Use of Nutrient on Algae Growth to Produce Biodiesel

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Abstract: Algae biodiesel is now widely accepted as a future source of biodiesel worldwide. In this study culture collection of algae was done from different places of Nepal. Main focus was given in to collect as much as greater no. of species from similar climate zone. More than 60 algae sample collected and taken at lab for identification and analysis. Out of 60 samples 11 types of algae species were identified with majority of filamentous groups of green algae. Out of identified algae oedogonium was chosen for detail analysis. Two batch of algae growth was carried with 5 experiments and 3 repetition of each type. 5 set of experiments are initial dose of Agroliv/water 0ml/L, 0.15ml/L,0.3ml/L,0.45ml/L,0.60ml/L and by name A, B, C, D, and E. 21 days of algae growth observation was carried out on each batch. DO maintained in the experiments are in the range of 5mg/l to 8 mg/l.

Temperature, pH, and DO were measured daily and Nitrate, Phosphors and algae growth were measured periodically in the interval of 3 days. The maximum concentration of Oedogonium was found at 14.34 g/L with pH 7.27 mg/L at experiment C. Maximum nitrate removal efficiency of oedogonium has found 86.25 %at and maximum phosphate removal efficiency has found 66.5% on 21 days growth period. Maximum Oil yield capacity of Oedogonium is found 32.90% of its dry weight at experiment D and minimum oil yield capacity of Oedogonium is found 23.33%.of its dry weight at experiment E. Up to 90% of Oedogonium oil is converted in to Biodiesel. high and low dose of nutrient is unsuitable for biodiesel production. Fatty acid and glycerol production is high 12% in Oedogonium oil in experiment C and low in 9% in experiment A.

Keywords: Oedogonium; Nitrate; Phosphate; Algae Oil; Biodiesel; Agroliv; Fatty Acid

1. Introduction

Microalgae are one of the most promising alternative and renewable feedstock sources for producing biodiesel. These are the third generation fuels. The algae-for-fuel concept has gained renewed interest recently with the world wide fluctuation in energy prices. The term "algae" encompasses a diverse group of simple organisms, ranging from large multi cellular seaweeds to microscopic single-celled organisms and includes both prokaryotic and eukaryotic phyla (Mata et al., 2010). Algae are typically photoautotrophic and generate organic compounds for growth and reproduction through photosynthesis. Algae play a very important role in ecosystems as primary producers and contribute 40 to 50% of the oxygen in the atmosphere (Anderson, 2005). Algae range in size from a few micrometers to over 30 m in length. There are two classifications of algae: macro algae and micro algae. *Oedogonium* is filamentous algae and chosen in this study. Biodiesel production capacity of *Oedogonium* is shown greater than that of Spirogyra and Cladaphora (Khola and Ghazala, 2012).

Similarly in another research Oil production capacity of *Oedogonium* is found 9.2 ml per 30 gram dry weight of algae which is greater than Spyarogyara 7.3ml per 30 gram (Hossain et.al, 2008). In recent day, algae research and farming for biodiesel production is being increased. Chicken manure, dairy manure, urea & vegetable compost are some of the inexpensive and locally available fertilizer sources for algae cultivation. Algae cultivation by using chicken manure (Lee, 2011). Urea was used as cheapest sources of nutrient for algae cultivation (Zuka et.al.2012). Algae cultivation is done by using natural water source without nutrient (Ramraj et.al, 2010). Dairy manure is also a cheap and feasible source of nutrient for algae growth (Mulbery and Wilkie, 2001). In this research, algae growth study was done by using varying dose of liquid fertilizer agroliv. There are many methods of oil extraction adopted by different research scientists. Ultrasonic extraction assisted with solvent (Wiyarno et.al, 2008). Lab grade solvents, chloroform, methanol, ethanol and hexane used in microalgae biomass extraction and comparison of yield (Blanc, 2008). (Tomson et.al, 2012) comparing 8 different solvents, N-hexane, ethyl acetate, diethyl ether, 2-propanol, acetone, ethanol (95%), ethanol / water / acetic acid (80/20/1 v/v/v) and ethanol / water (80/20 by volume) and they found ethanol is best solvent for oil extraction.

