

# GC-MS and FTIR analysis of bio-oil obtained from freshwater algae (spirogyra) collected from Freshwater

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**Abstract**— Algae are gaining broad consideration as a substitute renewable source of biomass for the manufacture of bioethanol, due to this reason categorized under the “third generation biofuels”. In this work, GC-MS analysis and FTIR has been done of bio-oil obtained from fast pyrolysis of Freshwater Algae (Spirogyra) in this paper we have shown a simple process of converting biomass of fresh water algae to bio-oil through pyrolysis and explained it with the help of graphs and tables. Pyrolysis is a thermal process for converting various biomasses, residues and wastes to produce high-energy-density fuels (bio-oil, biochar). The bio-oil was obtained in two step pyrolysis in which temperature of the system kept 25°C and then increased up to 650°C time by time. After pyrolysis these fractions were analyzed by gas chromatography/mass spectrometry (GC-MS) and FTIR which show different peaks and data of different compounds and functional groups present in this bio-oil

**Keywords**— Biomass, Bio-oil, Freshwater Algae, Pyrolysis, GC-MS, FTIR

## I. INTRODUCTION

Recently biodiesel are getting more and more importance as a engine fuels and one of the most promising alternative energy. biodiesel or bio-oil are becoming more attractive for the users in all over the world because of their Usefulness for the people and environment around us, Biodiesel is an oxygenated fuel consisting of long chain fatty acid which contain 10–15% oxygen by weight to find and develop an alternative energy is a major issue to our environment due to the depletion of fossil fuel [1,2,3]. Among many kinds of alternative energies, biodiesel can be directly applied to nowadays diesel engines with benefits low pollutions, high environmental friendliness and sustainability. and it contains neither sulfur, nor aroma. These facts lead biodiesel to enhance more complete combustion and less emission of particulate matter. [4] Recently, catalytic pyrolysis has aroused a great interest for the advantages of operating at atmospheric pressure and the lack of need for hydrogen [5], which has been demonstrated by many researches. The experiments on catalytic pyrolysis of biomass were generally carried out in a fixed bed reactor or fluidized bed [6, 7] On the other hand biodiesel has higher molecular weight, density, viscosity and pour point than conventional diesel fuel [8,9]. Higher molecular weight and viscosity of biodiesel causes low volatility and poor fuel atomization, injector coking, piston ring sticking and leading incomplete combustion [10] as well as it has cold flow property which is a barrier to use it in cold or chill weather [11] anyhow the best benefit of Bio-oils is that they are preparing from renewable sources like corpse, plants, trees and residues etc. approximately 100 years ago, Rudolf Diesel tested Bio oil as the fuel for his engine that was available with him [12,13] Soybean oil was transesterified into ethyl and methylesters, and comparisons of the performances of the fuels with diesel were made. [14,15]. Also, methylesters have been prepared from palm oil by transesterification using methanol in the presence of a catalyst (NaOH) or (KOH) in a batch reactor [16] Ethanol is a preferred alcohol in the transesterification process compared to methanol because it is derived from natural agricultural products and is renewable and biologically less objectionable in the environment. The success of rapeseed ethylester production would mean that biodiesel's two main raw materials would be agriculturally produced, renewable and environmentally friend [17]

Methyl, ethyl, 2-propyl and butyl esters were prepared from canola and linseed oils through transesterification using KOH and/or sodium alkoxides as catalysts. In addition, methyl and ethylesters were prepared from rapeseed and sun flower oils using the same catalysts [18,19] Biomass is a renewable source which has received attention due to various characteristics, particularly its low cost and wide availability. Biomass can be converted into bio-fuel by means of different processes, e.g., reductive combustion, liquefaction, pyrolysis and gasification [20]. The use of biomass is particularly interesting when it involves waste products such as waste vegetable oil, fruit seeds, sugarcane bagasse, sugarcane straw, rice husks, coconut fibers, and coffee grounds, which are also potential sources of energy [21]. Taking into account that bio-oil from pyrolysis contains a very complex mixture of products, its chromatographic analyses requires the coupling of these instruments with a mass selective detector to aid the identification of eluted products [22]. The mass spectrum, sometimes, is not sufficient to

identify unknown compounds, due to the similarity of their spectra, thus the use of reference compounds may be an aid to overcome this problem. Furthermore, the use of analytical standards may allow quick quantification through one of several techniques, such as the internal standard method [23,24]. However bio-oils from pyrolysis may include up to several hundred of compounds [25,26,27].

