

# Solution for Sustainable Development of Biodiesel Production through Microalgae

Ashutosh Kumar Pandey<sup>\*</sup>, Priyanka Singh<sup>\*</sup>, Anita Kushwaha<sup>\*</sup>, Manish Singh Rajput<sup>\*</sup>, Noopur Singh<sup>\*</sup>, Kesav Kishor<sup>\*\*</sup>

#### Abstract

Energy plays a vital role in global economy. The use of fossils as energy source is increasing day by day, however continuous usage of the fossils is hazardous for the future generations due to the accumulation of greenhouse gases in the environment. So we must curb the use of fossils for the energy and fuel production in order to preserve our ecosystem. Another reason is that being a non-renewable energy source, fossils takes millions of years to renew. To overcome this problem we should use such type of energy sources that are renewable and eco-friendly. Renewable and carbon neutral biodiesel are necessary for environmental and economic sustainability. There is also an alternative way of production of biodiesel by microalgae as provide sufficient energy. Demand of biodiesel is constantly increasing as the reservoir of fossil fuel are depleting. However, the question arises on the viability of first generation biofuel production due to its conflict with food supply. Microalgal diesel production appears to be a viable alternative. As a third generation biofuel, microalgal biodiesel has emerged with an enormous amount of economic and environmental benefits. It has been noticed recently that microalgae is the most favorite feedstock for triacylglycerol (TAG), the storage neutral lipid, for renewable and sustainable biodiesel production, due to their comparable lipid contents, faster growth rates and lesser land requirements in comparison to the non-conventional and non-edible oilseed crops. Many microalgae exceed the productivity of oil of the best producing oil crops. In this article the method applied of biodiesel production has been implemented in such a way to increase its production.

Keywords: Algae, Biodiesel, Feedstock, Sustainable, Triacylglycerol.

## Introduction

The world has been confronted with an energy crisis due to depletion of finite resources of fossil fuel in recent years. The global concern over energy and environmental security has thrown many technological challenges before the world's scientific community for driving

<sup>&</sup>lt;sup>\*</sup>Department of Biotechnology, Dr. Ambedkar Institute of Technology for Handicapped, Kanpur-208024. <sup>\*\*</sup>Assistant Director, Future Biosciences Academy and Labs, Kanpur-208024.

*Correspondence to*: Mr. Kesav Kishor, Assistant Director, Future Biosciences Academy and Labs, Kanpur 208024. *E-mail Id:* keshav1016654017@gmail.com

sustainable solutions through the discovery and development of feedstock for alternative green energy. According to annual world primary energy consumption report in 2008, fossil fuels accounted for about 88% of the total primary energy consumption in which oil and coal are the major participant fuels contributing [1] to the generation of greenhouse gases (GHGs) which eventually lead to global warming and climatic changes. With the advent of modern industrialization and ever increasing demand of energy, the paradigm has shifted toward the production of carbon neutral energy sources [2]. Biodiesel attracts considerable attention as an alternative to petroleum diesel, produced by chemically reacting vegetable oil with a short chain alcohol.

During the First generation, vegetable oils are renewable fuels they have become more lucrative recently because of the environmental benefits and the fact that they are procured from renewable resources. Vegetable oils are renewable and potentially inexhaustible source of energy, with energy content close to that of diesel fuel. Global vegetable oil production increased from 56 million tons in 1990 to 88 million tons in 2000. Global consumption rose from 56 million tons to 86 million tons leaving world stocks comparatively tight [3].

Second generation saw the advent of lignocellulosic biomass viz. Stover, Straw, Wood Bagasse, Grass etc in biodiesel production. Lignocellulosic materials are geographically more evenly distributed than the fossil fuels; thus, the sources of energy will, to a larger extent, be domestic and provide security of supply. Lignocellulosic raw materials minimize the potential conflict between land use for food (and feed) production and energy feedstock production. The raw material is less expensive than conventional agricultural feedstock and can be produced with lower input of fertilizers, pesticides, and energy. Biofuels from lignocellulose generate low net greenhouse gas emissions, reducing environmental impacts, particularly climate change.

