CHAPTER-2

2.1. Feedstocks and chemicals

Barium nitrate (99%), strontium nitrate (99%), titania (99.5%) and ceria (99.9%) were procured from Alpha Aesar, Thermo Fisher Scientific, U.S.; Acetone (99.5%), bromothymol blue (95%), benzoic acid (99.5%), calcium chloride (99.9%), cresol red (95%), deuterated chloroform (99.8% Atom D), ethylene glycol (25% v/v), neutral red (ACS grade), nile blue (85%), phenolphthalein (>98%), trapeolin (Microscopy hist.), sodium carbonate (99%), sodium sulfate (99%), and sulphuric acid (95-98%) were purchased from Sigma Aldrich (now Merck) Ltd, US; citric acid (99%) and methanol (99%) was bought from SD Fine-Chem limited, Mumbai, India; potassium hydroxide (85%) was facilitated from HiMedia LPL, Mumbai, India. All the chemicals were of analytical grade and they were used as received. *Millettia pinnata* oil, *Madhuca longifolia* oil, and *Schleichera oleosa* oil were procured from local market of Ranchi, Jharkhand, India. Waste cooking oil was fetched from the hostel canteen, mess and restaurants at the outskirts of institute campus nearby. The following feedstocks were applied in transesterification reaction for FAME production.

2.1.1. *Madhuca longifolia* oil

Mahua longifolia (Mahua) is an Indian tropical tree found largely in the central and north Indian plains and forests. It is a fast-growing tree that grows to approximately 20 meters in height, possesses evergreen or semi-evergreen foliage, and belongs to the family 'Sapotaceae'. Mahua is an oil plant whose seeds yield 35 to 47% oil (Puhan et al., 2005). In India, potential mahua oil production could be up to 60 million tonnes/year.

2.1.2. *Millettia pinnata oil*

Millettia pinnata (Karanja) is one of the species of tree in the pea family, Fabaceae, native in Indian subcontinent. Common names include Indian beech and Pongam oil tree. It is having potential oil production of 13500 million tonnes per annum but only 6% is being utilized. *Millettia pinnata* tree has the rare property of producing seeds of 25-40% lipid content of which nearly half is oleic acid (Patel and Sankhavara, 2017).

2.1.3. *Schleichera oleosa* oil

Schleichera oleosa (Kusum) belongs Sapindaceae family and members of the family are rich in oil containing toxic cyanogenic compound (HCN). It is abundantly found in India, China, Malaysia, Indonesia and Sri lanka and grows in rocky, gravelly and slightly acidic well drained soil. The annual production of *Schleichera oleosa* oil is 66000tonnes/year in India which escalates it as a potential feedstock for biodiesel production (Yadav et al., 2017).

2.1.4. Waste cooking oil

WCO is basically composed of used vegetable oil, mainly of domestic origin sunflower and soybean oil (Banerjee et al., 2019). Generally, cooking oil contains triglycerides which consist of glycerol ester of fatty acids. The physicochemical characteristics and composition profile of WCO may differ from those of fresh cooking oil because of oxidation, thermal decomposition and reduction at elevated temperature and the extent of difference in these properties often vary with frying temperature and cooking time. WCO might get enriched in diglycerides, mono glycerides and free fatty acids (FFA) as a result of hydrolysis in presence of water at high temperature. That's why, the repeated reuse of recycled oil is harmful for human health. As per survey of Food Safety and

Standards Authority of India (FSSAI), currently India is biggest consumer of vegetable oils; therefore significant amount of waste cooking oil is expected to be generated every year. Infact, on the occasion of world biofuel day on 10 August, 2019, FSSAI in coordination with Biodiesel Association of India (BDAI), has proposed an effective and well planned mechanism to collect WCO from various sources and its conversion into biodiesel. According to recent data published by FSSAI, India uses tentatively 225 lakh tons of cooking oil and around one third of it is sold to small scaled vendors at very low rate which is completely undesirable as it makes the processed food inedible. Hence, there should be a proper dumping mechanism of waste cooking oil produced at this much large scale. Fortunately, this problem can be effectively resolved by transesterification of WCO for production of FAME from it. This way, it solves the problem of dumping and provides feedstock for biodiesel simultaneously. The physicochemical properties of feedstocks were evaluated and enlisted in Table 2.1.

Table 2.1

Physicochemical properties of various feedstocks.