Similarly, in comparison of hexane and ether for extraction of Navicula sp (Vincent et.al, 2010) found hexane is best solvent for extraction of Navicula sp.oil. In this study, Oil extraction of algae was done by using hexane and ethanol (1:1 ratio). After extraction of oil biodiesel was made by transesterification method.

2. Materials and Methods

2.1 Culture Collection

Algae sample was collected from different parts of country. The pH of water was tested in the field by digital pH meter. The identification of green algae sample was performed by observing under the compound microscope. Identification characters were followed as per the descriptions made by (Kumar, 1990 and AFC, 2011).

2.2 Algae Growth Media

The algae of choice for this study were Oedogonium. Commercial liquid fertilizer "Agroliv" having Nitrogen 2.5%, Phosphate 1.5%, Potassium 3%, Copper 0.5%, Manganise 0.2%, Zinc 6%, Boron 0.3% and Iron 0.5% was used for this study. Simple 3W/220V, 50 Hz aquarium air pump with 150 L/h air injection capacity was used to maintain the oxygen supplies in the algae growth vessels to maintain dissolved DO level of water is above 5 ppm. Algae were grown 20 L capacity open PVC vessels with 6:1 surface area and depth ratio. All cultures were held at a room temperature. Algae growth in 12 hour day cycle and 12 hour night cycle was done.

2.3 Experiment Design

This study used five treatments, three test replicates within each treatment. The five treatments varied relative to N and P content in media with all other media components held constant. Temperature and light intensity were varying throughout all experiments. The experiment setups are A 0 ml/L, B 0.15ml/L, C 0.3ml/L, D 0.45ml/L and E 0.6ml/L of agroliv/water.

Temperature, pH and dissolved oxygen were measured daily. Nitrogen, phosphorous and algal growth measured twice a week over 21 day's algae growth period.

2.4 Measurement of Algae Biomass

Growth of algae was measured by weighting its fresh and dry weight. Wet weight is taken after gentle filtration of algae biomass from dried and pre weighted sonex filter paper no.1 and takes a wet weight of sample and continues drying it an oven at 100^0C overnight.

Biomass (g) =

Total weight (g) – Filter paper weight (g)

Dry Coefficient =

(Dry weight)/ (Wet weight) of sample

Dry weight =

Dry coefficient * Total Wet weight

Specific growth rate =

(Final weight – Initial weight)/100

2.5 Oil Extraction and Biodiesel Production

Oil extraction of algae was done by using hexane and ethanol solvent (1:1 ratio). First algae biomass was pestles as much as possible by grinder. After grinding algae biomass was dried at oven in 3 hours at 80^0C, after drying algae biomass, it was mixed with solvent in 1:8 ratio by hexane ethanol (1:2) ratio and kept the mixture in 24 hour. After 24 hour algae biomass was rest at the bottom and solvent and extracted algae oil was rest at upper level. Separate the layer by flask separator and measure algae biomass and oil mixture separately. Mixture of oil and solvent was heated on continuous magnetic stirrer at 60^0C until all solvent was removing by evaporation. After evaporating solvents, measure oil in terms of volume. Algae oil is further processed in to biodiesel. In this process 200ml methanol per liter algae oil plus sodium hydroxide (6g/L of algae oil) was mixed and properly stirred until all sodium hydroxide was dissolved on methanol. Methanol and sodium hydroxide solution was mixed with hot algae oil, properly stirred and the solution was kept for 24 hours. After 24 hours glycerine, fatty acids, algae pigment and other organic residue settled at the bottom and biodiesel was separated at the top. Biodiesel layer was carefully separated by flask separator and measured. Washed biodiesel by 5% of water until it was cleaned and finally kept under warm environment. After this process, biodiesel was ready for analysis.

