



Original Research Article

Biodiesel Production from *Thevetia peruviana* Seed Oil using FTIR and GC-MS Techniques to Investigate the Conversion Process

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ABSTRACT

Biodiesel production from *Thevetia peruviana* seed oil was investigated using Fourier transform infrared (FTIR) and gas chromatography-mass spectrometry (GCMS) techniques. Sample seed kernels were processed using mechanical press and the oil extracted. Agilent technologies FTIR spectrophotometer was used to obtain the oil infrared spectrum. A fused silica column, filled with Elite-5MS (95% dimethylpolysiloxane 5% biphenyl, 30 m × 0.25 mm ID × 250 μm df) on GCMS-QO2010 Shimadzu Japan equipment. The NIST library database was used to compare and identify the chemical compounds. The FTIR results of crude oil and the derived biodiesel indicates functional groups reduction from 8 peaks to 7 peaks, while the GCMS data showed an increase in the chemical compounds from 9 peaks to 10 peaks. The GCMS analysis confirmed the presence of fatty acids containing most of the functional groups identified in the FTIR. The dominant FTIR compounds as characterized were 73.57% fatty acids and 19.04% naturally occurring fatty acid methyl esters in the crude oil. The produced biodiesel is mainly oleic acid (C₁₈H₃₄O₂) (36.60% area), palmitic acid (C₂₆H₅₂O₂) (28.31% area), and 11-octadecenoic acid, methyl ester (C₁₉H₃₆O₂) (10.97% area). The GC-MS dominant biodiesel compounds were 62.87% fatty acid methyl esters, 19.28% fatty acids, 11.42% alkanals, 4.14% alkanols, and 2.28% ether groups, having good fuel properties. The useful biodiesel fuel from this feedstock seed oil will reduce waste and add value.

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1. INTRODUCTION

Emissions from internal-combustion (IC) engines have constituted a major source of greenhouse gases. Other toxic pollutants like particulate matter (PM) and nitrogen oxides (NO_x) are also generated from the use of IC engines (Hellier *et al.*, 2018). These toxic pollutants have devastating effects on the global climate. Due to increase in population of humans globally, energy demand is continuously increasing and fossil fuels reserves are depleting (Adama, 2021). There is an urgent need to replace petroleum liquid fuels because of

the energy issues and environmental problems associated with fossil fuel combustion in IC engines (Chizoo *et al.*, 2018). The depletion of fossil fuel reserve has given biodiesel an increased interest because of its potential to serve as an alternative fuel (Shah *et al.*, 2016). Biodiesel is an alkyl ester of fatty acids synthesized from animal fats and vegetable oils (Kaisan *et al.*, 2018). Other advantages of biodiesel include direct use in diesel engines with limited modification to the engine, biodegradable, environmental friendly, renewable and it does not contain sulfur nor aromatic compounds (Shah *et al.*, 2016).