Properties	Unit	*Millettia pinnata* oil	*Madhuca longifolia* oil	*Schleichera oleosa* oil	Waste cooking oil
Color	Gardner color	15	10	17	10
Acid Value	mg KOH/g	8.0	13.4	12.4	1.13
Density (at 15 ºC)	g/cm^3	0.921	0.871	0.869	0.913
Kinematic viscosity (at 40 ºC)	mm^2/s	39	23.7	19	34.2
Saponification number	mg KOH/g	161	192	182	185.9
Calorific value	MJ/kg	39.01	42.13	41.59	41.3

2.2. Catalyst synthesis

To see the effect of synthesis route on biodiesel production, catalyst was synthesized by three different methods and was checked for its catalytic activity in biodiesel production.

2.2.1. Sol-gel auto combustion method

In sol-gel auto combustion method, nitrate precursors of metals are taken and dissolved in aqueous media. Later an equimolar amount of citric acid as a complexant was dissolved in distilled water and added to aforementioned metal ion solution. Nitrate-citrate salt was stabilized through dropwise addition of 25% ammonia solution till pH 7 was reached or remained in acidic medium left without addition of ammonia. The solution was stirred continuously at 60˚C which resulted in formation of nitrate-citrate salt. Afterwards, sol was poured into a container and heated slowly to 130˚C causing evaporation of excess water. Further, it was continuously heated at 200-350˚C depending upon the required condition. This resulted in further removal of volatile carboneous matters causing a gradual change in viscosity as well as color. Continued heating formed brown fluffy porous dry gel which further got automatically ignited owing to thermally induced oxidation-reduction reaction. Finally, the solid oxide powder was obtained by auto-ignition method.

2.2.2. Co-precipitation method

In coprecipitation method, metal nitrate precursors were taken in stipulated stoichiometric ratio in distilled water. Mixed metal hydroxides were precipitated by dropwise addition of equimolar sodium carbonate solution till required pH at stirring speed of 600 rpm. The precipitated material was separated from its mother liquor using

Whatman filter paper followed by extensive washing with distilled water to de-contaminate it from nitrate ions and then, dried in an oven at 80°C overnight. Onwards, mixed metal hydroxide powder was calcined in a muffle furnace at required temperature for definite time at a certain heating rate. Synthesized sample was ground with agate mortar and sieved to obtain fine powder before it was used as a catalyst.

2.2.3. Solid state route

In this technique, nitrate precursors were taken in a fixed stoichiometric ratio. Metal precursor materials were ball milled with acetone as binding agent and solvent for 7h. The crude catalyst underwent for calcination at required temperature for prefixed time interval of 4h. Finally, prepared catalyst was placed in desiccator for further use.

2.2.4. Polymer precursor methods

The polymer precursor method is slightly modified sol-gel auto combustion method to prepare mixed metal oxides. The stipulated amount of metal nitrate precursor was dissolved in the saturated aqueous solution. This aqueous solution of strontium nitrate was added to the ultrasonicated aqueous solution of metal oxide. An equimolar aqueous solution of citric acid to metal concentration was appended as a complexing agent with continuous agitation at 600 rpm on hot plate at 65°C. Afterwards, nitrate-citrate salt was made unlikely to be unstable by putting 25% ammonia solution into reaction mixture unless pH 7 was achieved. Afterwards, resulted sol was stirred vigorously at 120°C to form gel. Onward complete gelation, entire reaction mixture was put for auto combustion at 350°C. The extensive firing of reaction mixture at this elevated temperature triggered the evolution of volatile carbonaceous products and

nitrate degradation changing the color of dry gel to grey. The crude catalyst underwent for thermal treatment at certain calcination temperature in muffle furnace for the optimized time interval with heating rate of $10^{o}C/min$. Finally, the calcined catalyst was applied in transesterification after grinding with agate pestle and mortar.

2.3. Catalyst characterizations

2.3.1. Thermal analysis

Thermal analysis explores the dependency of physicochemical properties of catalyst over temperature change right from an ambient temperature to a elevated temperature. Thermogavimetric analysis (TGA) signifies the weight loss over the fluctuation in temperature as a result of oxidation and thermal degradation whereas differential scanning colorimetry reflects any kind of heat flow involved in occurring physicochemical process in crude catalyst material. Thermal analysis of catalyst was conducted on thermogravimetric differential thermal analyzer (TG/DSC/DTG) SII 6300 EXSTAR. The catalyst sample was mounted in an alumina crucible and heated from 30˚C to 1200˚C at heating rate of 10˚/min in nitrogen atmosphere.