3. Results and Discussions

3.1 Algae Identification and pH Measurement

The algae isolated from water bodies and identified by microscope and characterized as per literatures. The pH of water is measured by digital pH meter in algae sample collection point. The identified algae species are Spirogyra(Lalitpur, pH 8.2), H. Reticulatum (Lalitpur, pH 8.5), H.africanum(Kavrepalanchowk, pH 8.3), Pediastrum Simplex(Pokhara, pH7.8), Cladaphora(Kirtipur,pH8.4), Oedogonium(Lalitpur, pH 7.7), Stigicolonium (Pokhara, pH7.6), Oscilatoria(Pokhara, pH 8.3), Ulva laecuta(Pokhara, pH 7.8), Cyanobacteria(Kavrepalanchowk, pH 8.2)

3.2 Growth Media Test and Selection

Algae were grown in ground water. The initial properties of ground water were pH 7.2, DO 5 ppm, Nitrate-N 20 ppm, Phosphorus 15 ppm, Potassium 10 ppm and chlorine 0 ppm. In this study different doses of commercial fertilizers had tried, Different doses of granular urea, granular N, P, K, Liquid fertilizer plant grower, Nitro King, phosphate king, Organic liquid fertilizer "NPK", Agroliv etc were used as a nutrient for initial algae growth. Out of those fertilizers "Agroliv" with doses 0.15ml/L to 0.60 ml/L of water was shown satisfactory and used this as a nutrient media.

3.3 Nutrient Removal by Algae

Nutrient removal pattern of algae is shown in the Figure.1

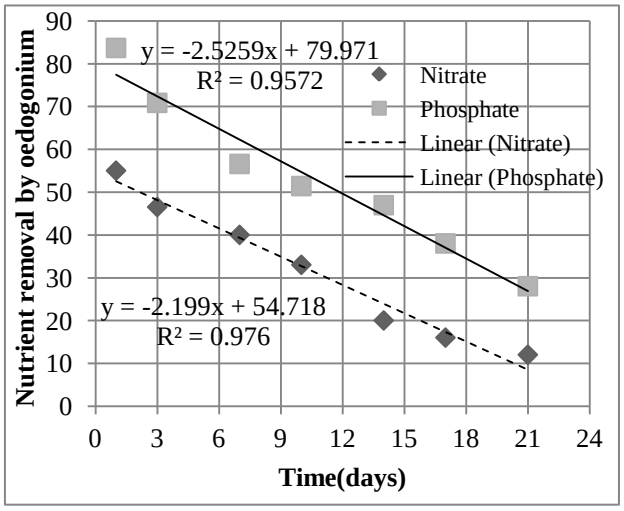

Figure 1: Nutrient Removal by Algae

From Figure 1, it is shown Oedogonium removes nitrate-n from 55 ppm to 12 ppm and phosphorous removal from 84 ppm to 28 ppm. Slope of phosphorous removal trend line is shown higher than that of nitrate-n removal so that it can be concluded that oedogonium is best for phosphorous removal.

3.4 Algae Biomass on Different Nutrient Dose

Algae biomass on different nutrient dose is as shown in Figure 2. It shows that Maximum growth occurs on experiment D in day 4. Highest algae growth rate is shown on experiment C and lowest on experiment A.

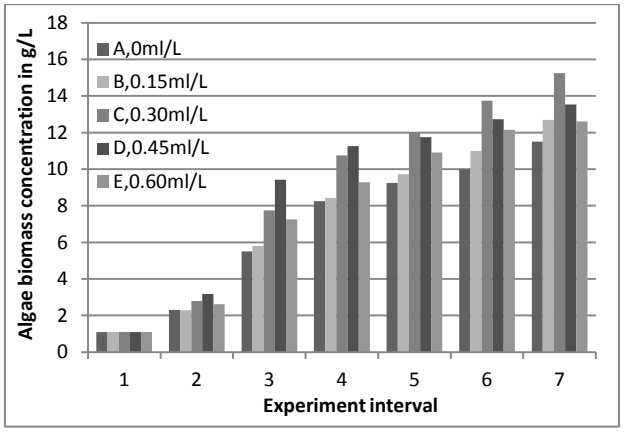

Figure 2: Algae Biomass

3.5 Algae Oil on Different Nutrient Dose

Algae oil on different nutrient dose is shown on Figure 3.

