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Biodiesel Preparation, Process Optimization and Characterization from Neem seed Oil

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Abstract: ***The consumption of edible oil is very high in the country and still the indigenous production does not meet the demand and considerable amount of edible oil is imported. Also, it is not advisable to divert these sources for biodiesel production. On the other hand, the non-edible oil resources could be a solution for biodiesel production. Non-edible oil from the plant seeds is the most promising alternative fuel for internal combustion engine because it is renewable, environment friendly, non-toxic, biodegradable has no sulphur and aromatics, has favourable combustion value and higher cetane number. Extensive work has been done on the transesterification of non-edible oils; however, no significant work has been done on the optimization of transesterification process, oil characterization and fuel analysis of most of the non-edible seed oils. Low cost and abundantly found non-edible oils such as Neem oil could be a better option for biodiesel processing. In the present work, optimization of transesterification process and analysis of biodiesel from non-edible oil was done; based on optimized protocol for biodiesel production from Neem seed oil converted into fatty acid methyl esters (FAME) through base catalyzed trans esterification using an optimum ratio of 1:6 (Oil : Methanol) at 60^{0}C. Biodiesel from these sources was analyzed for qualitative and quantitative characterization by using, GC-MS and FT-IR techniques. Based on qualitative and quantitative analysis of biodiesel, it is concluded that the biodiesel from these species can be feasible, cost effective and environment friendly.***
Keywords: Neem oil, **Biodiesel,** ***Tran's esterification, GC-MS, and FT-IR.***

I. INTRODUCTION

India occupies second place in population and 7th place into area in the World. Due to large population and the need of transportation would made India top 5th country in the World in consumption of petroleum products. The yearly consumption of diesel in India is approximately 40 million tones, which constitutes about 40% of the total petro-products consumption (1). As these carbon sources are limited and the consumption of petro-products is increasing day by day, there is a need of alternative resources, which includes solar energy, thermal energy, hydro energy and bio-energy. One of the most prominent alternative energy resources, attracting more and more interest in recent years with the price for crude oil reaching record heights, is biodiesel, which is a possible substitute for petroleum-based diesel fuel. Production of biodiesel using plant sources is good alternative resource through which we can meet the demand for petroleum products. Biodiesel is an alternative to diesel which is made from renewable resources such as vegetable oils (or) animal fats (2). The oilseed production in the country presently meets only 60-70% of its total edible oil requirements and the rest is met through imports (3). India also has a potential of collecting 5 million tones of tree-borne oilseeds (TBO) of which only one million tons are being collected presently (4).

In Tran's esterification reaction base chemical catalyst processes are more practical compared with the enzymatic method. Alkali process can achieve high purity and yield of biodiesel product in a short time (5, 6, and 7). Methyl esters are the product of Trans esterification of vegetable oils with alcohol (methanol) using an alkaline catalyst. In addition, the process yields glycerol which has great applications in the pharmaceutical, food and plastics industries (8, 9, and 10).

Biodiesel offers a number of interesting and attractive beneficial properties compared to conventional petroleum-based diesel. Most important, the use of biodiesel maintains a balanced carbon dioxide cycle since it is based on renewable biological materials (11). Additional environmental benefits are reduced emissions (carbon monoxide, sulphur, aromatic hydrocarbons, and soot particles) during combustion. Biodiesel is non-toxic and completely biodegradable (12). Due to its high flash point, it is of low flammability and thus its use is very safe and non-hazardous. Furthermore, it provides good lubrication properties, thereby reducing wear and tear on engines (13). Purpose of the present study is to optimize the processing parameters for improved production of biodiesel by using Tran's esterification process (14). Neem oil was the raw material with methanol and sodium hydroxide as the catalyst and to evaluate the produced biodiesel as a fuel (15). While production of biodiesel, the optimization parameters of the process were determined.

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The main objective of this research is production, process optimization and characterization of biodiesel from the neem seed oil which was collected from local sources, i.e., Anthapurumu of Andhra Pradesh. The aim of present work was to produce biodiesel in the form of fatty acids methyl esters (FAME) from neem seeds oil (NSO) by Tran's esterification through NaOH catalysis. It was also to experiment the biodiesel production process by evaluating different parameters for optimization of Tran's esterification process.

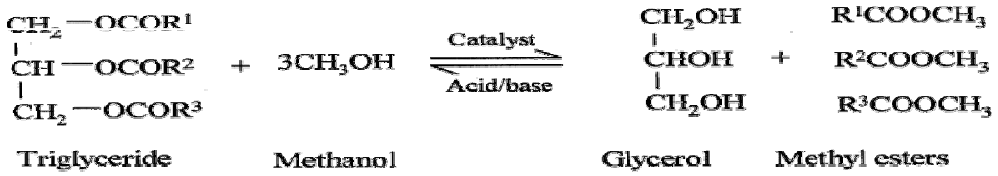
II. EXPERIMENTAL INVESTIGATIONS

A. Experimental Work

The Neem seed oil was used as raw material for biodiesel production and which was collected from local sources. Seeds of these plants were expelled by using electric oil expeller (KEK P0015-10127), India. Methanol 99.9% purity, sodium hydroxide (NaOH) and anhydrous sodium sulphate (Na_2SO_4) were of analytical grade obtained from Merck, Bangalore, India.