2.3.2. Powder X-Ray diffraction (PXRD)

Powder XRD analysis is non-destructive analysis which identifies the phase as well as the structural formula of polycrystalline material alongwith lattice parameters. In diffractogram, the prominent peaks are evolved through constructive interference of incident X-Ray with diffracted rays from atoms residing in a particular plane with specific (h, k, l) index (Borchert et al., 2005). The XRD pattern was observed by Rigaku miniflex 600 bench top X-ray diffractometer (Cu K_{α} radiation with λ, 0.154nm) over the 2θ range of 20˚-80˚ with step width of 0.02. The diffraction patterns observed

in diffractogram were manually matched with their respective Joint Committee of Powder Diffraction Standard (JCPDS) files mentioned in XRD subsection.

2.3.3. Scanning Electron Microscopy (SEM)

SEM analysis is a potential technique (destructive analysis) which records high magnification image of material surface through high energized electron bombardment to analyze the surface topography or roughness. It tentatively gives the idea about particle size. Furthermore, Energy dispersive X-ray spectroscopy unit predicts the elemental composition of materials. SEM image associated with Energy dispersive X-ray spectroscopy (EDS) histogram was recorded on ZEISS EVO 18 SEM with the coating of quorum Q150R ES (Model 51-ADD0048).

2.3.4. X-ray Photoelectron Spectroscopy(XPS)

It is surface sensitive spectroscopic tool which informs about elemental composition with very low limit of detection (the parts per thousand), empirical formula, and oxidation state of elements present in compound. XPS also manifests the presence of different oxidation states of single element. XPS spectrum is recorded by illuminating the compound with high energy X-rays and successively monitoring the kinetic energy of electrons escaping from the surface of material at the height of 10nm from the surface. The elemental composition and oxidation state of individual element present in synthesized catalyst was probed by XPS analysis employing PHI 5000 Versa Probe II, FEI, Inc model of instrument.

2.3.5. Fourier-Transform Infrared (FT-IR) spectroscopy

It is an important absorption spectroscopy which tells about the presence of any exclusive functional group especially in solids and liquids. An FT-IR Spectrometer

synchronically segregates spectral data with high resolution ($4cm^{-1}$) over a wide Infra red range (400-4000 cm^{-1}). The IR spectral region of 400-1400 cm^{-1} is known as fingerprint region which represents the backbone of organic or inorganic materials. The remaining spectral region beyond 1400 cm^{-1} is called functional group which manifests the vibrations of functional groups. Here, Fourier Transform converts the raw data i.e. counts per seconds into actual spectrum. FT-IR spectra of the catalyst samples were obtained from Varian 3100 FT-IR spectrometer over 500-4000 cm^{-1} using spectral-grade KBr pellets.

2.3.6. Brunauer-Emmett-Teller (BET) surface area analysis

This analysis is based on the application of Brunauer Emmett Teller (BET) theory of multilayer adsorption of gases onto solids. Unlike Langmuir theory, this theory emphasizes on physical process of multilayer adsorption of analyte on active sites present on surface. It measures the specific surface area of material assimilating total external surface area along with pore area available for adsorption. The BET (Brunauer Emmett Teller) surface area was assessed by Micromeritics, ASAP 2020, USA. The BET surface area was enumerated from the adsorption isotherm linear plots while the pore diameter alongwith pore volume was calculated from the BJH pore size-pore volume distribution.

2.3.7. Basic strength determination

Here, catalyst acts as a heterogeneous basic catalyst to accelerate the transesterification for biodiesel production. Since, basic sites residing on the catalyst surface cause formation of acyl acceptor which finally combines with acyl moiety to produce methyl ester, higher basicity would definitely ameliorate FAME conversion

during transesterification (Arora et al., 2014). In the present study, barium and lanthanum-based catalyst acts as a Lewis base which cleaves of the O-H bond in methanol. The basic strength of catalyst was evaluated through Hammett indicator-benzoic acid titration method (Banerjee et al., 2019). Basically, catalyst solution (0.5g of catalyst in 20 mL of benzene) was added with 2 mL of an indicator solution and then left to equilibrate for 2 h until no further color changes were observed. According to the literature, the basic strength of samples is defined as being stronger than a weaker indicator that exhibits a color change, and weaker than a stronger indicator that produces no color change. The basicity values (mol/mol) of inorganic salts were determined by the method of Hammett indicator-benzoic acid (0.1 mol L^{-1} anhydrous benzoic acid solution in benzene) titration until the color changed back to the original color. In this titration, indicators with 6.8-18.4 pK_{BH}^{+} (bromothymol blue (pk_a=7.2); neutral red (pk_a = 6.8); nile blue (pk_a = 10.2); phenolphthalein (pk_a = 9.8); 2,4-dinitroaniline (pk_a=15.0); and 4-nitroaniline (pk_a=18.4))were employed to calculate the number of basic sites.