For instance; Mustapha *et al.*, (2020) used alkaline refinement to produced biodiesel from waste cooking oil. The alkaline treated cooking oil gave improved fuel properties than waste cooking oil. The leaves of *Pandanus candalabrum* were characterized by Ogwuche and Edema, (2020) using FTIR and GC-MS analyses. It showed that Kaur-16-ene was the most abundant component with 84.62% percentage abundance. Carboxylic, and renes functional group were identified at 2958.08 cm^{-1} , and 1457.68 cm^{-1} wavenumber respectively. Samuel *et al.*, (2019) produced biodiesel from rubber seed oil. The highest yield of 92.5% rubber seed oil ethyl ester was obtained at 40 min reaction time, KOH of 4.5wt% and ethanol/oil molar ratio of 6/1. FTIR and GCMS analyses on *Canna indica* was investigated by Bolale *et al.*, (2019). The chemical compounds in African Mahogany were established through GCMS analysis by Ishaya *et al.*, (2020a). The physico-chemical and GC-MS analysis of *Thevetia peruviana* seed oil was investigated by Warra, (2017) for soap production. The results support the application of the seed oil for cosmetics. The GC-MS result showed presence of di-alpha-tocopherol. The FTIR results showed presence of aromatic O-H stretch (3300 cm^{-1}) and aromatic C=C stretch (1451 and 1640 cm^{-1}). Kaisan *et al.*, (2018) carried out comparison of Jatropa and Neem seed biodiesel, Results showed that Jatropa biodiesel contained 91.9% methyl ester and Neem biodiesel contained 70.2% methyl ester. Kavipriya *et al.* (2018) used FTIR and GC-MS to analyze the leaf extract of *Cassia alata*. The GCMS results showed 1-butanol, 3-methyl-1,6-anhydro-beta-D-glucopyranose, oxirane, and oleic acid. The FTIR results showed sulfates, sulfonamides, sulfones, aromatic, alkane, alkenes, nitrile, and amide functional groups. Ofogebu and Kelle, (2013) determined the edibility of *Thevetia peruviana* seed oil using GC-MS, FTIR and UV-VIS technique. The results showed that the oil has nutritional benefits provided the traces of cyanide are removed.

Thevetia peruviana plant is a member of the *Apocynaceae* (dogbane) family in the *Gentianales* order. *Thevetia* or yellow oleander or “Be-Still Tree” as it is commonly known, originate from the tropical and subtropical zones of Central America. *Thevetia peruviana* is a quick growing evergreen ornamental shrub or small tree, having upper range of 3.6 m to 4.7 m in height. It tolerates a wide range of environmental conditions, even in limestone conditions if not in volcanic island. *Thevetia* plants contain the poisonous alkaloids (*thevenine*). *Thevetia* has no known disease problems. It has a prolific seed production of more than a thousand seeds per square metres (>1000/m²). It is dispersed by water, birds and other animals. In this study, *Thevetia peruviana* seeds oil was used to produce biodiesel. This is because it is non-edible due to its toxic nature, and its oil is known to contain saturated and monounsaturated fatty acids. FTIR and GC-MS techniques were used in this study to identify the functional groups present in the fluids via FTIR, and the GC-MS specify and quantify each chemical compound in the fluid. The tests are used to investigate the transesterification of some essential organic compounds from fatty acids to fatty acids methyl esters, and the implication on the biodiesel. The contribution of this study is to convert non-edible, toxic seed oil into useful biodiesel fuel. This will reduce waste, add value as it can serve as alternative fuel to depleting fossil diesel fuel and for safer environment.

2. MATERIALS AND METHODS

2.1. Seed Collection and Preparation

Thevetia peruviana seeds were collected along Stephen Shekari Road, Barnawa, Kaduna, Nigeria. The collected seeds were separated and cleaned of fruits pulp/fibres as preparation for the oil extraction. The seeds kernels were sun dried for fifteen (15) days for 6 hours daily, starting 10:00 hour to 16:00 hour to reduce their moisture content. The dried kernels were roasted, deshelled. The kernels (Endosperm – the soft

fleshy part) were further dried by heating at 50 °C using a dryer for one hour before crushing in a grinder to powder form. The dried and ground powders were weighed, and values were recorded.

2.2. Oil Extraction Procedure

For this study, the mechanical expeller press method outlined by Kaisan *et al.* (2016) was adopted to extract the oil. The machine was operated at 120 bars to extract oils from *Thevetia peruviana* kernels powder, preheated at 50 °C. The block diagram illustrating the oil extraction process is shown in Figure 1.

