**6. Biodiesel Production using *Chlorella Vulgaris***

Microalgae are used to treat textile wastewater and other bio refinery applications such as human food, animal feed, cosmetic products, pharmaceutical products, fertilizers and biofuels [1-2]. Biofuels are recognized as renewable energy since its precursors are derived from biodegradable resources such as corn kernels, sugar cane, sweet sorghum, cassava, woody biomass, vegetable oils, microalgae, waste cooking oil and etc. [3-4]. Bioethanol is produced from sugar cane, cassava flour, corn starch [5-7] whereas biodiesel is generated from oil crops [5] or waste cooking oil [8-9]. Due to the food for fuel dilemma, non-food crops and aquatic microorganisms such as microalgae have been proposed as an alternative feedstock for biodiesel production, whereas lignocellulosic biomass [10] are highly used in the bioethanol production [11-12]. Biodiesel is a clean renewable source of energy because it can be utilized in any compression ignition engine without any requirement of modification [13]. Biodiesel is a mixture of long chain fatty acid methyl esters (FAME) and is derived by the transesterification of lipids (Fig. 1).

Microalgae

Microbial Fuel Cell

Thermochemical conversion

Biochemical Conversion

Transesterification

Acid/Base Catalysis

Anaerobic Digestion

Fermentation

Supercritical fluid

Photo-biological Hydrogen production

Biodiesel

Bioelectricity

Biomethane/Biogas

Bioethanol

Bio hydrogen

**Fig 1:** Microalgae biomass conversion processes into biofuels [14]

Biodiesel can also be derived from nontoxic and biological resources such as vegetable oils, animal fats, used cooking oil and algal oils. Biodiesel yield can be obtained about 80% of the

Volume of extracted algal oil [15]. Biodiesel from microalgae oil has received significant attention recently as it is renewable, environmentally friendly and represents the ability to convert CO2 to oil Microalgae based biofuels can be divided into four categories based on conversion technologies (thermo-chemical conversion, biochemical conversion, transesterification and photosynthetic microbial fuel cell). Microalgae are photosynthetic microorganisms that needs CO2, water and nutrients (e.g. nitrogen, phosphorus and potassium) for their growth. The produced microalgae biomass contains a significant amount of lipid in the form of fatty acids, which can be extracted for subsequent biodiesel production. Microalgae do not require much cultivation land as compared to vegetable plants; however, it proves a better feedstock for biodiesel. Microalgal biodiesel is the most important option because it possesses similar physical and chemical characteristics to petroleum as shown in table.

|  |  |  |  |
| --- | --- | --- | --- |
| **Source** | **Crop Name** | **Oil yield**  **(L ha−1)** | **Biodiesel productivity**  **(kg biodiesel/ ha-year)** |
| First generation | Corn  Soybean  Canola  Sunflower  Palm oil | 172  446  1190  1070  5366 | 152  562  862  946  4747 |
| Second  generation | Jatropha  Castor | 741  1307 | 656  1156 |
| Third generation | Microalgae (wet biomass)  Microalgae (dry biomass) | 58,700  136,900 | 51,927  121,104 |

**Table 2. Biodiesel production of different feedstock** [16, 17, 18].

The nutrients supply and medium composition impact the quality and quantity of microalgae-biodiesel [19]. The two major processes used for the production of biodiesel by *Chlorella Vulgaris* is lipid extraction and transesterification. For lipid extraction, harvested biomass is used for biodiesel production. Harvested microalgae contains lipid, carbohydrates, proteins and other valuable products [20]. Triglycerides (TG) and free fatty acids (FFA), glycolipids and phospholipids can be extracted from *Chlorella Vulgaris.* Lipids can be converted into biodiesel through transesterification [21]. The residual microalgae after lipids extraction can further be used to produce methane gas from anaerobic digestion. The lipids extracted are converted into biodiesel through transesterification process. Lipids react with an alcohol (methanol, ethanol, propanol, butanol and amyl alcohol) to form fatty acid methyl esters (FAME) and glycerol as shown in **Eq.1**

CH2-O-CO-R1 (Catalyst) CH2-OH R-O-CO-R1

CH-O-CO-R2 +3ROH CH-OH + R-O-CO-R2

CH2-O-CO-R3 CH2-OH R-O-CO-R3

**Triglyceride Alcohol Glycerol FAMEs**

Generally, chemical (acids, bases) or biological (enzymes) catalysts are used in transesterification reaction [23]. Transesterification reaction is reversible therefore; excess alcohol supply is used to ensure the forward reaction. Biodiesel is obtained by mixing an alcohol with TAG in a 3:1 M ratio. The transesterification reaction is carried out in various steps. TAG reacts with alcohol and stepwise gets converted into diglycerides (DG) which further converts into monoglycerides (MG). Finally, monoglycerides liberate one mol of FAME (biodiesel) at each step and glycerol at the end as shown in the following Eq 2 [22].

**Eq.**

TG +R’OH DG+R’COOR1

DG +R’OH MG+R’COOR2

MG+R’OH Glycerol + R’COOR3

R1, R2, R3 are long chain hydrocarbons also known as fatty acid chains [23]. There are six types of fatty acid chain in triglyceride of *Chlorella Vulgaris* such as palmitic, palmitoleic, stearic, oleic, linoleic, and linoleic acids. Several factors account for biodiesel production such as reaction temperature, reaction time, conc. of alcohol and catalyst. For the production of biodiesel *Chlorella vulgaris* is a suitable candidate because the lipid profile, lipid composition and level of unsaturation meets requirement of oil suitable for biodiesel production.

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