2.4. Methyl ester production

Initially, feedstock was filtered to eject the dissolved food ingredients from it using Whatman filter paper no. 42. The moisture content present in feedstock was eliminated by thermal treatment at 90°C for 2 h. In spite of this, a stipulated amount of activated carbon was put in feedstock for complete removal of moisture residue and also to decrease free fatty acid content of feedstock (Banerjee et al., 2019; Ma et al., 2015). The combination of feedstock and activated carbon was allowed to stand for 1h at 75°C with constant agitation. The free fatty acid quantity in feedstock and biodiesel was measured by following equation:

Acid Value (mg KOH/g) = V_{KOH} * 56.1 * N_{KOH} /weight of oil sample   (2.1)

where V_{KOH} represents the volume of KOH solution and N_{KOH} denotes the normality
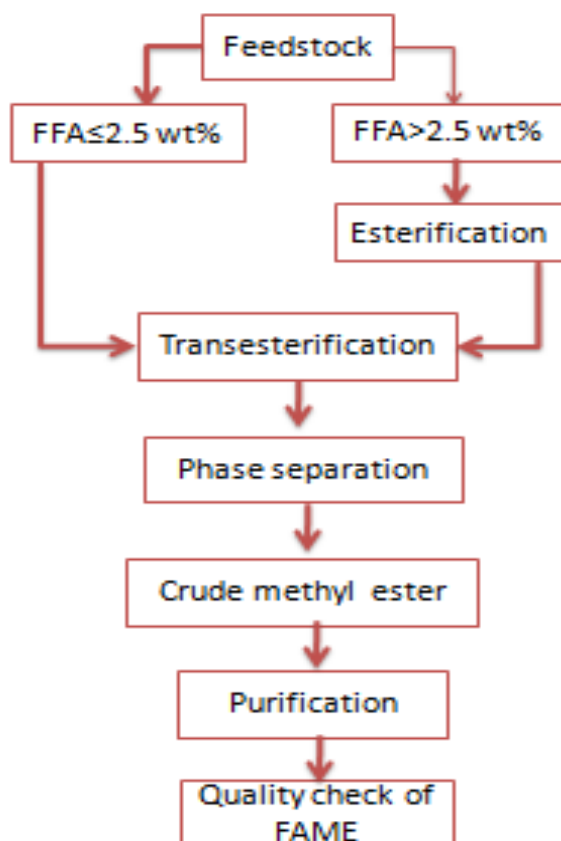
of KOH solution.

Figure 2.1 Transesterification reaction protocol.

If the feedstock was found to consist the FFA value more than 2.5wt% (3mg

KOH/g of oil), it was subjected to pretreatment called esterification, and otherwise, it

was directly fed for the transesterification reaction for FAME production. Except waste

cooking oil, all other feedstocks used in present study, had FFA value more than

permissible limit mentioned by ASTM standards, so they underwent for esterification

to lower down the FFA content.

During esterification, feedstock oil and methanol in an optimized molar ratio

were taken in three necked round bottom flask attached to water-cooled reflux condenser, mechanical stirrer and thermometer followed by addition of stipulated sulfuric acid into the mixture. Afterward, the whole content was stirred at 600 rpm for optimized time period to enhance the mass transfer rate in triphasic system on water bath at 65˚C. This esterification process led to the lowering of acid value within ASTM standards. The free fatty acid conversion was calculated by following formula.

FFA conversion (%) = (AV_1 - AV_2)/AV_1*100   (2.2)

where AV_1 and AV_2 represent the acid value of feedstock before and after esterification, respectively.

2.4.1. *Madhuca longifolia* oil

The free fatty acid content was also determined in terms of KOH required (in mg) and was found to be 14mg KOH/g using Equation (2.1). The suitable conditions optimized for pretreatment, also known as esterification are as follows: oil to methanol; 1:12, catalyst dose (sulfuric Acid); 2.0 wt %, stirrer speed; 500rpm, contact time; 75 min, reaction temperature; 60˚C. This pretreatment assuaged FFA value from 14 mg KOH/g to 0.55 mg KOH /g well below the threshold limit. Using Equation (2.2), free fatty FFA conversion was calculated to be 95%.