## II. EXPERIMENTAL

### 2.1 Biomass of Spirogyra (freshwater algae) and other reagents

The Spirogyra biomass was first converted to powder form then it was dried in fresh air and sunlight for more than 96 hours until 10% moisture remained, the fine powder spirogyra was introduced into a plastic jar and one liter of tap water was added to it which mixed well. the plastic jar was closed through lid in such a way that it became air tight. the jar was kept at room temperature after 7 days,. Then everyday water was sprayed on its surface and mixed well, this treatment was maintained for 14 days then the sample was dried and used as biomass sample, after this process this biomass was introduced to pyrolysis system in the presence of catalyst calcium oxide(CaO).

### 2.2 Production of bio-oil from Algal biomass by pyrolysis

The bio-oil was produced from the pyrolysis of spirogyra (freshwater algae). A round block shape structure of sample was made inside the filter paper from biomass ( filter paper as side wall of the sample block to keep the biomass tight) from the spirogyra biomass while the weight of this sample is kept 120 grams also some amount of sand was added to the mixture for some purposes(as heat resistant), after preparation of this sample block it was kept inside a stainless steel chamber of pyrolysis system which is further connected to other condensation chambers which are shown in diagram in figure 1.

The temperature of chamber which has biomass was increased from 25c to 650c, through which biomass was converted to biogas and then the biogas was condensed in condensation chamber which condensed fractions of biogas to bio-oil. The condensed fractions from this chamber were collected introduced to fractional distillation. The pyrolysate obtained at the optimum conditions was separated into fractions using distillation. It was carefully watched to accurately separate the fractions. The pyrolysate was also separated into four fractions including residue. These fractions were found in the temperature range of 65-200<sup>0</sup>C and the residue of distillation. The first fraction was obtained in the temperature range of 65<sup>0</sup>C to 100<sup>0</sup>C, the second fraction was collected in the range 100<sup>0</sup>C -150<sup>0</sup>C, the third fraction was found in the range of 150<sup>0</sup>C to180<sup>0</sup>C,the fourth fraction was at 180<sup>0</sup>Cto 200<sup>0</sup>C .The first fraction is flammable while the remaining all fractions are nonflammable. However the residue of the fractional distillation was a sticky organic material and believed as combustible. That's why it was paralyzed. The product of this pyrolysis was introduced to second step catalytic pyrolysis.

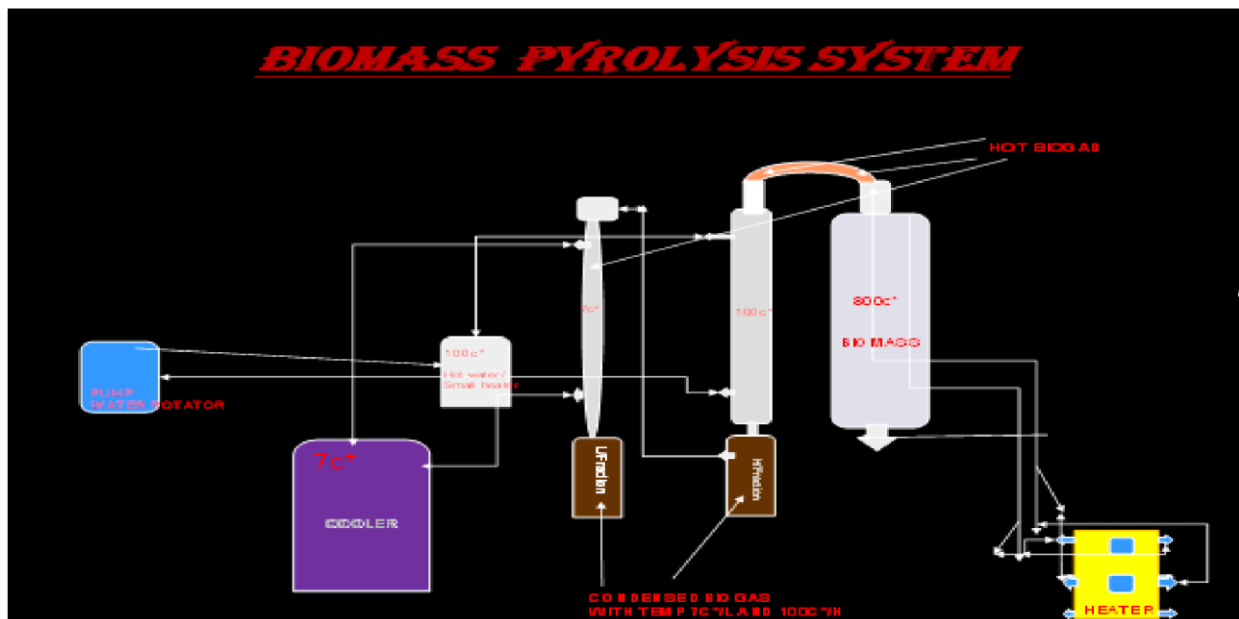
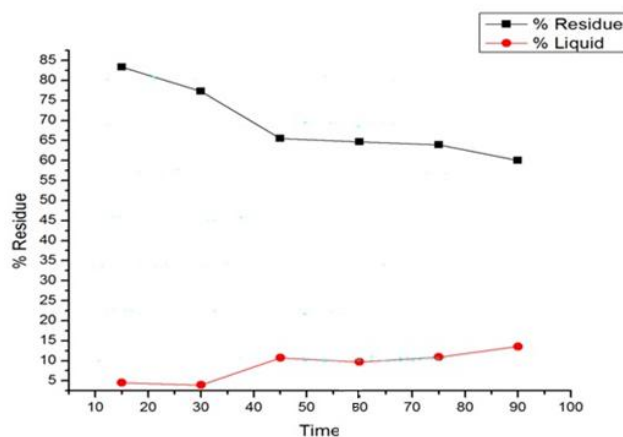