Third generation research emphasized biodiesel production by microalgae and other type of green algae. The use of aforesaid raw materials can give a solution for an environmental and economic perspective. Microalgae have recently emerged as a better bioenergy feed-stock as compared to conventional energy crops.[4]

Algae can grow practically anywhere there is enough sunshine; some algae can grow in saline water. All algae contain proteins, carbohydrate lipids and nucleic acids in varying proportions, while the percentages vary with the type of algae [5]. The most significant characteristic of algal oil are its yield and hence it is biodiesel yield. According to some estimates, the yield (per acre) of oil from algae is over 200 times the yield from the best performing plant vegetable oil [6], micro algae are the fastest growing photosynthesizing organisms. They can complete an entire growing cycle every few days. Approximately 46 tons oil/hectare/year can be produce from diatom algae. Different algae species produce different amount of oil. Some algae produce up to 50% oil by weight. The production of algae to harvest oil for biodiesel has not been undertaken on a commercial scale. Specially bred mustard varieties can render reasonably high oil yield and have the added benefit that the meal left over after the oil has been pressed out can act as an effective a biodegradable pesticide [7].

Microalgae have significant advantage for biodiesel production, due to their rapid biomass production rate high photosynthetic efficiency and the ability to convert light energy into storage lipid reserve [8]. In microalgae 70% of algal biomass is usable oil's, microalgae able to produce much more biodiesel as compared to other resources of biodiesel. Microalgae produced 5000 to 20,000 gallon of biodiesel per year acre in an open pond culture system. Major environmental factors such as temperature, light humidity affects. Actually the sun light driven cell factories convert morphemic CO<sub>2</sub> into complex value added molecules and carbon rich lipids that, subsequently transform into biofuels, particularly biodiesel, bioethanol and biobutanol without the requirement of any arable land for its cultivation [9]. An amount of biodiesel varies to microalgae species to species. Depending on species, microalgae produce many different kinds of lipids, hydrocarbons and other complex oils [10].

| S. No. | Сгор       | Oil yield (L/ Ha) | Land area needed (M ha) |
|--------|------------|-------------------|-------------------------|
| 1.     | Corn       | 172               | 1540                    |
| 2.     | Soyabean   | 446               | 594                     |
| 3.     | Canola     | 1190              | 223                     |
| 4.     | Jatropha   | 1892              | 140                     |
| 5.     | Coconut    | 2689              | 99                      |
| 6.     | Oil palm   | 5950              | 45                      |
| 7.     | Microalgae | 136,900           | 2                       |

#### Table 1.Comparison of some other sources of biodiesel

Microalgal oils differ from most vegetable oils in being quite rich in polyunsaturated fatty acids with four or more double bonds (Belarbi et al., 2000).

# **Material and Method**

# Sample Collection & Isolation

Green algae were collected from the Motijheel,

Kanpur, the samples were collected in 50ml. vials. The samples were inoculated on agar plates with different screening media composition. The plates were kept then kept in cultured room frequently and analyzed for morphological studies by using microscope. The colonies of Mougeotia species were identified and then isolated and inoculated on a new agar medium.



Figure 1&2.Sample Collection & Isolation

#### **Media Preparation**

Define medium was used for the culture of fresh water algal species of Mougeotia. It is cultured in B9 Media. 1L of defined medium was prepared by mixing Ca  $(NO_3)_4.4H_2O$  (94mg), NaH<sub>2</sub>PO4 (180mg), MgSO<sub>4</sub>.7H<sub>2</sub>O (715mg), CaCl<sub>2</sub>.6H<sub>2</sub>O (416mg), FeSO<sub>4</sub>.7H<sub>2</sub>O (200mg), AIK (SO<sub>4</sub>)<sub>2</sub>.12H<sub>2</sub>O (260mg), Na<sub>2</sub>SO<sub>4</sub> (780mg), MnSO<sub>4</sub>.4H<sub>2</sub>O (45mg) and sodium silicate solution (0.53ml) in distilled water in the following order. pH of medium is ~4.6 using 1N HCL after mixing of compounds.