2.4.2. *Millettia pinnatta* oil

The FFA value turned out to be 8.05mg KOH/g, much above the permissible limit which is needed to get lessened via esterification. The esterification was processed at optimized value of process parameters using 0.6 wt% catalyst dose and 1:8 oil to methanol molar ratio at 60˚C with constant stirring of 500 rpm for 1.5h. This led to a

massive reduction in FFA content in composition. This FFA values got mitigated to 0.55 mg KOH/g well below the permissible limit laid by ASTM.

2.4.3. *Schleichera oleosa* oil

The FFA value of *Schleichera oleosa* oil was evaluated as 12.4mg KOH/g, much above the permissible limit as per ASTM standards. Therefore, esterification reaction was performed with 1.5 wt % and 1:10 oil and methanol molar ratio at 60°C for 1.5 h at 650 rpm stirring speed. Using Equation (2.2), free fatty acid conversion was calculated to be 92.2%. The treated oil feedstock had further undergone for transesterification.

2.4.4. Waste cooking oil

The FFA value of waste cooking oil were determined to be 1.13 mg KOH/g and were found well below permissible limit (Sahani and Sharma, 2018; Banerjee et al., 2019). Therefore, WCO was directly fed to transesterification reaction without any pretreatment.

Transesterification reaction was conducted in a batch reactor. The temperature programmed batch reactor was assembled with a reflux condenser and mechanical stirrer integrated with a tachometer to measure out the agitation speed. In a batch reaction, the desired amount of catalyst was actuated by keeping it with required amount of methanol for reaction in three necked round bottom flask of 250 ml capacity submerged in water bath at 60°C for 30 min. Then the measured amount of feedstock was poured into round bottom flask and transesterification reaction was started by keeping the reaction system under reflux conditions at optimized temperature and agitation speed. After the completion of reaction, the reaction mixture was transferred

to separating funnel where reaction mixture got independently settled down in three layers according to density of constituents with FAME residing in top layer. FAME layer was decanted and washed repeatedly with hot water to remove polar compounds from biodiesel. It was subsequently heated at 105°C to get rid of moisture content from biodiesel. The methyl ester conversion of this transesterification was quantified as follows (Banerjee et al., 2019).

Methyl ester conversion (%) = (2 A_{OCH3} / 3$A_{\alpha\text{-}CH2}$) * 100 (2.3)

Here, A_{OCH3} denotes the integration value of methyl protons present in FAME and $A_{\alpha\text{-}CH2}$ shows the integration value of methylene protons.

Nonetheless, used catalyst was recovered from glycerol phase and firstly washed with 5 ml of hexane three times followed by washing with methanol and ethanol. Afterwards, used catalyst was regenerated by heating it at 110°C. The only side product of this process i.e. glycerol was collected and stored for its further value addition. It is notable that all experiments which were carried out in optimization process were repeated three times to assuage the experimental error. The process was optimized for the highest methyl ester conversion.

2.5. Effect of process variables on FAME conversion

Balat and Balat (2010) reported that the most important factors influencing transesterification reaction are namely; methanol to oil molar ratio, catalyst dose, reaction temperature, reaction time, presence of water and free fatty acid content in feedstock. Furthermore, usage of heterogeneous basic catalyst limits the interphasic mass transfer in the triphasic reaction mixture, consequently, inhibits proper mixing (Dai et al., 2015). Therefore agitation speed also plays a role to facile the mass transfer

in triphasic system. So there are several important process variables concerned in the present thesis work such as catalyst dose, methanol to oil molar ratio, reaction temperature, reaction time, and agitation speed which affect the transesterification reaction tremendously. For Ba-La mixed metal oxide, the optimization process was carried out over the range of oil to methanol molar ratio (1:6 to 1:22) using 0.5-2.5 wt% of the catalyst. The reaction temperature was varied from $45^{\circ}C$ to $85^{\circ}C$ and time was varied 0.5-2.5 h with 300-700 rpm stirring speed. For Ba-Ce mixed metal oxide, the optimization process was accomplished at following reaction circumstances over a precise range: oil to methanol molar ratio (1:3 to 1:23), catalyst dose (0.3-1.8 wt%), reaction temperature (35-85°C), reaction time (20-120 min), and agitation speed (400-800 rpm). For Sr-La mixed metal oxide, the optimization was carried out at 1:5 to 1:20 molar ratio (oil: methanol) using 0.5–2.5 wt% of catalyst. Reaction temperature was varied from 40°C to 90°C and time was varied 20–120 min with 300–800 rpm stirring speed. For Sr-Ti Mixed metal oxide, the process parameters namely catalyst dose (0.5-1.5 wt%), methanol to oil molar ratio (5-15), and reaction time(30-120min) were optimized by RSM using CCD at 65°C and 600 rpm.

