during the rainy season (Robledo, 2005). The highest protein content (22%) is produced by Caulerpa Pasppaloides that occur during the dry/hot season. Protein is also contained in Caulerpa Lentillifers, it levels between 20-67% (Matanjun 2009), while Sulfate Polysaccharide is found in Caulerpa Cupressoides (Eshaq, 2010 and Rodrigues, 2011). This carbohydrate can be converted into bioethanol by fermentation (Eshaq, 2010) and by using acid hydrolysis pre-treatment as well as enzymatic treatment (Lee, 2011). According to Pattama (2006), Caulerpa Lentillifera and Ulva Reticulata contain 21.06% and 12.49% of protein respectively. Kumar examined the biochemical composition of 18 species of macro algae that are grouped in Chlorophyta (one of them is Caulerpa Racemosa), Phaeophyta, and Rhodophyta. He found that the levels of carbohydrates, proteins, amino acids, phenols, and lipids are different in each species. The highest carbohydrate content is found in Chlorophyta group (Kumar, 2010).

Research using seaweed Gracilaria Salicornia and Caulerpa Lentillifera as a biofilter in aquaculture circulation system was conducted by Chaitanawisuti. It states that these two types of seaweed change the water quality such as its temperature, conductivity, salinity, pH, oxygen dissolved and total suspended solids. It also states that Caulerpa growth is much faster than Gracilaria Salicornia (Chaitanawisuti, 2011). According to Zuljecic, the overall regeneration of algae is occurring in any part of its thallus - the fronds, stolons, and rhizoids. The thallus formation happens on the 10^{th} day with a temperature around 25°C (Zuljetic, 2002). Strain Caulerpa taken from a metre depth and cultivated using 200 µ $Em^{-2}.s^{-1}$ light intensity produces a higher photosynthesis rate compared to the strain that is taken 37 metres depth, with the same light intensity, as well as one metre depth with 50 µ light $Em^{-2}.s^{-1}$ light intensity (Riechert, 1986). The growth of Caulerpa at 20 m depth in Salt River Canyon, Virgin Islands, US is obstructed by macro fauna such as shrimps and crabs (Williams, 1985).

Poespowati in 2010 ran a study of kinetics and pyrolysis of microalgae Chlorella sp. Dry. It was conducted using TGA (Thermogravimetry Analyzer). The TGA analysis covers differential scanning calorimetry, thermogravimetry and differential thermogravimetry. Argon was used as the carrier gas. With warming temperature range between 30°C to 800°C, heating rate of 10, 20, and 30°C, and variety of algae particle sizes, it is found that there is a change in the activation energy against the temperature in the kinetics reaction. And in a larger particle size, the exothermic reaction is decreasing. The used Chlorella sp. was obtained from Tasmania, Australia (Poespowati, 2010).

The purpose of this study is to determine the growth rate of Caulerpa Taxifolia that is cultivated in the aquarium by weighing before and after study. This is a preliminary study before making a computerized growth rate measurements using a microcontroller.

2. Research Methodology

Strains of green macro algae Caulerpa taxifolia is derived from saltwater fish collector or hobbyist, where algae is usually used as a counterweight marine life. After the acclimation to saltwater in the aquarium, strain is attached to the substrate. Substrate used is freshly live coral that is placed above the sea sand (see Figure 1) and the cultivation carried out for 2 months. Clown fish is used as a nutrient provider for the Caulerpa. Before its placing in the aquarium, clown fish is also acclimated in advance for about 2-3 hours to avoid shock.

Cultivation is conducted in nine 60x30x30 cm^3 sized aquariums made of glass as shown in figure 2. Because the cultivation is conducted indoor, the light is obtained by fitting 20W warm white fluorescent lamps that have suitable spectrum for the growth of Caulerpa. The light is mounted above the water surface to a height of about 10 cm. In that distance, the light can still penetrate the saltwater in the aquarium.

A starter bacterium is introduced to accelerate the cycle. This cycle is aimed to stabilize the content of ammonia in saltwater at the beginning of the setting. Saltwater used is a mixture of fresh sea water and sea water wastage from saltwater fish collector/hobbyist.

The cultivation is conducted at a room temperature, where the temperature ranges between 25-28 °C. Overheat temperature can cause losses on the fronds and rhizome fragments. Water analysis for salinity, Ca, Mg, NO_3, PO_4 is done once a week and control over the ideal conditions for the growth of Caulerpa is maintained. Analysis of cultivation results is done visually by observing the increase of fronds, rhizome and its rhizoids. As well as that, a digitally weighing process is done before and after the cultivation.

During the cultivation process it is necessary to take a special care including 10% salt water replacement per week, insertion of additive such as Ca, Mg and sodium bicarbonate once a fortnight to control the hardness of the water, provide fertilizer and supplement for roots, stems and leaves growth, removing attached microalgae from aquarium glass, feeding the fish every 2-3 days, and adding fresh water to replace the evaporated salt water.

