



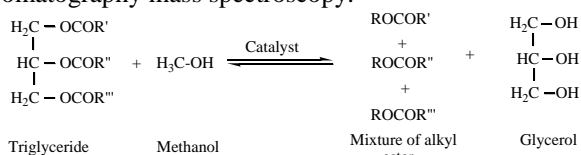
## 2.2 Drying of the Macro green Algae

The Sun-drying is the important method for drying of macrophytes [2] [12] [31]. This method save the fossil fuel but it depends on climatic condition and volume of biomass. Sun-drying in tropical region may take 2-3 days in sunny weather and may take up to 7 days in rainy seasons [4].

## 2.3 Biodiesel synthesis by In-situ Transesterification Method

The “In-situ transesterification” is “reactive extraction” process for synthesis of fatty acid methyl ester. Biodiesel content obtained from algae powder without extraction of oil. The alcohol acts as an extraction solvent and extraction reagent which enhance the product yield. These reactions are often catalyzed by the addition of an acid or base catalyst. Transesterification is three steps reaction while In-situ transesterification is one step reaction (Figure 1), there is oil extraction step skips hence saving the solvent like hexane as well as reduce the atmospheric pollution and global warming [26]. According to Hass and Wagner (2011) [18], In-situ transesterification method has high heating value with higher product yield. It is quick and simple operation process without loss of lipid. This method reduces the process cost due saving solvent, absence of extraction process and it reduce the waste water pollutants.

The base catalyzed reaction according to Dalvi et al. (2012) [8] and methanolic KOH catalyst used. The mixture of KOH and MeOH was added in round bottom containing dried macro-algae powder. The reaction mixture was continuously stirring. The reaction carried out at 60°C for 60 minute. At room temperature reactions mixture was cooled and separates the algal remnant and product. The product was wash water to remove water soluble impurities [32] and heat the product at 85°C. The fatty acid methyl esters were analyzed by standard method Infrared spectroscopy and gas chromatography mass spectroscopy.



**Figure 1:** Transesterification reaction of the Triglyceride.

## 3. Results and Discussion

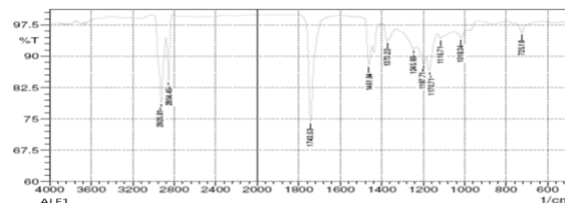
### 3.1 Characterization of Isolated Algae Sample

The isolated algae were belonging from the division of Chlorophyta. The order of Volvocales, Tetrasporales, Chlorococcales, Chlorosacinales, Ulotriches, Sphaeropleales, Charophorales, Trentopohiales, Oedogoniales and Ulvales species were observed.

### 3.2 Infrared Spectroscopy Analysis

The IR Spectroscopy is the method to determine the function group present in the obtained product. The macro-algae fatty acid methyl esters are analyzed by infrared spectroscopic method (Figure 2). Infrared Spectrum of Base catalyzed In-situ transesterification of Macro-algae Biodiesel shows the  $\nu$

as ( $\text{sp}^3$  C-H) stretching of ester group of fatty acid methyl ester at  $2925.51 \text{ cm}^{-1}$ ,  $\nu$  (C=O) stretching of ester at  $1743.63 \text{ cm}^{-1}$ ,  $\delta_s$  (CH<sub>2</sub>) bending of methylene group at  $1461.94 \text{ cm}^{-1}$ ,  $\delta_s$  (CH<sub>3</sub>) bending of methyl group absorption frequency at  $1373.22 \text{ cm}^{-1}$ ,  $\nu$  (C-O) stretching of alkyl carbon and oxygen from fatty acid methyl ester at  $1170.71 \text{ cm}^{-1}$ ,  $\delta_r$  (CH<sub>2</sub>) bending of methylene group was observed at  $726.10 \text{ cm}^{-1}$ .



**Figure 2:** FT-IR Spectrum of Macro-algae Biodiesel

### 3.3 Gas Chromatography Mass Spectroscopy Analysis

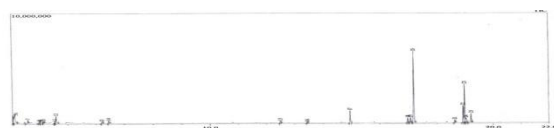
Gas chromatography mass spectroscopy is used to separate and identify the chemical ingredients in the biodiesel. The Figure 3 shows Gas chromatographic spectrum with the various peak obtained at different retention time. In Base catalyzed Biodiesel of Macro-algae, seven types of ester obtained at retention time (min) at 12.486, 14.952, 17.071, 17.190, 18.922, 18.976 and 19.201. Mass spectrum fatty acid methyl ester of energy liquid fuel shown in figure 4, 5, 6, 7, 8, 9 and 10 respectively. The four components of biodiesel at retention time (min.) 12.486, 14.952, 17.190 and 19.201 shows the base peak at  $m/z$  74.05 [Figure 4, 5, 7, 10]; the two components at 17.071 and 18.976 min. shows base peak at  $m/z$  55 [Figure 6, 9], while one component at 18.922 shows the base peak at  $m/z$  67 [Figure 8]. The experimental test results and the ester were confirmed with MS library. Table 1 shows the peak obtained with various retention times, percentage area, name of the obtained compound and molecular formula.