2.6. Kinetics and Thermodynamics of transesterification

The kinetics of transesterification reaction was investigated by performing the kinetic experiments over the saturation time of reaction. Fate of reaction was also interpreted by analyzing the temperature dependence of reaction. According to above mentioned mechanism, the entire reaction lasted in three major steps i.e. adsorption of reactant, surface chemical reaction followed by desorption of product molecules from the catalyst surface (Putra et al., 2018). Though surface chemical reactions were performed on the interface of a heterogeneous catalyst, mass transfer factor could come

into picture but the proper agitation speed of reaction mixture at most favorable temperature condition reduced the mass transfer resistance (Banerjee et al., 2019). Moreover, the first and last surface phenomena did not affect the fate of reaction as these were known to be faster processes due to being physical processes. In this situation, chemical reaction (Figure 1.7) was rate determining step. Hence the rate law expression for the reaction could be given in Equation (2.4).

$$\text{Rate of transestrification reaction(r)} = -\frac{dC_{TG}}{dt} = k'C_{TG}C_M^3 \qquad (2.4)$$

Here, k' is the rate constant, C_{TG} is concentration of triglyceride and C_M represents the concentration of methanol fed in transesterification.

Though the concentration of methanol used in actual reaction differed from the ideal stoichiometric ratio of 3:1 methanol to oil molar ratio, instead excess of methanol was provided to reaction (Putra et al., 2018). Thus, the rate of change in the concentration of methanol did not affect the course of reaction. Therefore, Equation (2.4) was modified to the following equation.

$$r = -\frac{dC_{TG}}{dt} = k'C_{TG}C_M^0 \qquad (2.5)$$

Consequently, a pseudo first order kinetic model was adopted and analyzed for its validation through kinetic experiments. Thus, Equation (2.5) further got converted to following one.

$$r = -\frac{dC_{TG}}{dt} = kC_{TG} \qquad (2.6)$$

After integration on both side of Equation (2.6),

$$-\ln\frac{C_{TG}}{C_{TG}^0} = k.t \qquad (2.7)$$

Where C_{TG} represents the concentration of triglyceride at time t while C_{TG}^0 depicts concentration of triglyceride at t=0 and k (k $= k' C_M^0$) denotes the pseudo-first order rate constant (min^{-1}).

To the fact that during reaction C_{TG} keeps on transforming into FAME, that's why, C_{ME} is convertible into $\frac{C_{TG}}{C_{TG}^0}$ as the following.

$$C_{ME} = \frac{C_{TG}^0 - C_{TG}}{C_{TG}^0} = 1 - \frac{C_{TG}}{C_{TG}^0} \qquad (2.8)$$

$$(1 - C_{ME}) = \frac{C_{TG}}{C_{TG}^0} \qquad (2.9)$$

After substituting fraction of triglyceride concentrations in Equation (2.7),

$$-\ln(1 - C_{ME}) = -kt \qquad (2.10)$$

Where C_{ME} is concentration of FAME.

The reaction rate constant (k) could be enumerated from Equation (2.10). Besides, the extent of temperature dependence of k was deduced through the application of Arrhenius equation written as the following Equation (2.11).

$$\ln k = \ln A - E_a/RT \qquad (2.11)$$

Where, k is the rate constant of the reaction; A is the frequency factor or pre-exponential constant (min^{-1}); E_a is the activation energy (kJ/mol) of process; R is the gas constant (8.314 J mol^{-1} K^{-1}), and T is the absolute temperature (K) of the reaction. Freedman et al (1986) reported that the activation energy of reaction following pseudo-first-order kinetics usually falls in the range of 33.6-84.0 kJ mol^{-1} in case of base-catalyzed transesterification reaction. Thermodynamic functions such as enthalpy change ($\Delta H°$), entropy change ($\Delta S°$), and Gibb's free energy change ($\Delta G°$) were

derived from Eyring-Polanyi equation written below (Banerjee et al., 2019).

$$k = \frac{k_b T}{h} \exp\left(-\frac{\Delta G^\circ}{RT}\right) \tag{2.12}$$

$$\text{Since, } \Delta G^\circ = \Delta H^\circ - T\Delta S^\circ \tag{2.13}$$

Equation (2.12) was modified to the following equation after taking natural logarithm on both side and substituting the value of ΔG° from Equation (2.13).

$$\ln\left(\frac{k}{T}\right) = -\left(\frac{\Delta H^\circ}{RT}\right) + \left[\ln(\frac{k_b}{h}) + \frac{\Delta S^\circ}{R}\right] \tag{2.14}$$

where, k is the rate constant (min^{-1}); T is the absolute temperature (K), R is the universal gas constant (8.314 J mol^{-1} K^{-1}), h is known as Planck's constant (6.626×10^{-34} J s^{-1}) and k_b is termed as Boltzmann's constant (1.38×10^{-23} J/K).