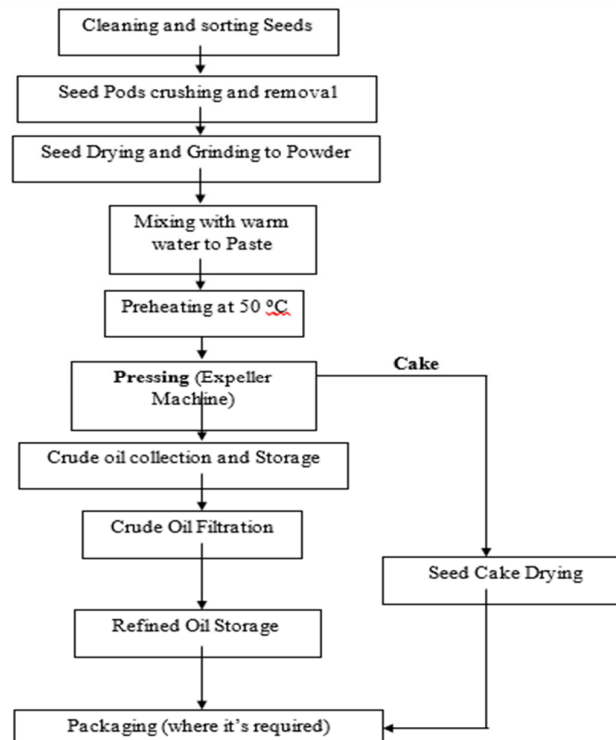


Figure 1: Flow diagram showing the oil extraction process

2.3. Biodiesel Production Process

For high free fatty acid (FFA) feed stocks, pre-treatment by acid-catalysis process – esterification was conducted. The next stage is the transesterification process, which involves crude glycerine and biodiesel separation, crude biodiesel purification by neutralization, methanol and catalyst recovery, drying of the biodiesel, conducted in this order as shown in Figure 2.

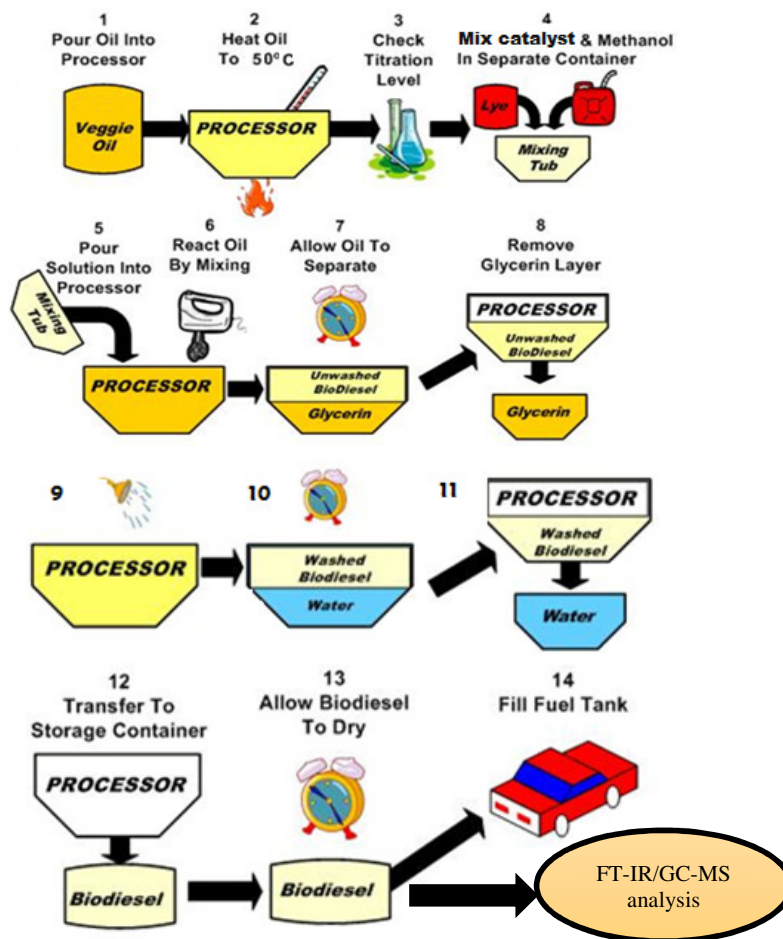


Figure 2: A simplistic approach for production of biodiesel

2.4. Fourier Transform Infrared (FTIR) Spectroscopy

Agilent technologies FT-IR Spectrophotometer was used to obtain the infrared spectrum of samples at stage 1 and stage 13. Potassium bromide disc method was used to obtain the IR spectra of samples. 650 to 4000 cm^{-1} scanning range was used. In this study, functional groups were identified in the crude oil (veggie oil) and biodiesel samples using the FT-IR spectra.

