



Determination of Fatty Acid Methyl Ester Composition of Mango (*Mangifera indica* L.) Kernel Oil and Tribological Properties of its Triesters

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ABSTRACT: Conventional process of Soxhlet extraction with n-Hexane was used to extract Mango kernel (*Mangifera indica* L.) oil for determination of fatty acid methyl ester composition of mango (*Mangifera indica* L.) kernel oil and tribological properties of its triesters and a percentage extractable of 8.4 % was obtained. A two-step process of esterification and transesterification was employed to produce fatty acid methyl ester (FAME) from *M. indica* kernel oil refers to as Mango kernel methyl ester (MKME). Double esterification of the MKME with trimethylolpropane (TMP) in the presence of sodium methoxide yielded 95 % Mango oil based trimethylolpropane ester (MOTE). Another portion of the MKME was used to synthesize Biolubricant using the conventional epoxidation process to produce 70 % Mango oil based epoxidised biolubricant (MEB-L). Tribological test was conducted on the MOTTE and MEB-L at coefficient of friction ranging from 0.03 to 0.18 μ on Aluminum alloy with load of 8.00 N. The mean value of coefficient of friction were determined to be 0.083 μ and 0.080 μ for MOTTE and MEB-L respectively. Although MEB-L present a slightly better frictional performance than MOTTE, both values are within an acceptable mean with regards to the suitability of MOTTE and MEB-L as potential substitute to conventional lubricant.

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M. indica belong to the family Anacardiaceae and the genus *Mangifera*, the fruits of most of the different species within this genus are edible. They are tropical trees that grow well in Asia and Africa (Umaru *et al.*, 2014). The fruit is widely produced in Nigeria and Guinea in West Africa. The raw mangoes are utilized in making pickles, curries and juice. The seed kernel of the ripe fruit is in great demand in the cosmetic and confectionaries (Torres-León *et al.*, 2016). However, huge heaps of Mango fruits are left to rotten in villages where they are produced across Nigerian, as there are no means of moving them to urban or commercial/industrial centers for sales (Tank *et al.*, 2016). Mango kernel contains 7-15 % fat rich in stearic, oleic and palmitic acids (Akhter *et al.*, 2016). Mango kernel of a ripe fruit are rich in fatty acid compared to the unripe fruit which is rich in aromatic

compounds which possess some antioxidant properties. Research reports indicated that *M. indica* oil is a non-edible oil (Tank *et al.*, 2016 and Nigade *et al.*, 2015) and little has been reported on optimum utilization of the oil and its methyl ester. Mango lipid have fatty acids composed of oleic acid (43.8 % w/w), linoleic acid (3.6 % w/w), stearic acid (43.2 % w/w), palmitic acid (4.9 % w/w), linolenic acid (2.3 % w/w), and arachidic acid (2.2 % w/w) (Chakraborty & Das, 2017 and Dzinga & Kapoor, 1985). It has been shown that when a single step based catalyzed transesterification is done; no biodiesel will be formed (Ogunsuyi, 2012; Nigade *et al.*, 2015 & Oyelaran *et al.*, 2018). Transesterification which is the best among the process of biodiesel formation was carried out by Umaru *et al.*, 2014 in an experiment in which two process parameters were studied. The effect of

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temperature and catalyst concentration on the transesterification of *M. indica* seed oil with methanol using potassium hydroxide as catalyst was reviewed. Result shows that increase in temperature results in increase in the amount of biodiesel yield. A yield of 83 wt % was achieved at optimum temperature of 60 °C. Similarly, the same trend was obtained for the effect of catalyst concentration; with optimum yield of 80 wt % at 1 w/v % (Umaru *et al.*, 2014). The biodiesel obtained meets the ASTM and EN standard. However, two-step process involving the pre-treatment step (esterification) with concentrated acid and transesterification step are involved for synthesis of biodiesel from feedstock with relatively high free fatty acid value (Urrutia *et al.*, 2016). Biodiesel can be blended with petro diesel in varying percentages to produce a blend of fuel with reduced environmental hazard and better fuel performance. There has not been much reports regarding the use of *M. indica* as a potential substitute to conventional lubricant which are known for their non-biodegradability and toxicity as 40 % of these toxic oleo-chemicals are discharged into the environment (Obasa *et al.*, 2020). The need for environmentally friendly, biodegradable, non-toxic and renewable biolubricant cannot be overemphasized. The objective of this paper is to determine the fatty acid methyl ester content of Mango (*Mangifera indica* L.) kernel oil and the tribological properties of its triesters.