# **Trans Esterification Process**

Parent oil used in making biodiesel consist of triglycerides in which three fatty acid molecules are esterified with a molecule of glycerol in making biodiesel, tri glycerides are reacts with methanol in a reaction known as Trans esterification or alcoholysis.

Trans esterification produces methyl esters of fatty acid, which are biodiesel, and glycerol.



Trans esterification requires 3 mol of alcohol for each mole of triglycerides to produce one mole of glycerol and three mole of methyl ester the reaction is a equilibrium. Methanol can be used, it is the least expensive to prevent yield loss due to saponification reactions (soap formation).

Alkali catalyzed trans esterification is about four thousand time faster than the acid catalyzed the reaction (Fukuda et al., 2001). Consequently, alkalis such as sodium and potassium hydroxide are commonly used as commercial catalyst at a concentration of about 1% by weight of oil. Alkoxides such as sodium methoxide are even better catalyst than sodium hydroxides and re being increasingly used. Use of lipids offers important advantages, but is not currently feasible because of the relatively high cost of the catalyst (Fukuda et al., 2001).

### **Experimental Procedure**

For 80-100 g of dried algal biomass, 250 ml of n-hexane were used for the lipid extraction. The extraction of lipid was carried out in Soxhlate apparatus. At a time of loading dry algal biomass in extractor of soxhlate apparatus ethanol is poured into the dry algal biomass for wetting and left it for the 10 min, and thenn-hexane was poured in the 2L round bottom flask of the soxhlate apparatus and then it was operated for 240 min. After the completion of the extraction procedure we get dark green lipid sample. After that, alkali catalyzed trans esterification was carried out by giving continues water bath at 50°C under atmospheric pressure, 20% methanol and 1% NaOH by weight of oil. Under these parameters, it takes about 90 min to complete. The reaction occur stepwise triglycerides are first converted to di glycerides, than to mono

glycerides and finally to glycerol. Methanol and oil do not mix and the reaction mixture contains two liquid phases.

The oil and alcohol must be dried and the oil should have minimum free fatty acid content. Left the processed sample with water overnight for recovery of biodiesel and repeated washing 5-6 times with distilled warm water ( $50^{\circ}$ C) to remove glycerol and methanol. By distillation resultant solution was separated from solvent. The solvent was reused in next batch of extraction. Finally sample was dried in oven (50- 60 °C).

## **Research Findings and Analysis**

#### **FAME Analysis**

The fatty acid profiles were procured for the algal sample assayed via GC. The procured sample profile was then compared with the standard sample for identification of the unknown peaks. The concentration and percentage of the fatty acid methyl esters in the lipid sample is listed and analyzed. The algal sample is composed of monounsaturated fatty acid (MUFA), saturated fatty acid (SFA) and polyunsaturated fatty acid (PUFA) in different proportion.