2.5. Gas Chromatography-Mass Spectrometry (GC-MS)

GCMS-QO2010 Shimadzu Japan was used in this analysis. A fused silica column, filled with Elite-5MS (95% dimethylpolysiloxane 5% biphenyl, 30 m \times 0.25 mm ID \times 250 μm df). Helium as carrier gas flowing constantly at 1 mL/min was used to separate the components. Temperature of injector was set at 250 $^{\circ}\text{C}$ for the chromatographic run. The extract (1 μL) was injected into the instrument and the temperature was 70 $^{\circ}\text{C}$ (0 min); followed by 280 $^{\circ}\text{C}$ at the rate of 10 $^{\circ}\text{C min}^{-1}$; and 280 $^{\circ}\text{C}$, where it was kept for 5 minutes. The mass detector conditions were set at transfer line temperature at 250 $^{\circ}\text{C}$; ion source temperature of 200 $^{\circ}\text{C}$; and ionization mode electron impact of 70 eV, a scan time of 0.5 seconds and scan interval of 0.1 seconds respectively. In this study, compounds were detected in the crude oil (veggie oil in stage 1), biodiesel in stage 13 and their respective quantities with GC-MS spectrometer.

2.6. Compounds Identification

National Institute Standard and Technology (NIST) library was used as the database to compare and interpret results. Compounds were identified according to molecular mass, structure and fragments calculated. The unknown components spectrum was compared with that of the NIST library databases, and the closest match is taken.

3. RESULTS AND DISCUSSION

3.1. Fourier Transform Infrared Spectroscopy (FTIR) Analysis

3.1.1. Crude *Thevetia peruviana* oil

The crude *Thevetia peruviana* oil was subjected to FTIR test and the resulting spectra is presented in Figure 3 and the functional groups classification are presented in Table 1. From the analysis of the results from the FT-IR spectrum shown in Figure 3 and Table 1, =C-H bond was detected at 3008 cm⁻¹, C-H bond at 2855-2922 cm⁻¹, C=O at 1744 cm⁻¹, C-H terminal alkane bond at 1461 cm⁻¹, C-O carboxylic group at 1379 cm⁻¹, C-O ester group peaks at 1103-1236 cm⁻¹, and C-H bend at 723 cm⁻¹. Attia et al. (2020) observed absorption bands of =C-H at 3007 cm⁻¹, CH₃ and CH₂ at 2855 cm⁻¹ and 2925 cm⁻¹ respectively, C=O at 1742 cm⁻¹ and C-O at 1168 cm⁻¹. Singh and Rahin, (2020) also observed fatty acids with double bonds within the range of 3000 – 3010 cm⁻¹. Sharma et al. (2021) assigned 3009, 2923, 2853 cm⁻¹, to CH, CH₂ and CH₃ vibrations and 1708.48 cm⁻¹ to C=O vibration. This study finding is consistent with those of previous work (Attia et al., 2020; Singh and Rahin, 2020; Sharma et al., 2021). Based on this FTIR result, alkanes, alkenes, carboxylic and esters functional groups were detected in crude *Thevetia peruviana* oil. The alkane will not be affected by transesterification; therefore, they will be maintained in the biodiesel, likewise the alkenes and ester groups. The alkanes and alkenes implied saturation and unsaturation of the fatty acid (FA) chain. The carboxylic group denotes the triglyceride is composed of fatty acids, and it is this group that takes part in the transesterification reaction.