Fig 1, biomass pyrolysis system.

**TABLE 1**  
**IMPORTANT DATA OF TEMPERATURE FOR PYROLYSIS OF FRESHWATER ALGAE:**

S/no	Temperature	Weight of biomass (g)	% Residue	% liquid	% Gases
1	250	120	92.89	7.01	0.103
2	350	120	70.23	20.11	9.66
3	450	120	62.77	25.99	11.24
4	550	120	57.10	26.97	15.93
5	650	120	53.10	34.16	12.84

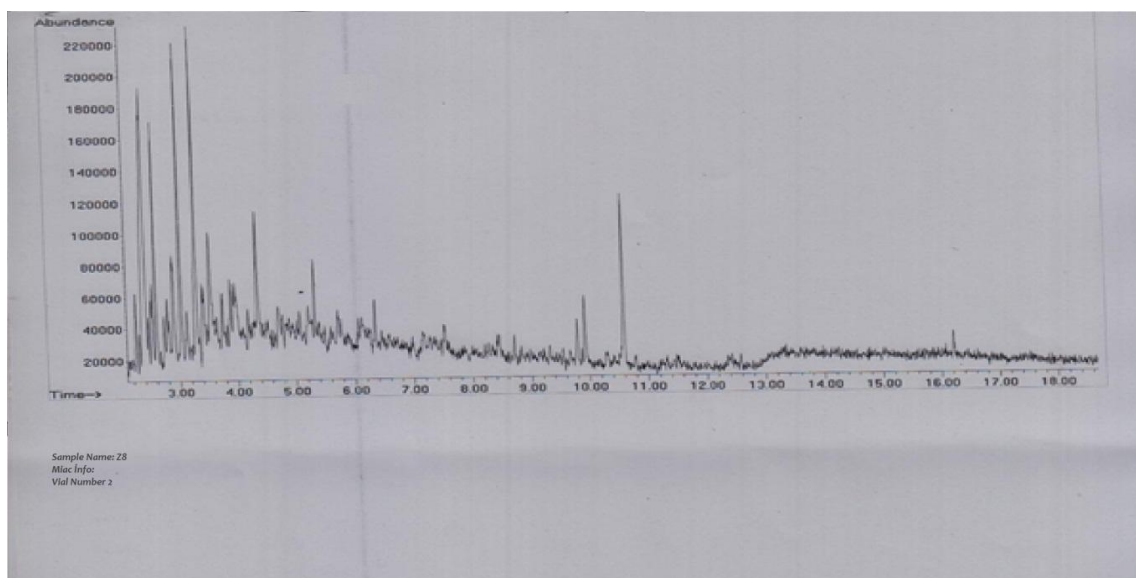


**FIG 2. BELOW GRAPH SHOWS THE RELATIONSHIP BETWEEN RESIDUES AND TIME.**

### 2.3 GC/MS CHROMATOGRAM OF THE FRESH BIOMASS FUEL AFTER 2<sup>ND</sup> STEP PYROLYSIS.

The GC/MS analyses were carried out in order to find out the hydrocarbons present in the fuel. The different biomass fuels which were obtained by second step pyrolysis was analyzed by GC/MS. Optimum pyrolysis temperature was 500<sup>0</sup>C. The main peaks identification of compounds has been performed by using NIST mass spectral data base. The samples of spirogyra biomass fuel which obtained after 2<sup>nd</sup> step pyrolysis were complex mixture and contain different class of hydrocarbons which are shown in the table 2.

After second pyrolysis was introduced to GC-MS analysis, where GC-MS had following peaks and results , as shown down in figure 3.



**FIG 3, GC/MS CHROMATOGRAM OF THE FRESH BIOMASS FUEL AFTER 2<sup>ND</sup> STEP PYROLYSIS.**

**TABLE 2**  
**GC/MS SHOWS THE COMPOUNDS PRESENT IN FRESH BIOMASS FUEL SAMPLE AFTER SECOND STEP PYROLYSIS.**

S/NO	Name of compounds	Retention Time	% composition
1	3,4,5- Tri-methyl pyrazole	2.979	20.086
2	2-pyrrilidinone	2.413	7.46
3	1-H imidazole1,2,4,5-tetramethyl	3.431	16.35
4	2-H imidazole,2,4,5-tetramethyl	3.531	15.12
5	2,3,4-Trimethyl-d-xylose	3.93	17.79
6	Pentadecane	6.218	5.36
7	Benzonitrile, 4-methyl	4.358	8.60
8	2-Hexadecene3,7,11,15-tetramethyl	9.926	5.73
9	Benzenamine,N-(1-methyl-2-propyl)	6.350	4.59
10	Hexadecanenitrile	10.595	14.06