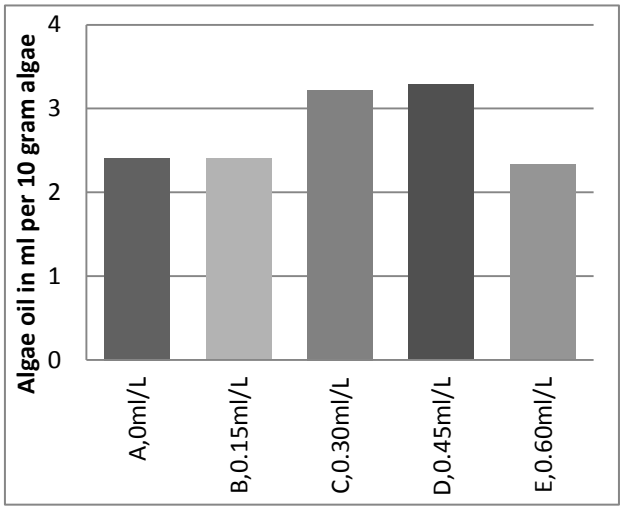

Figure 3: Oedogonium oil

Figure 3 shows the comparison of extracted algae oil on different doses of nutrient. Maximum algae oil was found in 3.29 ml per 10 gram dry algae in experiment D. The minimum oil yield was found is 2.33 ml per 10 gram fresh algae in experiment E.

3.6 Algae Biodiesel

Algae Biodiesel on different nutrient dose is shown on Figure 4. It shows algae biodiesel on different doses of nutrient. The highest amount of biodiesel production found from experiment C and lowest amount of biodiesel from experiment E.

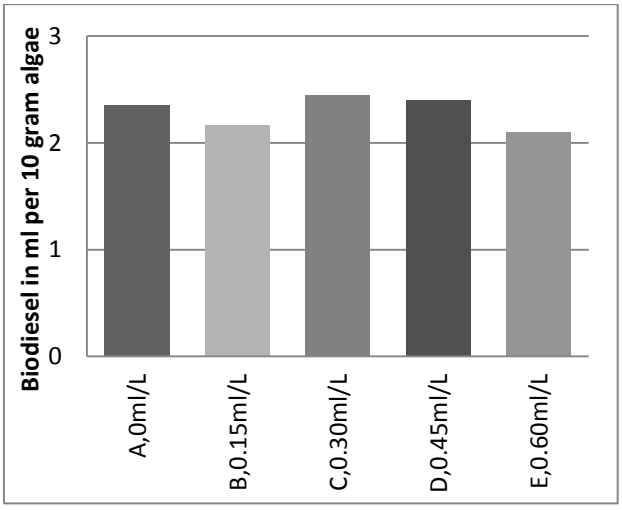

Figure 4: Algae Biodiesel

4. Conclusion

Out of 60 samples of algae 10 algae species were identified with majority of Filamentous group. The temperature measured in the range of 18.67^{0}C to 23.3^{0}C. The maximum concentration of algae biomass was found 15.25 g/L. Maximum nitrate removal efficiency of oedogonium has found 86.25 %at and maximum phosphate removal efficiency has found 66.5% on 21 days growth period. Maximum Oil yield capacity of Oedogonium is found 32.90% of its dry weight at experiment D and minimum oil yield capacity of Oedogonium is found 23.33%.of its dry weight at experiment E. Up to 90% of Oedogonium oil is converted in to Biodiesel. High and low dose of nutrient is unsuitable for biodiesel production. Fatty acid and glycerol production is high 12% in Oedogonium oil in experiment C and low in 9% in experiment A.

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