MATERIALS AND METHODS

Plant material: Mango fruit seeds were purchased from sellers of Mango fruit seeds at Ikanekpo, Ankpa local government area of Kogi State, Nigeria. The mango kernel was sun-dried at mean temperature of 33.5 °C, for two weeks to reduce the moisture content. The dried mango kernel was cleaned and sorted by removing the immature, broken kernel and unwanted materials. The kernel was pulverized with mortar and pestle; and extracted (25 g) with Soxhlet apparatus using n-hexane. The process was repeated five times to determine the mean oil yield.

Oil Extraction: The crushed pulp was packed into the extraction chamber of the soxhlet extractor and allowed to reflux for about eight hours at 65 °C. The extract filtered (to remove impurities) and evaporated using a rotary evaporator to isolate the free flow lipid from the solvent. The extracted oil was further evaporated on a magnetic stirrer at 120 °C to eliminate any moisture and residual solvent that may be present. The weight of the oil produced and the residue was measured to establish the percentage of the oil content. The oil gotten was weighed and the percentage oil yield calculated by Equation (1), as reported by Oyelaran *et al.*, 2018.

$$\text{Oil yield (\%)} = \frac{W_2 - W_1}{W} \dots\dots\dots (1)$$

Where, W = weight of mango kernel used (g); W_1 = weight of beaker (g); W_2 = weight of beaker with oil (g); $W_2 - W_1$ = weight of oil. (g).

Esterification: Esterification of mango kernel oil (MKO) was necessary due to high free fatty acid value, it was carried out in the presence of few drops conc. H_2SO_4 as a catalyst at 60 °C and excess methanol to obtain a mixture of fatty acid methyl esters and triglycerides. The sequence of the addition was MKO followed by mixture of methanol and conc. H_2SO_4 continuously stirred on a magnetic stirrer for one hour. The mixture was poured into the separating funnel for separation, the mixture separated into two layers of methanol at the top and fatty acid methyl esters/triglycerides mixture at the bottom. The bottom layer was collected and heated to remove remnant of methanol and allowed to stand for 24 hrs.

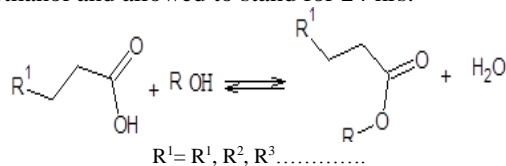


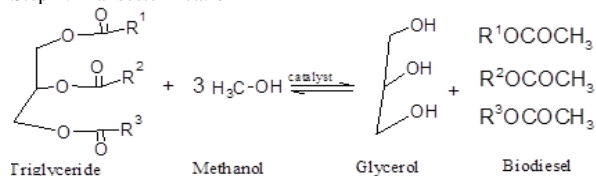
Fig 1. Esterification of Fatty acid.

Transesterification: The transesterification of MKO with methanol produces Mango kernel methyl ester (MKME) and glycerol catalyzed by both homogeneous and heterogeneous catalysts. Portions of MKO reacted with 25 cm³ methanol and 0.5 % catalyst calcium oxide (CaO), sodium hydroxide (NaOH) and potassium hydroxide (KOH) in separate beakers. Temperature was set at 60 °C on magnetic stirrer for 60 mins. Mixture transferred into the separating funnel and allowed to stand for 24 hrs for proper separation of the Biodiesel and Glycerol layer. The Glycerol layer was removed and the Biodiesel was washed with warm water to remove methanol and traces of the catalyst contained in the Biodiesel. Mango Kernel Methyl Ester (MKME) formed was heated to 120 °C to remove water and methanol still present before analyses commenced by FTIR, GC-MS and ASTM.