Table 2. is the fresh biomass fuel table which contain different hydrocarbons are shown. The sample GC/MS result indicate the presence of different classes of hydrocarbons which includes azole, aromatic amines and nitriles, carbohydrate, alkenes, and alkane. The most abundant compound 2, 3, 5-trimethyl pyrazole 20.086 % accounted for the total fuel. The azoles including 3, 4, 5-trimethyl-pyrazole, pyrrolidinone etc and aromatic amine and nitrile these compounds were assumed to be derived from protein degradation. While the 2, 3, 4-trimethyl-d-xylose derived from the hydrolysis of cellulose. The alkenes class compound 3, 7, 11, 15-tetramethyl-2-Hexadecene is probably generated from the conversion of unsaturated fatty acid in algal cell. Long chain compound Pentadecane accounted 5.36 % correspondingly of the total fuel. Which contributes better combustion property of the algal fuel?

### III. RESULTS AND DISCUSSION

#### 3.1 Theoretical bases of the pyrolysis of Biomass/ Spirogyra.

Spirogyra is a cellulosic biomass. Spirogyras is blue green algae and consider as third generation bio-fuel its use for the preparation of liquid and gaseous fuel is attractive. It contains small quantity of the lignin. Its growth and multiplication is very rapid. The pyrolysis of the cellulosic biomass gives water soluble oxygenated compounds. It is thought that the cellulosic biomass may manufacture methanol, anhydrosugars, furans and aldehydes compounds. These can be used as fuel if appropriately utilized. These products are formed along with large quantity of water therefore need treatment preceding to use as fuel. The product of pyrolysis also contain oil like liquid which is believed as composed of compounds containing less oxygen contents. These are partly soluble in the aqueous mixture. In this work major focus is on this product. Attempts were made to increase the amount of this product. This product was separated and further pyrolysed and converted into fuel oil. Different pretreatment procedures were used to increase the oily products and to decrease the oxygen containing compounds.

### 3.2 Infrared spectroscopy (IR) results , FTIR spectrum and Characteristics IR Absorption frequencies of organic functional groups of fresh biomass oil.