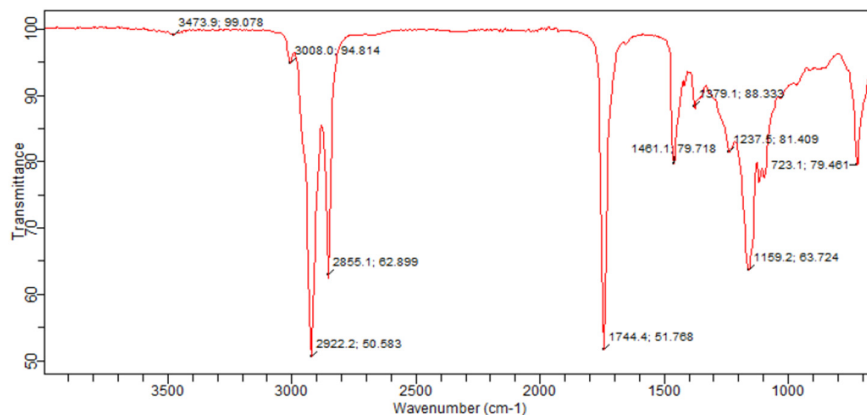


Figure 3: FTIR spectra of crude *Thevetia peruviana* oil

Table 1: Functional groups in crude *Thevetia peruviana* oil

Wavelength (cm ⁻¹)	Functional group
3474	N-H very weak stretch (amine)
3008	=C-H weak stretch (alkene)
2922, 2855	C-H strong stretch (alkanes)
1744	C=O strong stretch (alkanoic & ester)
1461	C-H medium stretch (terminal alkane bend)
1379	C-O weak stretch (alkanoic)
1237-1159	C-O medium stretch, (esters)
723	C=C-H medium stretch (alkene bending)

3.1.2. *Thevetia peruviana* biodiesel

The *Thevetia peruviana* biodiesel produced was subjected to FTIR test and the resulting spectra is presented in Figure 4. Also, the functional groups classification is presented in Table 2. Similarly, from the FT-IR spectrum in Figure 2 and results in Table 2, C-H bond was detected at 2855 and 2922 cm^{-1} , C=O at 1740 cm^{-1} , C-H terminal alkane bond at 1461 and 1438 cm^{-1} , C-O carboxylic group at 1364 cm^{-1} , C-O ester group peaks at 1244 and 1196 cm^{-1} , C-O-C at 1013 cm^{-1} and C-H bend at 723 and 879 cm^{-1} . Dass et al. (2018) observed that 1744.4, 1237.6, and 723.1 cm^{-1} were marched to C=O, C-O vibrations and C-H bending respectively in a biodiesel produced from Mahogany fruit shell. Cheah et al. (2020) assigned peaks at 1059 cm^{-1} to C-O stretching, 2900 cm^{-1} to alkane C-H stretching, and 1700 cm^{-1} to ester C=O stretching. This study is consistent with those of previous work (Dass et al., 2018; Cheah et al., 2020). Comparing Figure 1 and Figure 2, it was deduced that due to the transesterification reaction, there was a shift in C=O peak from 1744 to 1740 cm^{-1} , while an additional peak of C-H was observed at 1438 cm^{-1} . C-O carboxylic group shifted from 1379 to 1364 cm^{-1} , C-O ester shift from 1237 – 1159 cm^{-1} to 1244 – 1196 cm^{-1} . Based on this result, it shows the presence of alkanes, alkenes, carboxylic, and esters functional groups in the processed biodiesel. The implication of alkanes in the biodiesel is that the biodiesel will possess short ignition delay and high cetane number. The side effects of alkanes on the biodiesel are low pour and cloud point. Alkenes on the other hand will impart good fluidity to the biodiesel but will be challenged with oxidation instability and lower heating value.