Double Transesterification Method (Biolubricant Synthesis): Double transesterification of MKMS was done by weighing 50 cm³ of the MKMS reacted with 5 % Trimethylolpropane in a 100 cm³ beaker. The mixed substance was then heated to about 120 °C for 2.5 hours while continuously been stirred on magnetic stirrer. 1 % w/w catalyst potassium methoxide was added slowly to the reaction mixture to avoid spillage. The resultant product Mango oil based trimethylolpropane ester (MOTE) was monitored by

FTIR as few drop of the product was taken at intervals and analyzed for reaction completion. It was then allowed to cool before being filtered, washed and analyzed by FTIR and ASTM standard.

Step 1: Transesterification



Step 2: Double Transesterification

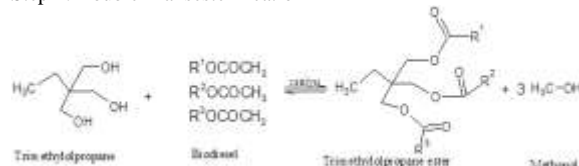


Fig 2. Transesterification and Double transesterification reaction

Epoxidation Method for Biolubricant Synthesis: The classical Prilezhaev method of epoxidation was employed for the synthesis of MEB-L. Portion of MKMS (40 cm³) was weighed into 250 cm³ beaker, 20 cm³ of acetic acid was added to the oil in the beaker and heated at 60 °C before 20 cm³ of hydrogen peroxide 30 % mixed with 1 % H₂SO₄ was added to the mixture dropwise to avoid a spillover of the reaction mixture, the sequence of these addition was reported by (Ashrafi *et al.*, 2017 & Cecilia *et al.*, 2020). The setup was heated for two hours on a magnetic stirrer and the progress of the reaction monitored at interval by FTIR for completion. The mixture was transferred into separatory funnel where oil layer (epoxide) was seen at the top and hydrogen peroxide/acetic acid clear aqueous layer was seen at the bottom. The aqueous layer was drained off and the epoxide washed repeatedly with warm water. The epoxide was heated to 120 °C to remove any water present.

Acetylation of MOTE and MEB-L: The FTIR of MOTE reveals there was some level of hydroxyl group present, the acetylation is to convert the hydroxyl group to an acetyl. 10 cm³ of the MOTE was introduced into a 50 cm³ beaker place on a magnetic stirrer and heated to 60 °C, 1 % acetic anhydride was added and allowed to heat for one hour. The completion of the reaction was confirmed by FTIR. Similarly, 10 cm³ of the epoxide was placed in 50 cm³ beaker and 1 % acetic anhydride was added and heated to 60 °C for 30 mins after which the mixture was transferred into the separatory funnel and 0.1 M of sodium hydroxide was prepared and added to the separatory funnel to neutralize the acetic acid formed. The white precipitate that appeared was drain off and

the tri/tetra esters formed washed with warm water severally until a transparent water layer not turbid was seen. The biolubricant MEB-L formed was drain off and heated above 120 °C to remove water and then analyzed by FTIR and ASTM standard.

Tribological Investigation Using Four Ball Tribometer: The tribometer has four balls, TR-30L-IAS, made by Ducom Instruments UK, was the tribological equipment used for the experiment. Each test lubricant was applied to a charged pot with three ball made of aluminum alloy of diameter 12.7 mm with hardness of 60 HRC and polish grade of 25 were loaded. The fourth ball was a fixed one and exert a load of 8 N on the bottom balls, chamber was kept at 80 °C according to ASTM D4172 and the reaction period was 460 s. The coefficient of friction (μ) experienced by the balls due to each lubricant was measured, the wear scare diameter on the balls was also measured. The results were presented as a plot. The coefficient of friction is denoted by $\mu = \text{Frictional force/Normal force}$.