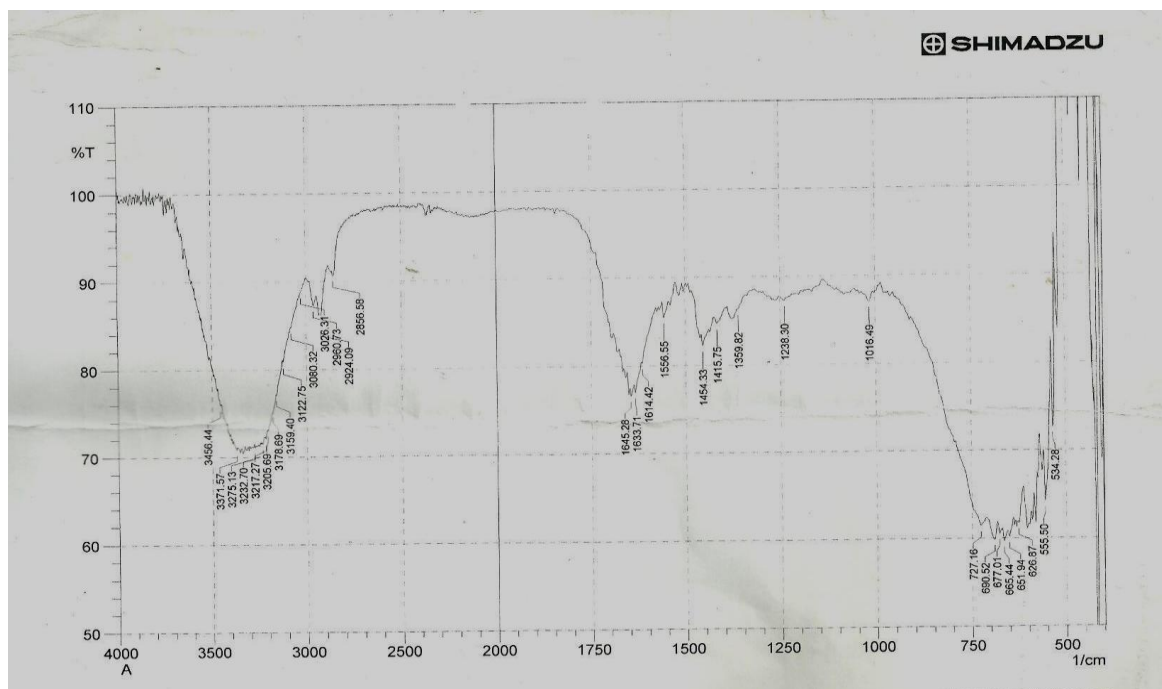


FIG. 4. FTIR SPECTRUM OF FRESH BIOMASS OIL SPECTRUM

TABLE 3  
CHARACTERISTICS IR ABSORPTION FREQUENCIES OF ORGANIC FUNCTIONAL GROUPS OF FRESH BIOMASS OIL.

Functional	Type of vibration	Characteristics Absorption (Cm <sup>-1</sup> )	Intensity
C-O (Ether)	Stretch	1238.30	Strong
-C-H (Alkane)	Bending	1359.82	Variable
C=C (Aromatic)	Stretch	1556.55	Medium-weak, multiple bands.
C-H (Alkane)	Stretch	2924.09	Strong
=C-H (Alkene)	Stretch	3080.32	medium
O-H (Alcohol)	Stretch (H-bonded)	3456.44	Strong, broad
Alkene	Stretch	1645.28	Variable

### 3.3 Infrared spectroscopy (IR) result of fresh biomass oil sample.

The table 3. result of fresh biomass oil sample indicates that (C-H) Alkane stretching at 2924.09 cm<sup>-1</sup> and intense broad peak and axial at 3000 cm<sup>-1</sup>. While alkane bending at 1359.2 cm<sup>-1</sup> occur and the peak intensity is variable. The (O-H) is also a weak broad peak is due to the presence of water content in bio-oil or may be due to alcohol which appear at 3456.44 cm<sup>-1</sup>. The strong intensity due to stretching vibration of ether appear at 1238.30 cm<sup>-1</sup>. The aromatic C=C stretching at 1556.55 cm<sup>-1</sup> which intensity is medium weak multiple band The alkene =C-H stretching at 3080.32 cm<sup>-1</sup> which intensity is medium while the alkenes stretching occur at 1645.28 cm<sup>-1</sup> and the intensity of peak is variable. It was concluded that the presence of different functional group of compounds indicate the presence of hydrocarbons in the fuel and also the (O-H) broad peak indicate the presence of water contents in bio-fuel. The broad O-H peak was due to the interaction (hydrogen bonding) among the water contents. This oil sample consist of Alkane, Alkenes', ether, alcohol, aromatic, the presence of this functional group indicate the presence of hydrocarbons in the bio-fuel.