B. Transesterification

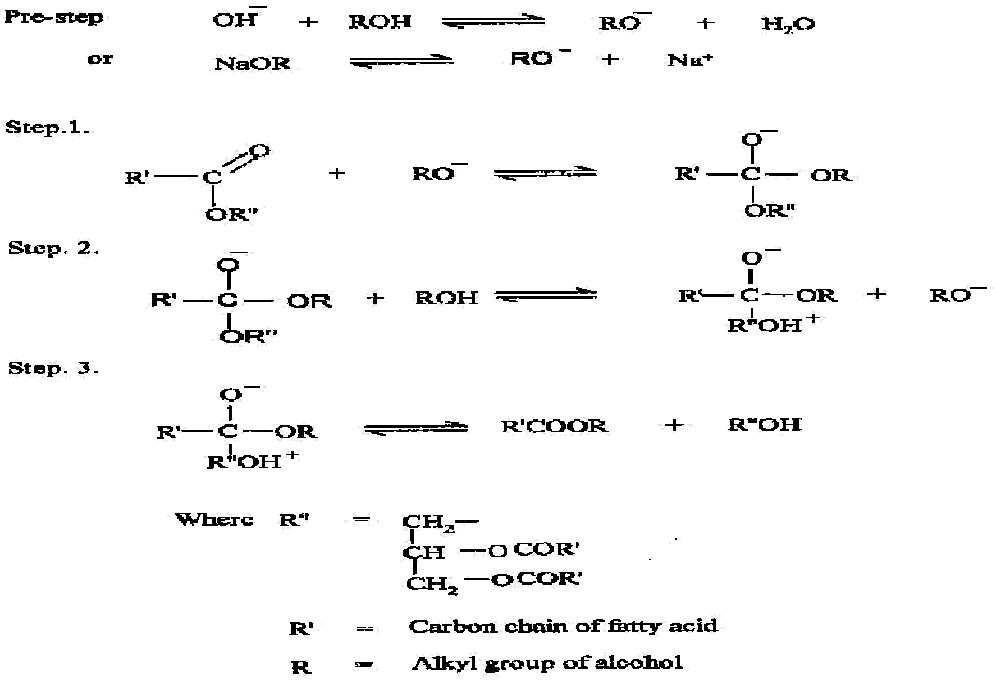
Transesterification is the displacement of alcohol from an ester by another. The process is similar to hydrolysis; in this process alcohol is used instead of water. This process has been widely used to reduce the high viscosity of triglycerides. The transesterification reaction is represented by



Equation 1. General equation for methanolysis of triglycerides

Transesterification is one of the reversible reactions and proceeds essentially by mixing the reactants (15). However, the presence of a catalyst (NaOH) accelerates the conversion. The mechanism of alkali-catalyzed trans-esterification is described in Scheme 1. First step involves the attack of alkoxide ion to the carbonyl carbon of triglyceride molecule, which results in the formation of a tetrahedral intermediate. The reaction of this intermediate with an alcohol produces alkoxide ion in the second step. In the last step, rearrangement of the tetrahedral intermediate gives rise to an ester and a diglyceride. The same mechanism is applicable to di-glyceride and mono-glyceride (16).

C. Mechanism of Trans Esterification Process



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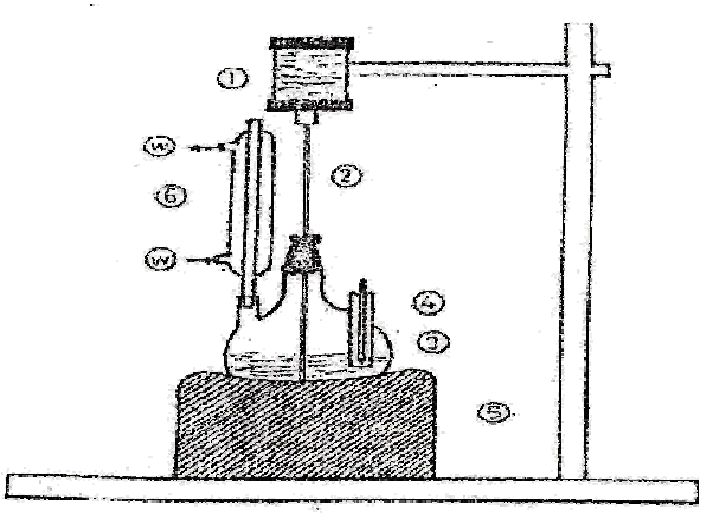


Fig. 1 Experimental setup for preparation of methyl esters from Neem and pongamia oil

D. Experimental Set up & Procedure

A 2000 ml three-necked round-bottomed flask was used as a reactor. The flask was placed in a water bath, whose temperature could be controlled within ± 2^0C. One of the two side necks was equipped with a condenser and the other was used as a thermos well. A thermometer was placed in the thermo well containing little mercury for temperature measurement inside the reactor. A blade stirrer was passed through the central neck, which was connected to a motor along with speed regulator for adjusting and controlling the stirrer speed.

1.	Electric Motor	4.	Thermo-well with Thermometer
2.	Stirrer	5.	Water bath
3.	Three-necked Round Bottom Flask	6.	Condenser

1) *Esterification (17, 18):* A known amount of Neem oil was taken in the above-mentioned setup. Required amount of sulphuric acid and methanol were added to the oil and stirred continuously maintaining a steady temperature of 64^0C. Intermittently samples were collected at regular intervals (30 min.) and acid value was determined. After the confirmation of complete reduction of acid value to less than 1.0, the heating was stopped and the products were cooled. The unreacted methanol was separated by separating funnel. The remaining product was analyzed for acid value and it was found that the acid value varied from 1.0 to 0.5. This oil sample was used for transesterification to obtain methyl esters.
2) *Experimental Procedure:* After thorough cleaning of the same setup, known amount of esterified Neem was charged. Required amount of catalyst NaOH was dissolved in methanol and the rest amount of methanol along with the catalyst solution was added to the oil sample. After proper closing of the flask it was put on the water bath. The system was maintained airtight to prevent the loss of alcohol. The reaction mixture was maintained at temperature 60-70^0C to speed up the reaction rate. Excess alcohol was used to ensure total conversion of the oil in to its methyl esters. The formation of methyl ester was monitored by measuring the viscosity of the reaction mixer. Reduction in viscosity confirms the formation of methyl esters. This procedure was followed for all the samples collected at regular interval of time to check the formation of methyl esters. After the confirmation of complete formation of methyl esters, the heating was stopped and the products were cooled and transferred to a separating funnel. Where the ester layer containing mainly methyl ester and methanol and glycerol layer containing mainly glycerol and water were separated.

