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Studies on microalgae bioengineering: From CO_2 fixation to lipid extraction

R. Saravanan[1], N. Madhivanan[2], R. Saravanane[3]

[1]*M.Tech student, Dept. of Civil Engg., PEC, Puducherry, India*
[2]*Ph.D research scholar, Dept. of Civil Engg., PEC, Puducherry, India*
[3]*Professor, Dept. of Civil Engg., PEC, Puducherry, India*

Abstract -*Increase in fossil fuel consumption and rapid depletion of fossil fuel resources, there is a need for alternate sources. In addition, the fossil fuel is directly related to air pollution, land and water degradation. In this circumstance, biofuel from renewable sources (i.e. from microalgae) can be a good alternative to reduce the fossil fuel consumption and to maintain a healthy and sustainable environment. Microalgae are photosynthetic microorganisms with simple growing requirements (light, sugars, CO_2, N, P and K) that can produce lipids, proteins and carbohydrates in large amounts over short period of time. These products can be processed into both biofuels and valuable co-products. In this pilot scale study microalgae were cultured in closed loop inclined photobioreactor systems with the supply of lignite burned CO_2 and growth nutrients. The cultured biomass was then harvested for the extraction of lipids by using Bligh and Dyer method. Further the extracted lipids can be converted into biodiesel through a process called transesterification.*

***Key Words*:CO_2 fixation, Lipids, Microalgae, photobioreactor, Spirulinaplatensis**

1. INTRODUCTION

Biodiesel is currently being recognized as a green and alternative renewable diesel fuel that has attracted vast interest from researchers, governments, and local and international traders. The rapid depletion of fossil fuel resources and increase in the greenhouse gas emissions cause the global climate change are creating much interest in the production of biodiesel [1]. In this study,a pilot scale experiment of CO_2 sequestration and lipid extraction from microalgae cultivated in closed loop inclined tube photobioreactor of 100 L capacity. The microalgae *spirulinaplatensis*species were used for lipid extraction and further it will be transesterified for biodiesel conversion. Microalgae are photosynthetic microorganisms that require simple growing requirements as sunlight, sugar, nitrogen, phosphorous and potassium for their growth [2]. With these nutrients microalgae can be cultivated in larger amounts and the lipid production capacity.

Also microalgae have the ability of CO_2 fixation from the atmosphere, industrial gas discharges and soluble carbonates. Most microalgae species can tolerate and utilize higher levels of CO_2, typically up to 150,000ppmv [2]. In general, from 1kg of dry microalgae biomass can absorb 1.83kg of CO_2 [3]. For lipid extraction, algae can be cultivated in two ways i) open ponds and ii) closed photobioreactors. Open pond system is the easiest way for culturing the species and it has the low initial investment compared to photobioreactors, but it requires more land surface. Photobioreactors are closed loop flat, vertical and inclined tubular systems, it requires high initial investment for installation with less land surface. Open pond systems are less efficient when compared to closed systems. Photobioreactors has distinctive advantages over open ponds because of evaporation loses, CO_2fixation, material loss, etc. can be eliminated.

Photobioreactors consists of an array of glass or plastic tubes that captures sunlight and can be aligned in horizontal, vertical and inclined tubes. The tubes are generally 0.1 m or less in diameter [4]. Algae cultures are circulated by mechanical pumps for the complete mixing of CO_2,the O_2 were released at the outlets. The tubes are served as a solar receiver; it provides a platform for the algae to grow by giving a high surface area to volume ratio. Agitation and mixing are very important to encourage gas exchange in the tubes.

2. MATERIALS AND METHODOLOGY

2.1. Materials

The wet biomass slurry of algae *spirulinaplatensis*was initially added in the photobioreactor and it was obtained from a local supplier in Puducherry, India. The reagents used for this study were of analytical grade and were purchased from Sri Rajendra's Scientific and Surgicals (P) LTD.

2.2. Methods

CO_2 Sequestration

Fig – 1: CO_2 sequestration setup at the front, tubular Photobioreactor at the back.

In this pilot scale experiment the lignite was burned into the chamber containing an open at the bottom and a metal pipe is connected to the pretreatment chamber in which the smoke to be passed through it. After all the burning process the smoke was captured in the pretreatment chamber (wet scrubber), water is sprayed at certain pressure so that the SOx, NOx, soot particles, etc. are removed in water droplets and collected at the bottom of the chamber. Only CO_2 is escaped out at the top of the pretreatment chamber and then passed to the bubbling tank. The CO_2 is measured before passing into the bubbling tank for measuring the CO_2 fixation rate by the microalgae biomass. The industrial exhaust gas contains 10-20% CO_2 and also small amounts of SOx and NOx [5].