2.7. Green chemistry metrics and Turn Over Frequency(TOF)

Green chemistry metrics measure the green aspect of a chemical process and correlate to the principles of green chemistry. To understand the impact of this methanolysis process on environment, Environmental Factor 'i.e. E-factor was estimated and discussed in present study. The greenness approach of this transesterification reaction was estimated by E- factor using following formula.

$$\text{Envionement Factor}\,(E-\text{factor}) = \frac{\text{weight of waste generated}}{\text{weight of product obtained}} \tag{2.15}$$

Additionally the potential of prepared catalyst was evaluated through the Turn Over Frequency (TOF) which quantified the catalytic activity of catalyst undergoing methanolysis followed by surface chemical reactions forming methyl ester. TOF is defined as number of moles of substrate undergone the reaction at unit concentration of active sites of catalyst over unit time in heterogeneous catalysis (Sahani et al., 2019).

This value is determined using the given formula below.

$$\text{Turn over frequnecy(TOF)} = \frac{\text{Number of moles of substrates reacted}}{\text{Number of moles of basic sites of catalyst} \times \text{time}} \qquad (2.16)$$

2.8. Methyl ester characterization

The synthesized methyl ester was inspected through Gas chromatography-Mass Spectroscopy (GC-MS), Proton Nuclear Magnetic Resonance (^{1}H-NMR) Spectroscopy, Carbon Nuclear Magnetic Resonance (^{13}C-NMR) Spectroscopy and Attenuated Total Reflectance Fourier Transform Infrared (ATR FT-IR) Spectroscopy.

2.8.1. Gas chromatography - Mass Spectroscopy (GC-MS)

Gas chromatography - Mass Spectroscopy is an analytical method that performs the task of both Gas chromatography and Mass Spectroscopy collectively. It identifies the presence of different substances present in analyte mixture of compounds (Likozar and Levec, 2014). Here, GC-MS analysis was carried out to determine chemical composition of feedstocks. Gas chromatograph separates the constituents according to retention time and constituents are further fed to online Mass spectrometer which analyzes the m/z ration of molecules and its further fragmented parts. Mass spectrum was interpreted by means of NIST database. GC-MS analysis of FAME was discharged on Perkin Elmer Clarus 680 Gas Chromatograph and SQ8T Mass spectrometer at inlet temperature of 200˚C, auxiliary temperature of 250˚C; initially raised to 50˚C for 10 min at the rate of 10˚C per min then 50˚C to 300˚C with same rate.

2.8.2. Proton and Carbon Nuclear Magnetic Resonance Spectroscopy (^{1}H-NMR) and (^{13}C-NMR)

Proton NMR spectroscopy informs about the different types of protons present

in organic material whereas carbon NMR spectroscopy signifies environment of carbon present in analyte compound (Satyarthi et al., 2009). Ultimately, NMR spectroscopy deciphers the structure of organic compounds. Moreover, Fatty acid methyl ester (FAME) in biodiesel was quantified from ^{1}H-NMR spectroscopy. In current study, ^{1}H-NMR analysis in combination with ^{13}C-NMR was attained using JEOL AL 300 instrument where Tetramethylsilane (TMS) was utilized as an internal standard and deuterated chloroform as a solvent at the scan rate of 32 with 512 scan.

2.8.3. Attenuated Total Reflectance Infrared Spectroscopy (ATR FT-IR)

Attenuated Total Reflectance (ATR) in combination with Infrared Spectroscopy allows the sample analysis without any sample (solids or liquids) preparation. It gives out the idea about the stretching and bending vibrations of the bonds present in methyl esters. ATR FT-IR spectrum of methyl ester was acquired on Alpha Bruker Eco-ATR attached with ZnSe ATR crystal over 500-4000 cm^{-1} with 24 scans at resolution of 4 cm^{-1}.