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For neutralization a known amount of sulfuric acid in methanol was added to both the layers separately to neutralize the sodium methoxide present in them. The traces of methanol present in ester layer were recovered in a distillation column under controlled vacuum. Distilled methanol was weighed and stored in sample bottle. Similar procedure was adopted to recover the traces of methanol present in glycerol layer. The methyl ester was washed and dried under vacuum to remove traces of moisture. A sample of esters was analyzed for acid value by using standard AOCS procedure. The sample of glycerol layer was analyzed for glycerol content by using AOCS procedure.



Fig. 2 Mixing the catalyst sodium hydroxide to methanol to liberate sodium methoxide



Fig. 3 Filtration process of esterification

E. Oil Properties (19 & 20)

The experimental investigations were carried out for different fuel properties and which were evaluated according to ASTM D-445, D-1298, D-93, D-1298, D-1500, and D-4294.

F. FT-IR Analysis (21)

The presences of fatty acids, organic and inorganic materials were identified through the Fourier Transform Infrared (FT-IR) Spectroscopy analyzer. Biodiesel samples were characterized by FT- IR, using a Bio-Rad Excalibur Model FTS3000MX. The resolution was $1cm^{-1}$ and 15 scans. The FT-IR spectra were recorded between 400 and 4000 cm^{-1}.

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G. GC-MS Method (22)

The fatty acid methyl ester (FAMEs) contents were determined by gas chromatography, model GC 6890N coupled with mass spectrometer, model MS 5973 MSD (mass selective detector). Separation was performed on a capillary column DB-5MS (30 m×0.32 mm, 0.25µm of film thickness). The carrier gas was helium with flow rate of 1.5 mL/min. The column temperature was programmed from 120-300^{0}C at the rate of 10^{0}C/min. A sample volume of 0.1µL NOB in chloroform was injected using a split mode, with the split ratio of 1:10. The mass spectrometer was set to scan in the range of m/z 50-550 with electron impact (EI) mode of ionization.

III. REMEDIAL MEASURES

The following parameters were studied in order to optimize the transesterification process.

A. Reaction Temperature (23)

The rate of reaction is influenced by the reaction temperature as per kinetics of reaction. Generally the reaction was conducted close to the boiling point of the methanol at the atmospheric pressure. The maximum yield of esters was observed at temperature ranging from 60 to 70^{0}C. Further increase in temperature has negative effect on the conversion. Studies have also indicated that sufficient time for reaction and temperature are important parameters for the better results.

B. Ratio of alcohol to oil (24)

Higher molar ratio of alcohol to vegetable oil interferes in the separation of glycerol. On the other hand with lower molar ratio, required more reaction time and conversion increases but recovery decreases. It was also found that optimum molar ratio depend upon type and quality of oils. The mixing effect is most significant during the slow rate region of the trans-esterification reaction. It is the most valuable tool for the transesterification process, while study the kinetics of reaction. In the reaction the different molar ratios of methanol and oil are being used.

C. Catalyst type & effect of concentration of catalyst (25)

Alkali catalysts are the most effective for Trans esterification compared to the other. The NaOH is more frequently used as catalyst in the process than acids and enzymes. The different amounts of catalyst give different yields of biodiesel, glycerin and soap. The different concentration of catalyst leads to formation of different yields of biodiesel. If the concentration of catalyst is increases biodiesel yield is decreases and formation of soap increases.

4. Time Duration (26)

Few investigations found that reaction starts very fast and almost 80% conversion takes place in the first five minutes. After that, the reaction slows down and after one hour 93-98% conversion of triglycerides into ester takes place (Ma and Hanna, 1999; Srivastava and Prasad, 2000). In the present work, effect of reaction time from 60-120 minutes on reaction yield was investigated it was found that the reaction time was increases more than two hours the ester yield is decreases slightly.

D. Effect of stirring speed (in rpm) (27)

The effect of stirring speed of the mixture was varied from 400 rpm to 700 rpm. Based on the experimental observations it can be seen the 600 rpm stirring speed gives maximum product output, therefore this is chosen as the optimum stirring speed for the remaining experiments.

IV. RESULTS AND DISCUSSION

A. Determination of Free Fatty acid (FFA)

The FFA has significant effect on the transesterification of oils with alcohol using catalyst. The high FFA content (>1%w/w) will cause soap formation and the separation of products will be exceedingly difficult, and as a result, low yield of biodiesel product would be obtained. It is important to first determine the FFA content of oil. The free fatty acid (FFAs) number of crude Neem oil 2.5% and biodiesel yield.

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1) *Experimental Results by Varying Molar Ratio of Methanol and Oil*

a) *Discussion:* The yield of methyl esters of Neem oil is investigated by changing molar ratios of methanol/oil (4:1, 5:1, 6:1 and 7:1).The stirring speed, reaction time, catalyst concentration and temperature were kept constant. The highest conversion rate of neem biodiesel (71%). The soap formation in the case of Neem is 0.6%. From the Figure 4 and Table 1, it is known that 6:1 molar ratio was optimized for biodiesel preparation.