Biomass growth

Fig – 2: Sample collected at the outlet valve.

Suitable condition for the growth of microalgae culture medium should be a pH of around 9.5 to 11. In addition, the supply of nutrients such as potassium, urea, magnesium, ferrous sulphate, sodium Bicarbonate, etc. to the growth medium.The CO_2 supply of flue gas 12% were injected into the reactor system. Biomass growths were measured by Total Volatile Solids present in the reactor sample.

Biomass harvesting

Fig – 3: Filtered biomass using 70 µm filter cloth.

Microalgae were cultured in the closed loop inclined photobioreactors with essential nutrients. After a certain period of time the cultured biomass slurry were collected at the outlet of the valves fitted in the photobioreactor. Then the biomass slurry is filtered using 70 µm filter cloth by gravity sedimentation method [2].

This method is only suitable for large (>70 µm) microalgae species such as *Spirulina.* The samples should be filtered approximately 3 to 4 times for obtaining maximum biomass yield. The filtered biomass contains 20 to 30 % of moisture content.Samples should be stored at 4˚C for preservation.

Dehydration process

Fig – 4: Oven dried algae sample.

The harvested biomass slurry was dried in an oven at 60˚C for 24h. The dried algae were ground in a coffee grinder until the particles were less than 150 µm. After that the algae were again heated to 100˚C to remove the residual moisture [6].

Lipid extraction (Bligh and Dyer extraction)

Lipids were extracted by mixing methanol, chloroform and distilled water. 10g of algae were taken in an Erlenmeyer flask, 100 ml methanol, 50 ml chloroform, and 40 ml of distilled water were poured into the flask. Then the mixture was shaken for few minutes, after that an additional 50 ml chloroform and 50 ml distilled water were poured into the flask. The mixture was shaken for few minutes, the mixture was immediately filtered to remove algae debris. After that the chloroform layer was evaporated using a rotary evaporator and the mixture was transferred to a separatory funnel to allow separation of the organic and aqueous layers. The weight of extracted lipids was then recorded gravimetrically.Crude lipid yield was calculated by dividing the weight of crudelipid by the weight of dry algae [6].

3. RESULTS AND DISCUSSION

The lipids were extracted by the modified Bligh and Dyer extraction method. The extracted lipids were quantified and it was found,160 ml of lipid from 10 g of dry microalgal powder.The average of Total Volatile Solids present in the photobioreactor sample was 0.11 g/l/d and pH was around 10.0 to 10.3. The CO_2 fixation abilitywas found to be a maximum of 0.313 g/l/d, based on the reduction of CO_2 measured at outlet valves.

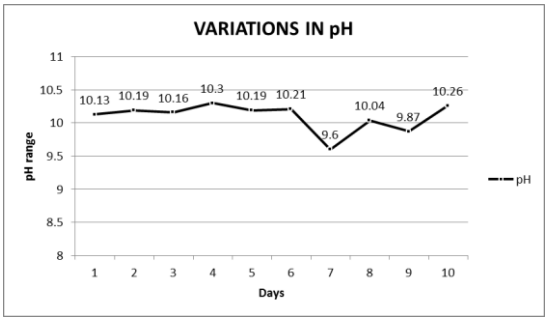

Chart -1: Day by day variations of pH in the reactor sample

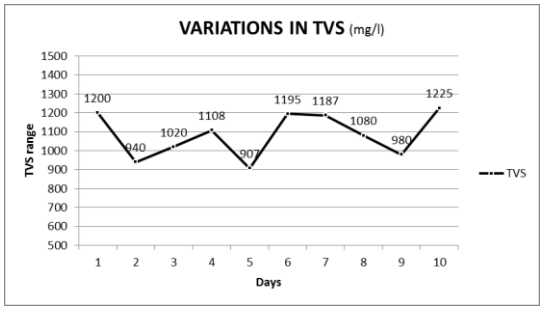

Chart -2: Day by day variations of TVS in the reactor sample

Fig – 5: Extracted lipids from dry microalgal powder

The extracted lipids can be further transesterified by adding 4 ml of BF_3 in methanol to determine the fame content. Transesterification is a process of reducing the viscosity of extracted lipids. The tranesterified final product should be analysed by Gas Chromatograph-Mass Spectrometer [7]. Finally the esterified biodiesel can be tested in light vehicles for performance evaluation.

4. CONCLUSIONS

Apart from several lipid extraction methods, Bligh and Dyer method of extraction has quite easy and more suitable for small scale productions. The experimental result shows that Bligh and Dyer method of extraction has significant advantages over other method of extractions. However, this method has several disadvantages when used on a large scalebecause it generates large quantities of waste solvent and this method is highly toxic.Recycle of organic solvent is costly and it is unsafe to handlein large amounts.

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