2.8.4. Physicochemical properties of methyl esters

The physicochemical and fuels characteristics of methyl esters were evaluated as per the norms laid down by ASTM standards. The compatibility of prepared methyl esters was decided by several important physicochemical and fuels properties such as acid value, calorific value, cetane number, density (at 15˚C), flash point, cloud point, fire point, and kinematic viscosity (at 40˚C) (Table 2.2) (Liu et al., 2009).

2.8.4.1. Acid value

The Acid value represents the number of milligrams of potassium hydroxide (KOH) required to neutralize the free fatty acids in 1 g of sample. According to ASTM

D6751 standard methods, an ideal substitute of conventional biofuel must have acid value less than 0.50 mg KOH/g.

2.8.4.2. Calorific value

The calorific value of a fuel is defined as the amount of heat energy liberated on the complete combustion of a unit amount of fuel. It is generally measured in units of energy per amount of material, e.g. kJ/kg. ASTM D4809 test method has been followed for the determination of calorific value. Generally, FAME acquired comparatively lower calorific value than that of diesel owing to high oxygen proportion.

2.8.4.3. Density

Since FAME has higher density than conventional diesel fuel owing to higher molecular formula and its mass, its vital application has been encouraged in form of blending with diesel fuel. However, density of FAME was analyzed according to ASTM D1298 standard test method.

2.8.4.4. Kinematic viscosity

Kinematic viscosity is obtained by dividing the absolute (or dynamic) viscosity to fluid density. It is incorporated by internal friction of fluid which causes disturbance in fuel injection into engine (Liu et al., 2009). It is determined by Redwood viscometer by following the ASTM D445 standard test method. Fluid with higher kinematic viscosity faces the problem of poor atomization inside the engine, carbon choking and hence incomplete combustion reducing the engine efficiency. Kinematic viscosity is directly correlated to density which can be mitigated by transesterification reaction remarkably.

2.8.4.5. Cetane number

The cetane number measures the delay in the ignition time of the fuel. The cetane number refers to the ease with which diesel fuel ignites easily at a relatively low temperature. Fuels with lower cetane number have longer ignition delays, requiring more time for the fuel combustion process to be completed. Hence, higher speed diesel engines operate more effectively with higher cetane number fuels (Liu et al., 2009). Running a CI engine on fuel with a lower-than-recommended cetane number can result in rough operation (noise and vibration), low power output, excessive deposits and wear, and hard starting. Here, Cetane number was determined by Ignition quality tester referring ASTM D 613 standard test methods.

2.8.4.6. Flash point and fire point

The flash point of a volatile material is the lowest temperature at which its vapors get ignited if given an ignition source. The flash point is sometimes confused with the auto ignition temperature, the temperature that causes spontaneous ignition where as fire point of a fuel is the lowest temperature at which the vapor of that fuel will continue to burn for at least 6 seconds after ignition by an open flame of standard dimension (Candeia et al., 2009). Usually, fire point is 20-30°C above flash point. Here, flash point and fire point have been estimated by Pensky-Martens Closed Tester adopting ASTM D93 standard test method.

2.8.4.7. Cloud point

Cloud point refers to the temperature below which wax in diesel or biowax in biodiesels forms a cloudy appearance. The presence of solidified waxes thickens the oil and clogs fuel filters and injectors in engines. The wax also accumulates on cold

surfaces and forms an emulsion with water. Here, cloud point has been estimated by Pensky-Martens Closed Tester adopting ASTM D 97 standard test method.

Table 2.2

Physicochemical properties of methyl ester derived from non-edible feedstocks.

Properties	Unit	ASTM standards range for biodiesel	Testing Apparatus	Uncertainty (%)	ASTM standards
Acid value	mg KOH/g	≤0.5	Titrimetric method	±1.5% (R.U.)[†]	D664
Color	-	Yellowish	-	-	Yellowish
Calorific value	(MJ/kg)	-	Bomb calorimeter	-	D4809
Copper strip corrosion	-	3a max	Copper strip corrosion apparatus	-	D130
Cetane number	-	48−65	Ignition quality tester	±1.38 (R.U.)	D 613
Density (at15°C)	g/cm^3	0.860 − 0.900	pycnometer	±0.0001 (A.U.)[‡]	D1298
Flash point	°C	100−170	Pensky-Martens Closed Tester	-	D93
Cloud point	°C	-3−12	-	-	D97
Fire point	°C	130−210	Pensky-Martens Closed Tester	-	D93
Kinematic viscosity (at 40°C)	mm^2/s	1.9 − 6.0	Red wood viscometer	±0.02(A.U.)	D445

[†] **Relative uncertainty**

[‡] **Absolute uncertainty**