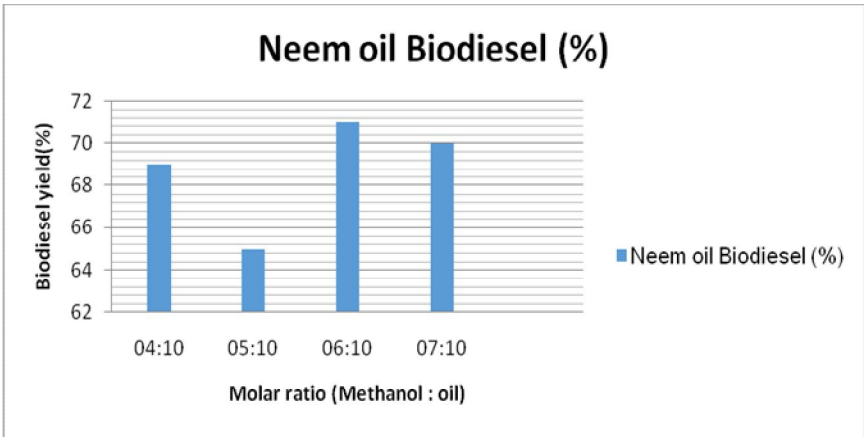


Fig. 4 Biodiesel yield with different ratios of methanol & oil

Table 1 Biodiesel Yield with Different Molar Ratios of Methanol and Oil

Oil type	Methanol:oil ratio	Biodiesel (%)	Glycerin (%)	Soap (%)
Neem oil sample	4:1	69	5.10	0.90
	5:1	65	5.80	1.25
	6:1	**71**	**7.10**	**0.6**
	7:1	70	6.15	1.0

2) *Experimental Results by Varying Temperature*

a) *Discussion:* The yield of methyl esters of Neem oil is investigated by changing temperatures (40^0C, 50^0C, 60^0C and70^0C). The results of yield of biodiesel with varying temperatures were given in Table 2 and Figure 5. The Stirring speed, reaction time, catalyst concentration and molar ratio of methanol/oil were kept constant. The highest conversion rate of neem biodiesel (71%) obtained at 60^0C. The soap formation at highest yield of Neem is 0.9%. The glycerine formation at highest yield of Neem oil is 9.20%.

Table 2 The yield of Biodiesel with Different Temperatures

Oil type	Temperature (^{0}C)	Biodiesel (%)	Glycerin (%)	Soap (%)
Neem oil sample	40	48	8	1.05
	50	50	8.10	1.0
	60	**71**	**9.20**	**0.9**
	70	59	6.50	1.10

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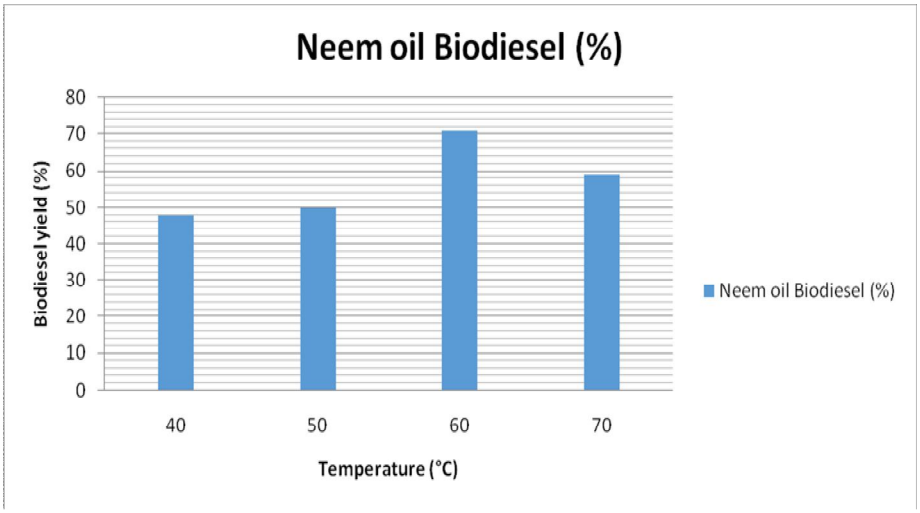


Fig. 5 Biodiesel yield of neem with different temperatures

3) Experimental Results By Varying Time

a) Discussion: The yield of methyl esters of Neem oil is investigated by changing time (1hr, 2hrs, 3hrs, and 4hrs). Table 3 and Figure 6 represent the results of biodiesel yield with different time intervals. The Stirring speed, Temperature, catalyst concentration and molar ratio of methanol/oil were kept constant. The highest conversion rate of neem biodiesel (71%) obtained at the time 2hrs. The soap formation at highest yield of Neem oil is 0.6%. The glycerin formation at highest yield of Neem oil is 7.10%.

Table 3 Biodiesel yield with different time intervals (HRS)

Oil type	Time(hrs)	Biodiesel (%)	Glycerin (%)	Soap (%)
Neem oil sample	1	62	9.0	1.5
	2	**71**	**7.10**	**0.6**
	3	69	6.0	1.05
	4	68	7.0	1.20