FTIR Analysis: The FTIR analysis was performed at the Multi-User Science Research Laboratory, Ahmadu Bello University, Zaria. The analyses were done by using the Agilent MicroLab PC, the sample scans were 30 and background scans 16, with range of 4000 – 650 cm⁻¹.

Gas chromatography – Mass Spectroscopy: GC-MS analysis was performed at the Multi-User Science Research Laboratory, Ahmadu Bello University, Zaria. This was done by using a Mass Hunter GC-MS 2014, the GC oven temperature was operated from 150 °C to 325 °C at the rate of 40 °C /min. The gas employed as carrier gas was Helium, the inlet pressure set at 4.4867 psi, average velocity was 26.042 cm/sec. Column flow was 0.4684 mL/min, injector temperature: 250 °C. Injection mode: split. MS scan conditions: source temperature 230 °C, maximum temperature 250 °C, scanning speed was normal. The composition of MKO and MKME were identified by comparison with spectra of pre-identified compounds stored in the memory NIST library (2014) presented in the (Figure 4-8). The relative percentage abundance of each component was determined by comparing its average peak to the total areas under the peak.

RESULTS AND DISCUSSION

The soxhlet extraction of the mango kernel using n-hexane as the extracting solvent yielded 8.4 % MKO with density of 0.8900 gcm⁻³ (Table 1), this is in line with the observation of Akhter *et al.*, 2016 who reported that mango kernel contains oil yield ranging from about 7 to 15 percent.

Table 1. Physicochemical properties of MKO and MKME

Specifications	MKO	MKME	ASTM	EN	ASTM/EN
Yield wt. %	8.4 - 13.5%	83.6%	-	-	-
Density g/cm ³	0.89	0.85	-	0.86-0.90	ASTM-1298
Viscosity @ 40°C cSt	4	3.8	1.9-6.0	3.5-5.0	ASTM-445
Viscosity @ 100°C cSt	2	1.8	1.9-6.0	3.5-5.0	ASTM-445
Cloud point °C	25	18	-	-	ASTM-97
Pour point °C	11	8	-	-	ASTM-97
Total acid number (mg KOH/g)	5.5	1.9	0.50	0.50	ASTM-664
Iodine value	150.62	125.50	-	120	EN 14111
Saponification Value	222.47	-	-	-	-

Table 2. Fatty acid component of the MKO and their relative percentages

Common name	Symbol	Percentage of total weight
Myristic acid	C14:0	0.40
Palmitoleic acid	C16:1	0.44
Palmitic acid	C16:0	21.78
Linoleic acid	C18:2	40.85
Oleic acid	C18:1	25.60
Stearic acid	C18:0	3.92
Eicosanoic/Arachidic/Icosanoic acid	C20:0	1.28
Docosanoic/Behenic acid	C22:0	0.41
M-cresyl acetate & Others		5.31

Table 3. Presentation of some major esters that were detected in MKME from GC-MS analysis

Name of methyl ester	Percentage of total weight
Methyl tetradecanoate	0.41
Methyl hexadec-9-enoate	0.45
Hexadecanoic acid, methyl ester	17.85
cis-10-Heptadecenoic acid, methyl ester	0.10
Hexadecanoic acid, 14-methyl-, methyl ester	0.12
9,12-Octadecadienoic acid, methyl ester	47.83
Methyl stearate	17.75
Cyclopropanoic acid, 2-octyl-, methyl ester	0.15
cis-13-Eicosenoic acid, methyl ester	0.36
Methyl 18-methylnonadecanoate	1.39
Tricosanoic acid, methyl ester	0.07
Tetracosanoic acid, methyl ester	1.00
Hexacosanoic acid, methyl ester	0.09
Octacosanoic acid, methyl ester	0.09
Other components	12.34