3. Results

Visual observation is carried out on the results of Caulerpa cultivation. Newly grown shoots on the fronds, rhizome and rhizoids are appearing to be bright green and fresh, as shown in figure 3 (before cultivation) and figure 4 (after cultivation). Whether Caulerpa grows or dies can be noted by the colour of its leaf tips. Brownish-black in colour is a sign that Caulerpa will die. But when it has the colour of whitish green and leaves stand erect then Caulerpa is vastly growing. Its best growth is achieved from adding fish at the cultivation process with coral and sand as its media.

Aside from the colour, the growth of Caulerpa can also be seen physically by its increase of length from the birth of new shoots. Caulerpa is weighed before and after cultivation. The result can be seen in table 1 below.

As can be seen from Table 1, the best growing progress is cultivation using fish with coral as its media, followed by; again cultivation using fish but this time is with sand and coral as its media. The lowest percentage is cultivation with coral as its media but without fish. With the percentage growth 554.82%, 329.3% and 201.47% respectively, out of the three parameters, the average per cent growth is 361.86%.

4. Discussions

With the above result, Caulerpa cultivation conditions during the study have met the requirements for its life. The conditions are:

a. Availability of adequate nitrate and phosphate. They are resulted from clown fish wastage and also from fish food. The amount of nitrate and phosphate is controlled by setting the feeding of fish for only once per 2 days. To sustain the life hood of fish, it needed to be adjusted before moved to the aquarium.
b. Sufficient light obtained from 20W warm white fluorescent lamp, or identical to 6500k spectrum that is adequate to the growth of Caulerpa. Lamp is set by placing it 10 cm above the water surface. With this distance the light is still able to penetrate the bottom of the tank so Caulerpa will receive enough light. Less intensity of lighting will cause a Caulerpa leaves and stems to fall.
c. Aquarium has a glass base or normally called bare bottom. It aims to ease the maintenance process. The removal of other habitats that grow against the glass is easily done. Also the cycling process is moving faster. Cycling process performed on the new tank aims to the removal of gases that released by saltwater, eg: ammonia, which is obstructing the growth of Caulerpa. For a faster cycling process it is

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necessary to add starter bacteria that can help speed up ammonia decomposition. Starter bacteria will consume the excess ammonia. Naturally saltwater already has ammonia-controlling bacteria, but for laboratory scale cultivation, providing starter bacteria is needed for the best result.

d. The water used in this study is a mixture of fresh saltwater and used saltwater provided by aquarium hobbyist. Used saltwater contains high phosphate and nitrate levels that are adequate to the process of cycling. Fresh saltwater is still yet not contaminated by marine life pollutants, and is balancing the pollutants level of used saltwater. Water is circulated under laminar condition with the installation of water pump. And to maintain the clarity of the water it needs to come with a filter. The filter has to be replaced on a regular basis. The water level in the tank is 15 cm.
e. Live rock is used as a medium for Caulerpa roots attachment. Live rock is not causing ammonia blooming as it has been covered with coraline algae or micro sponge that serves as a natural filter. Dead rock was not selected because it can lead to ammonia blooming which causes water to be toxic to Caulerpa as well as the fish.
f. The suitable temperature for Caulerpa growth is between 25-28°C. If the temperature is more than that range, it could lead to stem and leave falls. Thermometer is installed inside the water tank to monitor the temperature.
g. The influential parameter in the development of Caulerpa includes magnesium, calcium, phosphate, salt and nitrate levels should always be maintained in an ideal condition. In this study, phosphate and nitrate levels over 10 ppm, magnesium levels below 1500 ppm, 500 ppm calcium levels, and salinity level of 1026 ppm.

Subsequent research will be carried out by means of a computerized weight measurements using microcontroller, therefore an accurate result is gained.

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Tabel 1. Weight measurement before and after cultivation

Aquarium	Initial Weight, gram	Final Weight, gram	Growth Percentage
1	6	21,6	360,00
2	3,8	7,9	207,89
3	1	4,2	420,00
4	7,7	16	207,79
5	10,3	15,4	149,51
6	4,2	54,9	1307,14
7	12,2	25,2	206,56
8	3,9	8,8	225,64
9	1,8	3,1	172,22

Figure 1. Cultivation process of Caulerpa

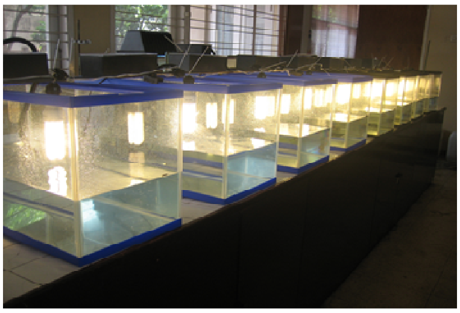

Figure 2. Cultivation process of Caulerpa in nine aquariums

Figure 3. Caulerpa before cultivation process

Figure 4. Caulerpa after cultivation process