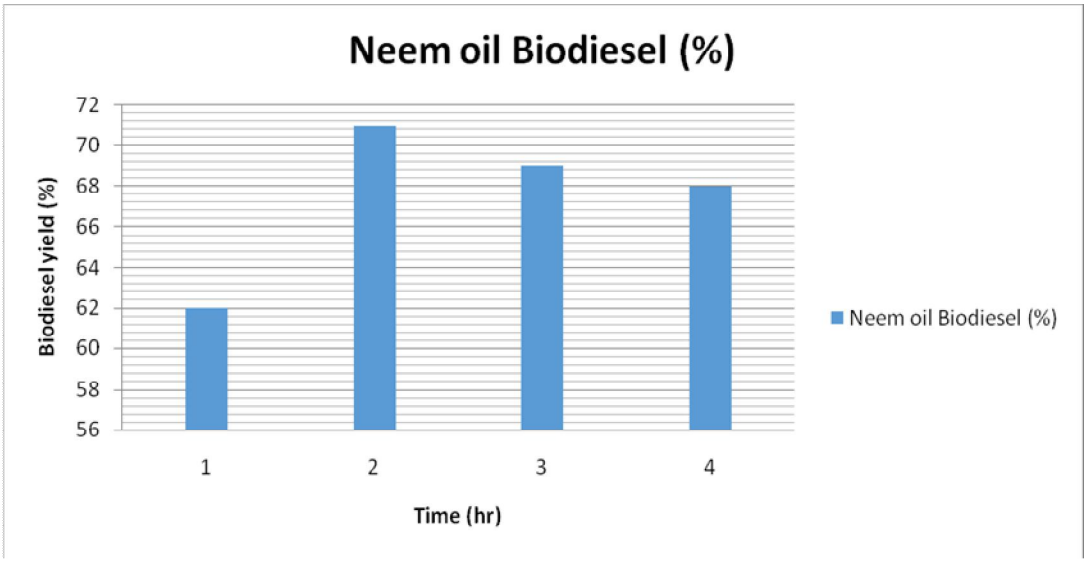


Fig. 6 Neem biodiesel yields with different time intervals

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4) Experimental Results by Varying Stirring Speed (In RPM)

a) Discussion: The yield of methyl esters of Neem oil is investigated by changing stirring speed (400 rpm, 500 rpm, 600 rpm and 700 rpm). The results of biodiesel yield with different stirring speeds were shown in Table 4 and Figure 7. The time, temperature, catalyst concentration and molar ratio of methanol/oil were kept constant. The highest conversion rate of neem biodiesel (71%) obtained at stirring speed 600rpm. The soap formation at highest yield of Neem oil is 1.05%. The glycerine formation at highest yield of Neem oil is 7.50%.

Table 4 Biodiesel yield with different stirring speed (in rpm)

Oil type	Velocity (rpm)	Biodiesel (%)	Glycerin (%)	Soap (%)
Neem oil sample	400	63	8.15	1.15
	500	67	6.10	1.25
	600	**71**	**7.50**	**1.05**
	700	69	6.0	1.1

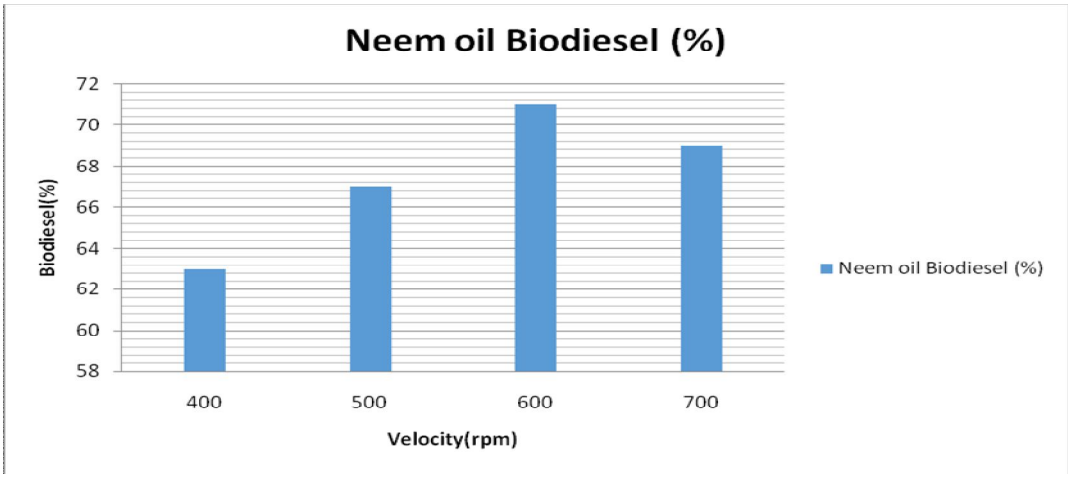


Fig. 7 Neem oil biodiesel yields with different velocities (rpm)

5) Experimental results by varying Catalyst Concentrations: The yield of methyl esters of Neem oil is investigated by changing catalyst concentrations (0.5gms, 0.6gms, 0.7gms, and 0.8 gms). The time, temperature, stirring speed and molar ratio of methanol/oil were kept constant. Table 5 and Figure 8 was depicts the results of biodiesel yield with different catalyst concentrations. The highest conversion rate of neem biodiesel (71%) obtained at 0.7gms. The soap formation at highest yield of Neem oil is 1.05% and 1.1% respectively. The glycerine formation at highest yield of Neem oil is 7.50%.

Table 5 Biodiesel yield with different concentrations of catalyst

Oil type	Catalyst NaOH concentration(in gm)	Biodiesel (%)	Glycerin (%)	Soap (%)
Neem oil sample	0.5	63.25	8.15	1.15
	0.6	67.40	6.10	1.25
	0.7	**71.0**	**7.50**	**1.05**
	0.8	63.50	12.10	2.90