| Table 2.00100111 and percentage of FAME in the algar lipit sample |              |             |                         |              |             |  |  |
|---|--------------|-------------|-------------------------|--------------|-------------|--|--|
| Fatty acid  | Conc (mg/ml) | Percent (%) | Fatty acid2             | Conc (mg/ml) | Percent (%) |  |  |
| C4  | 112.38178    | 98.1629     | C17                     | 4.96E-02     | 0.0434      |  |  |
| C10   | 1.79E-02     | 0.0156      | C17:1                   | 4.79E-02     | 0.0418      |  |  |
| C11   | 3.11E-02     | 0.0272      | C118                    | 5.56E-01     | 0.4857      |  |  |
| C12   | 3.79E-02     | 0.0331      | C18.1                   | 1.22992      | 1.0743      |  |  |
| C14   | 4.56E-02     | 0.0398      | C18:2                   | TANS1.78797  | 1.5618      |  |  |
| C14   | 11.00E-01    | 0.0876      | C18:2                   | CIS1.1714    | 1.0232      |  |  |
| C15   | 6.59E-02     | 0.0575      | C20                     | 2.32E-01     | 0.2029      |  |  |
| C15   | 12.43E-01    | 0.2126      | C20:01                  | 7.09E-02     | 0.0619      |  |  |
| C16   | 2.37E-01     | 0.2071      | C18:1                   | 5.85E-02     | 0.0511      |  |  |
| C16:1   | 1.32397      | 1.1565      | C207.7                  | 3E-02        | 0.0676      |  |  |
|   |              |             | C22:2                   | 5.36E-02     | 0.0469      |  |  |
| TOTAL   |              |             | 114.4849482             |              |             |  |  |
| SFA   |              |             | 94.92%                  |              |             |  |  |
| MUFA  |              |             | 2.51% For aviation fuel |              |             |  |  |
| PUFA  |              |             | 2.51% for biodiesel     |              |             |  |  |

#### Table 2.Concentration and percentage of FAME in the algal lipid sample



Figure 4.Gas chromatogram of sample prepared

Based on analysis from Table 2, it is seen that the lipid has an equal percentage of PUFA and MUFA content. The monounsaturated fatty acid between C12 to C15 are suited for their use as aviation fuels so algal biofuel can be a cheaper source of aviation fuel replacing the current expensive jet fuels.

The monounsaturated fatty acids between C15 to C20 are considered for being used as biodiesel. The high percentage of saturated fatty acid may be a problem at low temperature oil get freezed, for minimizing this problem is needed to to be solved by decreasing the concentration of SFA in the biodiesel.

# Conclusion

Algae are analyzed as the most suitable for the production of biofuel and replacement of the unsustainable source of fuel. But still lot of research and development is needed for the successful use of algal fuel at commercial level, selection and screening of the algal species is done based on the studies of high growth rate and high lipid content and biomass productivity are the few required characteristics for the species selection towards the commercialization of biofuel. Mougeotia species are suited for biofuel production.

After the selection of algal species next step is cultivation. Land, water, & nutrients, should be utilized in a sustainable and cost effective manner.

# References

[1] Brennan L, Owende P. Biofuels from microalgae-a review of technologies for

production, processing, and extractions of biofuels and co-products. *Renew Sust Energ Rev* 2010; 14: 557-77.

- [2] Sorest JP. Climatic Change: Solutions in Sight, a Dutch Perspective. Energy Policy Platform, Delft, 2000.
- [3] Demirbas A. Biodiesel production from vegetable oils via catalytic and noncatalytic supercritical methanol transesterification methods. *Prog Energy Combus Sci* 2005; 31: 466-87.
- [4] FAO. Sustainable bioenergy: a framework for decision makers. United Nations Energy, 2007.
- [5] Becker EW. In: Baddiley J et al. (Eds). Microalgae: biotechnology and microbiology. Cambridge, New York: *Cambridge Univ. Press*, 1994.
- [6] Sheehan J, Dunahay T, Benemann J et al. A look back at the US department of energy's aquatic species programbiodiesel from algae. National renewable energy laboratory (NREL) report: NREL/TP-580-24190. Golden, CO, 1998.
- [7] Demirbas A. New liquid biofuels from vegetable oils via catalytic pyrolysis. *Energy Educ Sci Technol* 2008; 21: 1-59.
- [8] Rasoul-Amini S, Montazeri-Najafabady N, Mobasher MA et al. Chlorella sp: a new strain with highly saturated fatty acids for biodiesel production in bubblecolumn photo bioreactor. *Appl Energy* 2011; 88: 3354-56.
- [9] Christy Y. Biodiesel from microalgae. *Biotechnology Adv* 2007; 25: 294-306.
- [10] Banerjee et al., 2002; Metzger and Largeau, 2005; Guschina and Harwood, 2006.