### 3.4 Infrared spectroscopy (IR) results, FTIR spectrum and Characteristics IR Absorption frequencies of organic functional groups of residue of fractional distillation of fresh biomass pyrolysate.

Fig 5. FTIR Spectrum of the fuel sample which obtained after the second step pyrolysis of the residue of fractional distillation of fresh biomass pyrolysate.

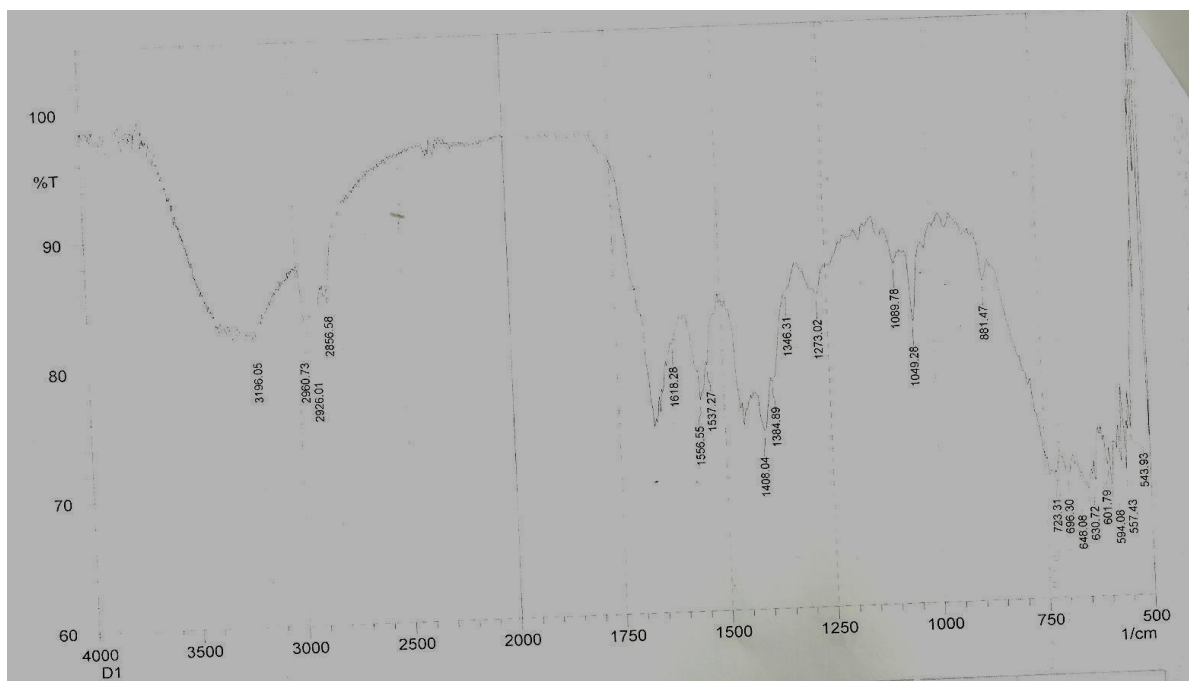


FIG 5. FTIR SPECTRUM OF THE FUEL SAMPLE

TABLE 4

CHARACTERISTICS IR ABSORPTION FREQUENCIES OF THE ORGANIC FUNCTIONAL GROUPS OF THE FUEL SAMPLE WHICH OBTAINED AFTER THE SECOND STEP PYROLYSIS OF THE RESIDUE OF FRACTIONAL DISTILLATION OF FRESH BIOMASS PYROLYSATE.

Functional group	Type of vibration	Characteristics Absorption ( $\text{Cm}^{-1}$ )	Intensity
C-O (Ether)	Stretch	1273.02	Strong
-C-H (Alkane)	Bending	1384.89	Variable
C=C (Aromatic)	Stretch	1537.27	Medium-weak,
C-H (Alkane)	Stretch	2926.01	Strong

### 3.5 Infrared spectroscopy (IR) result of the fuel sample which obtained after the second step pyrolysis of the residue of fractional distillation of fresh biomass pyrolysate.