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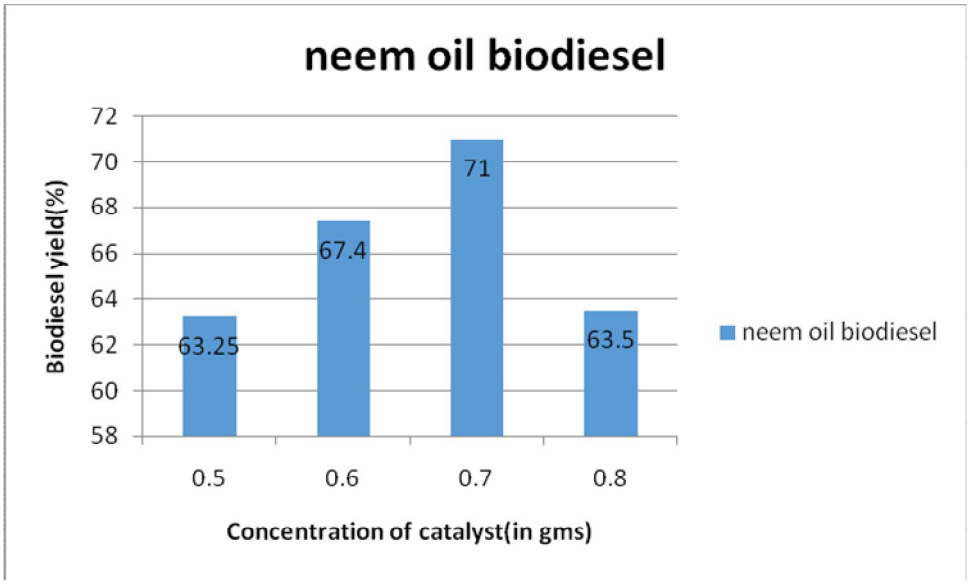


Fig. 8 Neem oil biodiesel yield with various catalyst concentrations

B. Discussion of final result

To achieve maximum yield of biodiesel from non-edible seed oils, alkali based transesterification was carried out. The yields of methyl esters of Neem oilis investigated by changing parameters like molar ratio (6:1), temperature (60^0C), stirring speed (600 rpm) and reaction time (2 hrs.). The highest conversion rate was obtained with the catalyst concentration of 0.7 g, under these conditions and the biodiesel yield was 71% for Neem oil. When the catalyst concentration increased to 0.8 g, it was observed that the ester formation decreased with the increase in sodium hydroxide concentration and soap formation was increased. This is because the higher amount of catalyst may cause soap formation. Soap formation reduces catalyst efficiency, causes an increase in viscosity, leads to gel formation and makes the separation of glycerol difficult. Table 6 gives the comparative results of Neem oil biodiesel with HS diesel.

Table 6 Comparative ANALYSES for fuel properties of Neem oil biodiesel with High speed diesel

Properties	Neem oil Biodiesel	Diesel
Kinetic viscosity @ 40^0C cst	4.81	1.3-4.1
Density @ 30^0C Kg/L	1.74	1.76
Flash point	124	60-80
Sulphur % wt	Nil	0.05
Color comparison	2.5	2.0
Pour Point (^{0}C)	9	-35 to 15
Cloud point (^{0}C)	12	-15 to 6

C. Viscosity

High viscosity is the major problem preventing the use of vegetable oils and animal fats directly in diesel engines as it affects the flow of fuel and spray characteristics. High Speed Diesel (HSD) has viscosity of 1.3-4.1 at 40^0C whereas the Neem oil biodiesel is 4.81 which were closer to HSD.

D. Density

The density of Neem oil biodiesels at 30^0Cwas found to be 1.74 Kg/L and 1.72 Kg/L which are closer to the density of diesel i.e., 1.76 Kg/lit can be used as an alternative fuel.

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E. Flash point

Flash point is the temperature that indicates the overall flammability hazards in the presence of air; higher flash points make for safe handling and storage of biodiesel. The flash points of Neem oil biodiesel is 124^0C, which are higher than that of HSD (60^0C-80^0C) which makes for safe handling and storage.

F. Sulphur contents

The most valuable result is the reduction and absence of percentage of total sulphur contents in Neem oil biodiesel is that will result in reduction of SO_x in exhaust gases which is one of the reasons of acid rain.

G. Cloud point and pour point

Cloud point is the temperature at which a cloud of wax crystals first appear in the oil when it is cooled. The pour point is the lowest temperature at which the oil sample can still be moved. These properties are related to the use of biodiesel in colder region. The cloud points of Neem oil biodiesel is 12^0C and the pour point are 9^0C. Pour point and cloud point of all oils were almost within the specified range. Argued that the cloud points were affected by the presence of monoglycerides while the pour points were not affected

H. FT-IR Analysis For Fatty Acid Methyl Esters

The FT-IR spectra in the mid-infrared region have been used to identify functional groups and the bands corresponding to various stretching and bending vibrations in the samples of oil and biodiesel. The FT-IR spectrum of biodiesel prepared from neem seed oil chemical profile/composition results as illustrated in Figure 9 and the various functional groups which were identified are tabulated in Table 7. The position of carbonyl group in FT-IR is sensitive to substituent effects and to the structure of the molecule. The methoxy ester carbonyl group in Neem oil biodiesel was appeared at 1741.1 cm^{-1} and 1743 cm^{-1} respectively. The band appeared at 3465 cm^{-1} showed the overtone of ester functional group. The peak observed at 1014 cm^{-1} which corresponds to C-O. This functional group transformation has confirmed the formation of biodiesel, as the major functional group is the methyl ester.