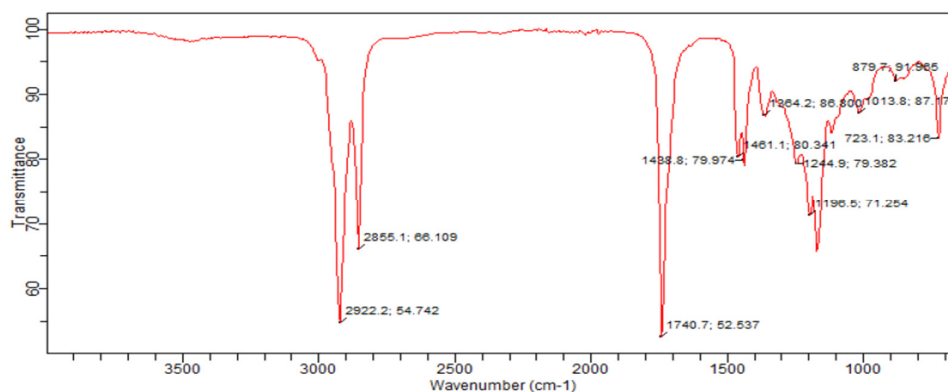


Figure 4: FTIR spectrum of *Thevetia peruviana* biodiesel

Table 2: Functional groups in *Thevetia peruviana* biodiesel

Wavelength (cm^{-1})	Functional group
2922, 2855	C-H strong stretch (alkanes)
1740	C=O strong stretch (alkanoic & ester)
1461, 1438	C-H medium stretch (terminal alkane bend)
1364	C-O weak stretch (alkanoic)
1244-1196	C-O medium stretch, (esters)
1013	C-O-C weak stretch (alkanoate)
723-879	C=C-H medium stretch (alkene bending)

3.2. Gas Chromatography-mass Spectrometry (GC-MS) Analysis

3.2.1. Crude *Thevetia peruviana* oil

The chemical profile of the crude *Thevetia peruviana* oil was established through the GCMS test. The resulting chromatograph is presented in Figure 5. The functional groups in the oil are indicated in Table 3. The result synthesis from the GCMS spectrum of crude *Thevetia peruviana* in Figure 5, and in Table 4. Nine (9) compounds were identified. Dominant compounds include oleic acid ($\text{C}_{18}\text{H}_{34}\text{O}_2$) at retention time of

18.117 min having 36.60% spectral area, palmitic acid ($C_{26}H_{32}O_2$) at retention time 16.432 min occupying 28.31% spectral area, 11-Octadecenoic acid, methyl ester ($C_{19}H_{36}O_2$) at retention time 17.426 min with a mass spectra area 10.97%, brassidic acid ($C_{22}H_{42}O_2$) occupying 8.66% mass spectra area at retention time 20.724 min, and 1, 15-pentadecanediol ($C_{15}H_{32}O_2$) at retention time 18.005 min with a mass spectra area of 5.03%. Ofogebu and Kelle, (2013) observed palmitic acid, trans-oleic acid and behenic acid methyl ester in *Thevetia* seed oil. Similarly, Warra, (2017) identified brassidic acid, oleic acid and palmitic acid in TP seed oil. This study results aligned consistently with those of previous researchers (Ofogebu and Kelle, 2013; Warra, 2017; Ishaya *et al.*, (2020b).