The residue sample 4.6.1 table indicates that (C-H) Alkane stretching at  $2926.01 \text{ cm}^{-1}$  and intense broad peak and axial at  $3000 \text{ cm}^{-1}$ . While alkane bending at  $1384.89 \text{ cm}^{-1}$  occur and the peak intensity is variable. The strong intensity due to stretching vibration of ether appears at  $1273.02 \text{ cm}^{-1}$ . The aromatic C=C stretching at  $1537.27 \text{ cm}^{-1}$  which intensity is medium weak multiple band.

The table 4.6 functional group belongs to the residue while 4.6.1 table functional group belongs to the fuel which obtained from the residue of fractional distillation of fresh biomass pyrolysate through second step pyrolysis at optimum condition. The advantage of second step pyrolysis was to convert large quantity of oxygenated compound into small quantity of oxygenated compound. When catalyst was again mixed with residue in order to carry out pyrolysis reaction called second

step pyrolysis. The again used of catalyst in order to facilitate cracking, dehydrogenation and dehydration of the pyrolysis product. Due to this reason when we compared the two table 4.6 and 4.6.1 there is a difference because the some functional group which present in table 4.6 disappeared in 4.6.1 this is due to second step pyrolysis. The disappearance or appearance of functional group was due to the conversion of one class of compound into another class of compound due to dehydrogenation, dehydration and cracking during pyrolysis. After the second step pyrolysis the fuel obtained and analyzed through GC/MS the fresh biomass fuel which obtained through second step pyrolysis of the residue of fractional distillation of fresh biomass pyrolysate contained long chain alkane, alkenes, heterocyclic nitrogen containing compounds which belongs to azole class, aromatic and carbohydrate class compounds.

### 3.6 Catalytic optimization

To choose a suitable ratio of catalyst with biomass at which maximum liquid oil while less amount of % residue is obtained. For this purpose numerous reaction were carried out in which the amount of catalyst was remain unchanged while biomass was varying. At different ratio of catalyst and biomass the pyrolysis reaction was carried out. Finally 1:10 of catalyst and biomass respectively was found proper for good product yield of pyrolysis.

## IV. CONCLUSION

Spirogyra as a biomass for bio-oil production was found to have noticeable and unignorable effect on the products of pyrolysis. The comparative amounts of the gases and bio-oil product was found to enhance with pretreatment. In addition to the pretreatment the catalyst (CaO) was found more efficient in increasing the amount of the bio-oil product. The amount of the oily product was found to increase with change in the amount of catalyst and temperature with respect to biomass. with different temperature different ratio of Residues, bio-oil and biogas was experienced, as the temperature increased the residues decreased while product of pyrolysis(bio-oil)was also increased with temperature. The most favorable amount for the formation of highest amount of the oily product was 47.1%. After second step pyrolysis of the residue of the fractional distillation of microbial treated biomass pyrolysates. Studies of The GC-MS peaks showed that the obtained bio-oil was mostly composed of hydrocarbons but also have many other compounds and our future work will be extraction important compounds from this bio-oil and will compare freshwater algae biomass, earth buried algae biomass and microbial treated algae biomass.

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## REFERENCES

- [1] Veses RC, Shah Z, Kuamoto PM, Caramão EB, Machado ME, et al. (2015) Bio-Oil Production by Thermal Cracking in the Presence of Hydrogen. *J Fundam Renewable Energy Appl* 5:194. doi:10.4172/2090-4541.1000194
- [2] Shahabuddin M, Kalam M, Masjuki H, Bhuiya M, Mofijur M. An experimental investigation into biodiesel stability by means of oxidation and property determination. *Energy* 2012;44:616–22.
- [3] Puhan S, Vedaraman N, Sankaranarayanan G, Ram BVB. Performance and emission study of Mahua oil (madhuca indica oil) ethyl ester in a 4-stroke natural aspirated direct injection diesel engine. *Renew Energy* 2005;30:1269–78
- [4] H.-M. Yang et al., Separation of phenols and ketones from bio-oil produced from ethanolysis of wheat stalk, *Separ. Purif. Technol.* (2015), <http://dx.doi.org/10.1016/j.seppur.2015.03.032>
- [5] S.R. Wang, Y.R. Wang, Q.J. Cai, X.Y. Wang, H. Jin, Z.Y. Luo, Multi-step separation of monophenols and pyrolytic lignins from the water-insoluble phase of biooil, *Sep. Purif. Technol.* 122 (2014) 248–255.
- [6] A.S. Pollard, M.R. Rover, R.C. Brown, Characterization of bio-oil recovered as stage fractions with unique chemical and physical properties, *J. Anal. Appl. Pyrolysis* 93 (2012) 129–138.
- [7] C. Amen-Chen, H. Pakdel, C. Roy, Separation of phenols from Eucalyptus wood tar, *Biomass Bioenergy* 13 (1997) 25–37.
- [8] Ashraf AM, Masjuki HH, Kalam MA, Rizwanul Fattah IM, Imtenan S, Shahir SA, et al. Production and comparison of fuel properties, engine performance, and emission characteristics of biodiesel from various non-edible vegetable oils: a review. *Energy Convers Manage* 2014;80:202–28.
- [9] Hoekman SK, Broch A, Robbins C, Cenicerros E, Natarajan M. Review of biodiesel composition
- [10] Bagby. Vegetable oils for diesel fuel: opportunities for development. *ASAE* 1987:87–1588.
- [11] Sivalakshmi S, Balusamy T. Effect of biodiesel and its blends with diethyl ether on the combustion, performance and emissions from a diesel engine. *Fuel* 2013;106:106–10.
- [12] Shay EG. Diesel fuel from vegetable oils: status and opportunities. *Biomass Bioenergy* 1993;4:227–42.