Table 7 Chemical Composition of Neem oil biodiesel based on FT-IR analysis

Sample type	Catalyst	C=O (cm^{-1}) Ester Carbonyl	C-O(cm^{-1})
Neem oil biodiesel	NaOH	1741	1014.2

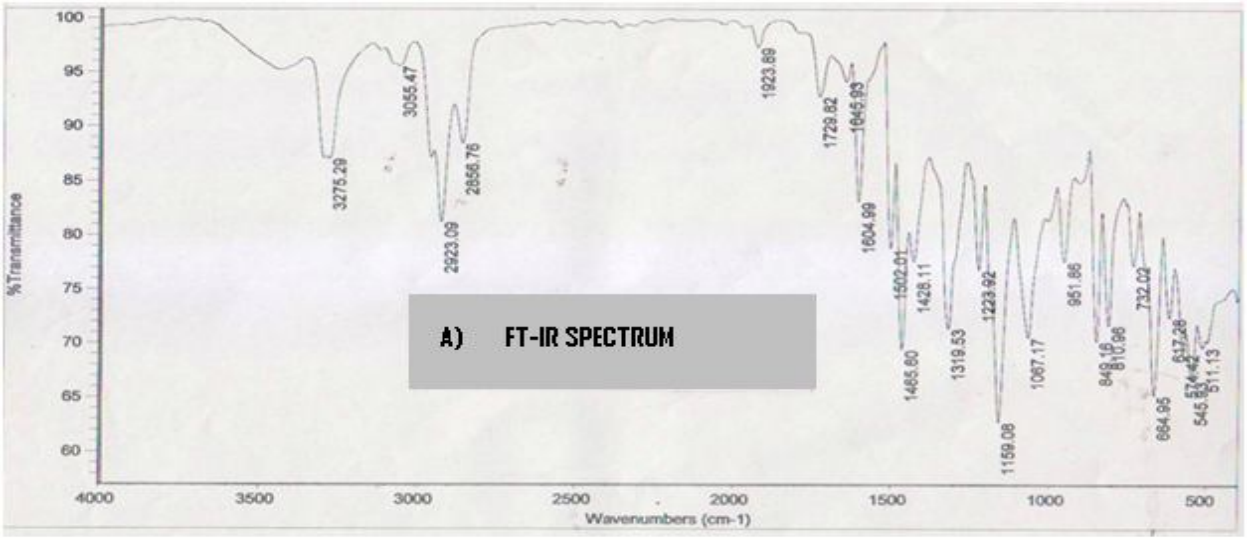


Fig. 9 Neem oil biodiesel chemical profile of FT-IR spectrum

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I. The FA compositions of Neem oils

The compositions of fatty acids of Neem are analyzed by using gas chromatography. The use of mass spectrometer would eliminate any ambiguities about the nature of eluting materials since mass spectra unique to individual compounds would be obtained. Fatty acids (FA) components of Neem oil mentioned in the table.

Table 8 Fatty acid composition of Neem oil

FA	Carbon number	Neem oil
Myristic Acid	C14:0	-
Palmitic Acid	C16:0	14.9
Stearic Acid	C18:0	14.4
Oleic acid	C18:1	61.9
Linoleic Acid	C18:2	7.5
Linoleic Acid	C18:3	-
Arachidic Acid	C20:0	1.3
Eicosenic acid	C20:1	-
Docosanoic acid	C22:0	-
Tetracosanoic acid	C24:0	-

Oleic acid was the most common FA in Neem oil. Neem oil containing 61.9 % oleic acid, Palmitic Acid (14.9), Stearic Acid (14.4), Linoleic Acid (7.5), Arachidic Acid (1.3).The fatty acids like Eicosenic acid, Docosanoic acid and Tetracosanoic acids were absent in Neem oil. Oleic acid is the major unsaturated fatty acids found in Neem oil.

The Gas Chromatography test analysis was performed to measure the various factors of bio-oil, presence of FFAs composition and different types of hydrocarbons. The fatty acids of bio-oil organic compounds were measured by the test method of gas chromatography.

GC-MS analysis is an extensively used analytical technique to quantify chemical composition, structure and type of FAMEs present in biodiesel. Gas chromatography analysis of neem oil biodiesel as discussed as follows. The flow rate of carrier gas fixed at 1 mL/min and the GC was operated at 58 min for the helium flow rate of 1 mL/min. The result of the chromatography analysis of neem oil biodiesel was shown in Figure 10. The percentage of un-saturated fatty acid is 80%, while the percentage of saturated fatty acid is 20%. It has been shown that biodiesel fuel with more unsaturated fatty acid composition has more density but has less viscosity, lower cetane number, and moderate color comparison. Further confirmation of transesterification of triglycerides to fatty acid methyl ester was obtained from gas chromatogram. Each peak corresponds to Methyl Ester and identified from the library match software. FAMEs were identified by arranging the retention time data; moreover, verification was made by GC internal standards.