The Macro-Green Algae Biodiesel contain higher content of Hexadecanoic acid methyl ester (29.15%), 9-Octadecenoic acid (Z) methyl ester (18.63%), 9, 12-Octadecadienoic acid(Z,Z) methyl ester (8.12%) and in small content of Tetradecanoic acid methyl ester (4.94%), 9-Hexadecenoic acid methyl ester (4.23%), Octadecanoic acid methyl ester (4.04%), Dodecanoic acid methyl ester (1.01%).

## 4. Conclusions

The present paper was study the macro green algae Chlorophyta species found in Ahmednagar district of Maharashtra State, India. The biodiesel energy liquid fuel of macro green algae was synthesis by In-situ transesterification Method. The obtained product biodiesel liquid fuel is characterized by FT-IR Spectroscopy and Gas Chromatography Mass Spectroscopy. The FT-IR shows the characteristic peak of ester at  $1743.63 \text{ cm}^{-1}$ . The component of biodiesel separated by Gas Chromatography and identify with standard data MS library. The base catalyzed biodiesel of macro algae biodiesel contain mixture of 30.98% unsaturated fatty acid methyl ester and 38.24% saturated fatty acid methyl ester. The fatty acid obtained was ranging between C13 to C19.

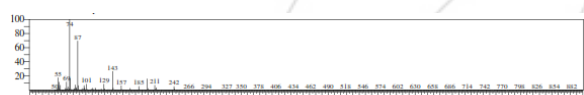
The macro-algae is a challenging material for biodiesel production due to its higher content of polyunsaturated fatty acids and unsaturated fatty acids [1] [9] [16], But the best solution to decrease polyunsaturated fatty acid is drying [27]. The GC-MS study (Figure 3) state that the biodiesel from macro green algae contains mixture of saturated fatty acid unsaturated fatty acid methyl esters, hence its oxidative stability is higher. Thus, Macro green algae can be considered as potential algae for biodiesel production because of their large availability and easy harvesting. It could be help for innovation in local market, to create job opportunity, sustainability and energy security of the nation.



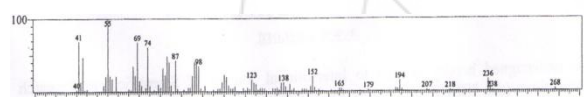
**Figure 3:** Gas Chromatographic spectrum of Macro-algae Biodiesel.



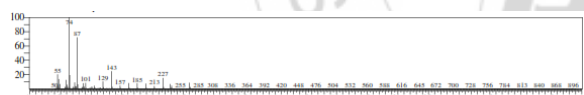
**Figure 4:** Mass Spectrum of Dodecanoic acid methyl ester



**Figure 5:** Tetradecanoic acid methyl ester



**Figure 6:** 9-Hexadecenoic acid, methyl ester (Z)



**Figure 7:** Hexadecanoic acid, methyl ester



**Figure 8:** 9, 12-Octadecadienoic acid (Z, Z) methyl ester



**Figure 9:** 9-Octadecenoic acid (Z) methyl ester



**Figure 10:** Octadecanoic acid methyl ester

**Table 1:** Macro-algae Biodiesel Components with Retention time, Percentage area, Name of the Ester Compound and Molecular Formula

Sr. No.	Retention Time (min.)	% Area	Name of the Compound	Molecular Formula
1	12.486	01.01	Dodecanoic acid methyl ester	C <sub>13</sub> H <sub>26</sub> O <sub>2</sub>
2	14.952	04.94	Tetradecanoic acid methyl ester	C <sub>15</sub> H <sub>30</sub> O <sub>2</sub>
3	17.071	04.23	9-Hexadecenoic acid methyl ester	C <sub>17</sub> H <sub>32</sub> O <sub>2</sub>
4	17.190	29.15	Hexadecanoic acid methyl ester	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>
5	18.922	08.12	9,12-Octadecadienoic acid(Z,Z) methyl ester	C <sub>19</sub> H <sub>34</sub> O <sub>2</sub>
6	18.976	18.63	9-Octadecenoic acid (Z) methyl ester	C <sub>19</sub> H <sub>36</sub> O <sub>2</sub>
7	19.201	04.04	Octadecanoic acid methyl ester	C <sub>19</sub> H <sub>38</sub> O <sub>2</sub>

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