- [13] N. Özbay, B. B. Uzun, E. A. Varol and A. E. Pütün, "Comparative analysis of pyrolysis oils and its subfractions under different atmospheric conditions," *Fuel Processing Technology*, vol. 87, pp. 1013–1019, 2006.
- [14] Diasakou M, Louloudi A, Papayannakos N. Kinetics of the non-catalytic transesterification of soybean oil. *Fuel* 1998;77:1297–302.
- [15] Adams C, Peters JF, Rand MC, Schorer BJ, Ziemke MC. Investigation of soybean oil as a Diesel fuel extender: endurance tests. *JAOCS* 1983;60:1574–9.
- [16] Darnoko D, Cheryan M. Kinetics of palm oil transesterification in a batch reactor. *JAOCS* 2000;77:1263–7.
- [17] Isigür A, Karaosmanoğlu F, Aksoy HA. Methyl ester from safflower seed oil of Turkish origin as a biofuel for Diesel engines. *Appl Biochem Biotechnol* 1994;45/46:103–12.
- [18] Freedman B, Pryde EH. Fatty esters from vegetable oils for use as a Diesel fuel. In: *Proceedings of the International Conference on Plant and Vegetable Oils as Fuels*, 1982. p. 17–122.
- [19] Fuls J, Hugo FJC. On farm preparation of sunflower oil esters for fuel. In: *Third International Conference On Energy Use Management*, 1981. p. 1595–602.
- [20] N. Laksmono, M. Paraschiv, K. Loubar and M. Tazerout, "Biodiesel production from biomass gasification tar via thermal/catalytic cracking," *Fuel Processing Technology*, vol. 106, pp. 776-783, 2013.
- [21] M. H. Dukua, S. Gu and E. B. Hagan, "A comprehensive review of biomass resources and biofuels potential in Ghana," *Renewable and Sustainable Energy Reviews*, vol. 15, pp. 404–415, 2011.
- [22] M.M. Abou-Zaid, C. Nozzolillo, A. Tonon, M. Coppens, D.A. Lombardo, High-performance liquid chromatography characterization and identification of antioxidant polyphenols in maple syrup\*±, *Pharm. Biol.* 46 (2008) 117.
- [23] J.-Y. de Saint Laumer, E. Cicchetti, P. Merle, J. Egger, A. Chaintreau, Quantification in gas chromatography: prediction of flame ionization detector response factors from combustion enthalpies and molecular structures, *Anal.Chem.* 82 (2010)
- [24] H. Wang, G.J. Provan, K. Helliwell, HPLC determination of catechins in tealeaves and tea extracts using relative response factors, *Food Chem.* 81 (2003)307.
- [25] A.V. Bridgwater, G.V.C. Peacocke, Fast pyrolysis processes for biomass, *Renew.Sustain. Energy Rev.* 4 (2000) 1.
- [26] A.V. Bridgwater, Renewable fuels and chemicals by thermal processing of biomass, *Chem. Eng. J.* 91 (2003) 87. 6457.
- [27] M. Staš, D. Kubička, J. Chudoba, M. Pospíšil, Overview of analytical methods used for chemical characterization of pyrolysis bio-oil, *Energy Fuels* 28 (2014)385.