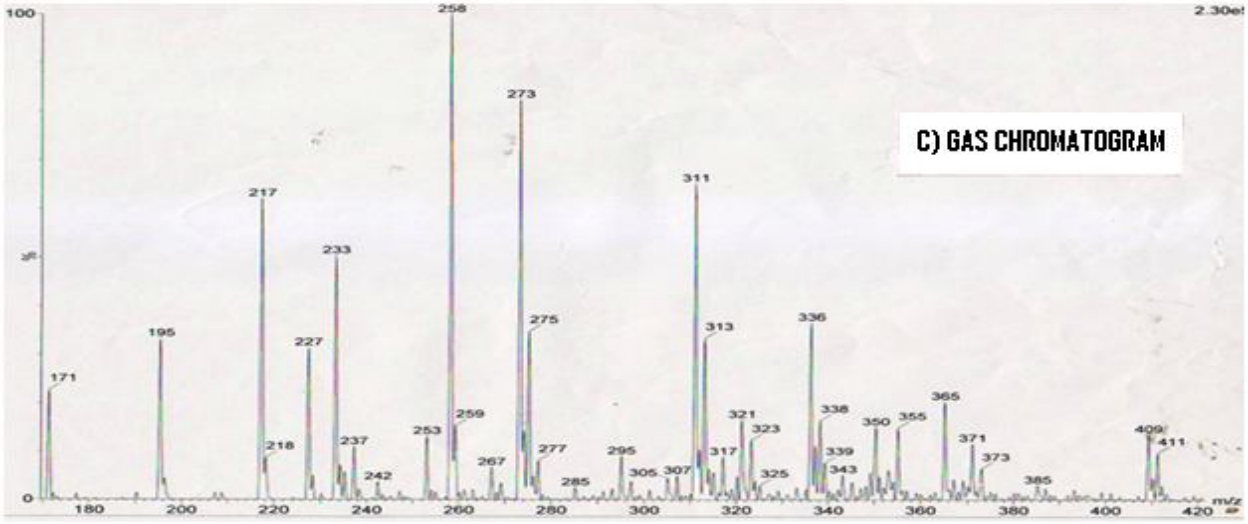


Fig. 10 Neem oil biodiesel chemical profile Gas chromatogram

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V. CONCLUSIONS

In this study, an optimized protocol for biodiesel production from non-edible seeds of Neem (*Azadirachta indica* A. Juss.) converted into fatty acid methyl esters (FAME) through base catalyzed Tran's esterification process. It can be concluded that maximum biodiesel yield from Neem can be achieved at 6:1 molar ratio of Methanol and oil,0.7%(w/w_{oil}) NaOH, at 60^0C reaction temperature and 2hr reaction time followed by 3-4 times gentle washing of the biodiesel with 60^0C distilled water (10% v/v). Biodiesel from these sources was analyzed by using Gas chromatography and FT-IR techniques. The growing demand for fuel and the increasing concern for the environment due to the use of fossil fuel have led to the increasing popularity of biofuel as a useful alternative and environmentally friendly energy resource. The presence of hydrocarbon groups indicates the potential usage of bio-oil as the alternate source of energy. The formation of biodiesel was confirmed by FT-IR analysis. The conversion of the ester functional group into methyl esters in biodiesel verified the success of the reaction. This optimized Trans esterification process gives a higher yield of biodiesel that has properties comparable to those of petroleum diesel.

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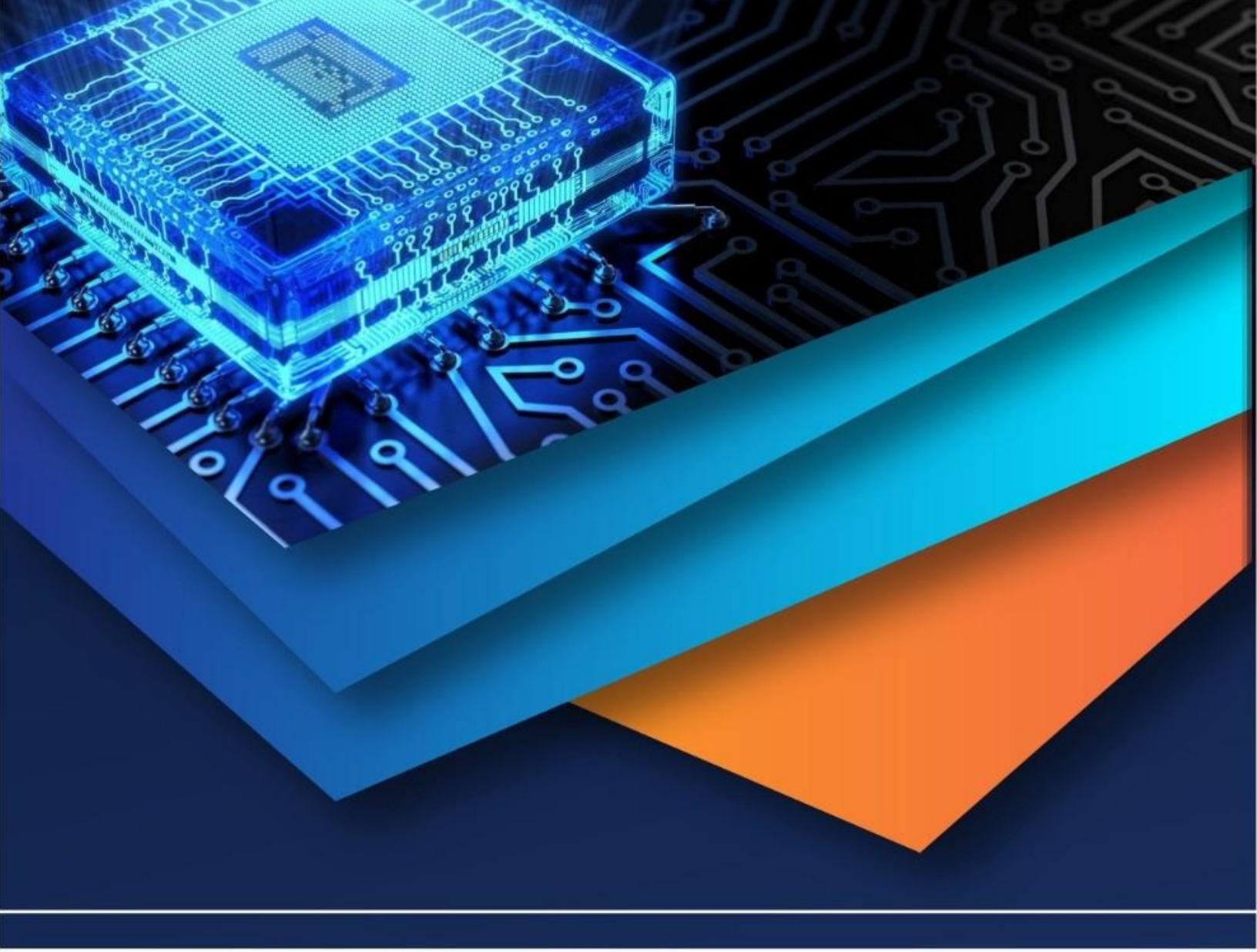
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