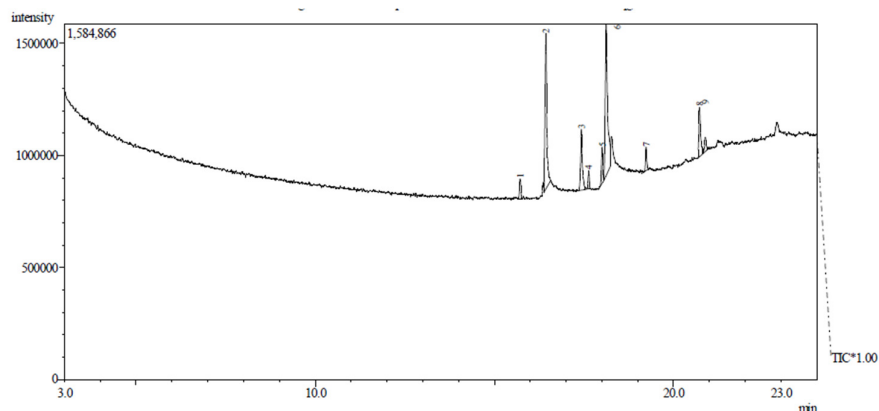


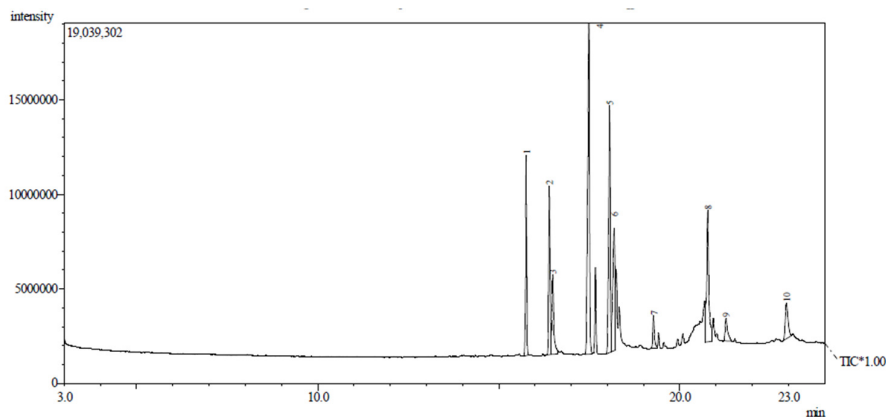
Figure 5: GC-MS spectrum of crude *Thevetia peruviana* oil

Table 3: Compounds present in crude *Thevetia peruviana* oil

RT	Area %	Library/ID	Formula
15.718	2.55	Butanoic acid, methyl ester	$C_5H_{10}O_2$
16.432	28.31	Palmitic acid	$C_{16}H_{32}O_2$
17.426	10.97	11-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$
17.629	2.47	Behenic acid, methyl ester	$C_{23}H_{46}O_2$
18.005	5.03	1, 15-Pentadecanediol	$C_{15}H_{32}O_2$
18.117	36.60	Oleic acid	$C_{18}H_{34}O_2$
19.233	3.05	Decanoic acid, decyl ester	$C_{20}H_{40}O_2$
20.724	8.66	Brassidic acid	$C_{22}H_{42}O_2$
20.886	2.36	Hexadecanohydrazide	$C_{16}H_{34}N_2O$

3.2.2. *Thevetia peruviana* biodiesel

Similarly, the chemical profile of the crude *Thevetia peruviana* biodiesel was established through the GCMS test and the resulting chromatograph is presented in Figure 6. Again, the functional groups in the oil are indicated in Table 4. Based on this GC-MS result, the oleic acid is a monounsaturated fatty acid with 18 carbon atoms. Mono-unsaturation implies the intended biodiesel will have good fluidity property. Palmitic acid is a saturated fatty acid with 16 carbon atoms. Saturation in the carbon chain implies the intended biodiesel with possess greater combustion efficiency and high cetane number. Although it can also contribute to higher viscosity and higher melting point. The long fatty acid chain (16-18 carbon atoms) will impart high heating value and short ignition delay on the intended biodiesel.

Figure 6: GC-MS spectra of *Thevetia peruviana* biodieselTable 4: Compounds present in *Thevetia peruviana* biodiesel

RT	Area %	Library/ID	Formula
15.756	10.16	Palmitic acid, methyl ester	C ₁₇ H ₃₄ O ₂
16.398	8.40	Palmitic acid, ethyl ester	C ₁₈ H ₃₆ O ₂
16.493	6.92	Palmitic acid	C ₁₆ H ₃₂ O ₂
17.485	25.19	11-Octadecenoic acid, methyl ester	C ₁₉ H ₃₆ O ₂
18.059	16.75	9-Octadecenoic acid, ethyl ester	C ₂₀ H ₃₈ O ₂
18.183	12.36	Oleic acid	C ₁₈ H ₃₄ O ₂
19.273	2.28	Stearyl vinyl ether	C ₂₀ H ₄₀ O
20.273	11.42	9-Octadecenal	C ₁₈ H ₃₄ O
21.272	2.37	Hexadecanoic acid, 2,3-dihydroxypropyl ester	C ₁₉ H ₃₈ O ₄
22.943	4.14	2-Methyl-Z,Z,-3,13-Octadecadienol	C ₁₉ H ₃₆ O

From the GC-MS spectrum of *Thevetia peruviana* biodiesel in Figure 6, and results in Table 4. Ten (10) compounds were identified. Dominant compounds include: 11- Octadecenoic acid, methyl ester (C₁₉H₃₆O₂) at retention time of 17.485 min having 25.19% spectral area, 9-Octadecenoic acid, ethyl ester (C₂₀H₃₈O₂) at retention time 18.059 min with a mass spectra area 16.75%, oleic acid (C₁₈H₃₄O₂) at retention time of 18.183 min occupying mass spectra area of 12.36%, palmitic acid, methyl ester (C₁₇H₃₄O₂) at retention time 15.756 min occupying 10.16% spectral area, palmitic acid, ethyl ester (C₁₈H₃₆O₂) at retention time 16.398 min occupying 8.40% spectral area, palmitic acid (C₁₆H₃₂O₂) at retention time 16.493 min occupying 6.92% spectral area. 9-Octadecenal (C₁₈H₃₄O) at retention time 20.273 min occupying 11.42% spectral area. 62.87% of the total area of the biodiesel is occupied by fatty acid methyl esters (FAME), 19.28% by fatty acids (FA), 11.42% by alkanals, 4.14% by alkanols and 2.28% by ether groups.

Based on these results the transesterification reaction caused the reduction of FA from 73.57% in the crude oil to 19.28% in the biodiesel. The formation of esters due to the transesterification reaction impart an increase in FAME from 19.04% in the crude oil to 62.87% in the biodiesel. The presence of FA in the biodiesel will increase the total acid number (TAN) of the biodiesel. This will aid chemical corrosion of moving engines parts. The presence of FAME in the biodiesel will allow good fuel properties such as high energy per unit volume, low viscosity, and short ignition delay. The presence of other oxygen bearing groups like alkanals, alkanols and ether in the biodiesel will increase the combustion efficiency of the fuel.

4. CONCLUSION

In this study, biodiesel production from *Thevetia peruviana* seed, using FTIR and GC-MS techniques to investigate the conversion process. The following conclusions are drawn from the results:

1. The FTIR analyses of the crude *Thevetia peruviana* seed oil showed the presence of alkanes, alkenes, alkanolic and esters functional groups. The GC-MS analyses confirmed the presence of fatty

acids containing most the functional groups identified in the FTIR. The dominant compounds were: Oleic acid ($C_{18}H_{34}O_2$) 36.60% area, Palmitic acid ($C_{16}H_{32}O_2$) 28.31% area, and 11-Octadecenoic acid, methyl ester ($C_{19}H_{36}O_2$) 10.97% area. The crude oil was characterized by 73.57% fatty acids and 19.04% naturally occurring fatty acid methyl esters.

2. The GC-MS analyses confirmed the conversion of fatty acids to fatty acid methyl esters, containing most of the functional groups identified in the FTIR. The dominant compounds were 62.87% fatty acid methyl esters, 19.28% fatty acids, 11.42% alkanals, 4.14% alkanols, and 2.28% ether groups.
3. From this study, it was observed that transesterification reaction can be used to process *Thevetia peruviana* oil to biodiesel, having good fuel properties.

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6. CONFLICT OF INTEREST

There is no conflict of interest associated with this work.

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