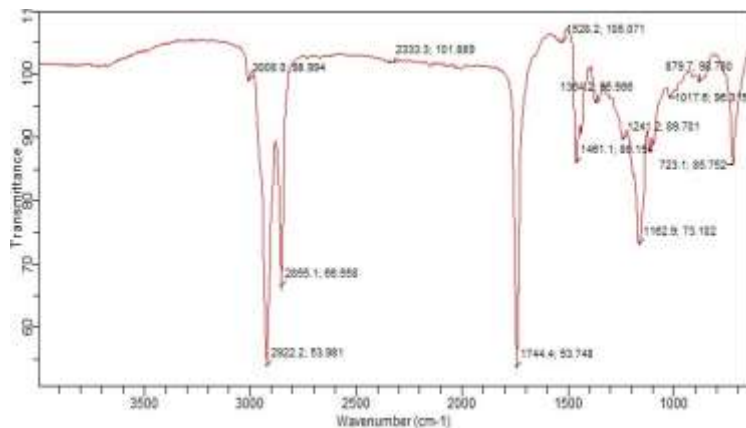


Fig 3. FTIR for mango kernel oil

It was also established in the work of Chakraborty and Das, 2017 that Ruhi mango has a yield of 8.11 percent and density of 0.8710 gcm⁻³ and this species is closely related to the species used in this work. The

viscosity at 40 and 100 °C of MKO and MKME which are 4.0 and 2.0, 3.8 and 1.8 respectively are within the ASTM standard of 1.9 to 6.0 (Table 1), making MKO suitable base stock for bio-lubricant formulation. The cloud and pour point values of MKO and MKME are relatively high as lower cloud/pour point values are preferred for low temperature fluidity, these would require blending with cold flow property improvers to enhance its fluidity. The total acid number (TAN), which is the measure of degree of acidity of the oil was determined to be 5.5 (Table 1) for MKO and free fatty acid value of 2.75 % which reduced to less than 1 % free fatty acid through esterification to enhance the yield of MKME. High value of TAN undermined the quality of the oil, as this could cause corrosion and wear on the metals of engines. The iodine value which is the measure of degree of unsaturation of the MKO decreases from 150.62 to 125.5 after formation of MKME. Saponification value for MKO was high as evidence in its high value (222.5) and this explains why it saponified into soap during transesterification when NaOH was used. The fatty acid composition from the GC-MS analysis of MKO revealed linoleic acid to be the most abundant in the oil with 40.85 percent, followed by oleic acid 25.60 percent, palmitic acid 21.78 percent and stearic acid 3.92 percent (Table 2).

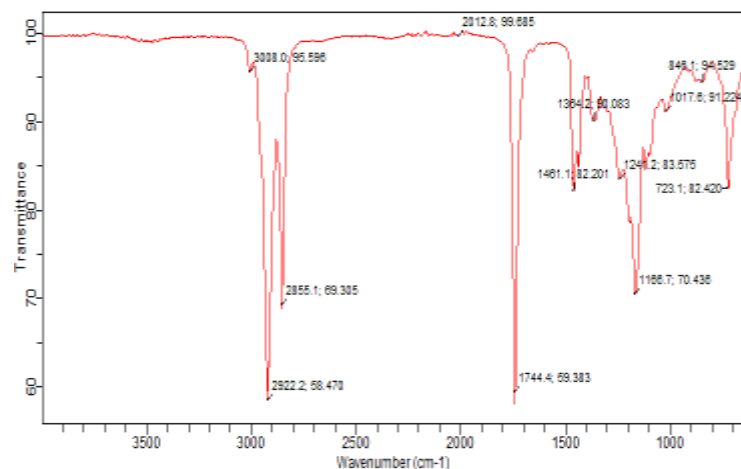


Fig 4. FTIR for mango kernel methyl ester.

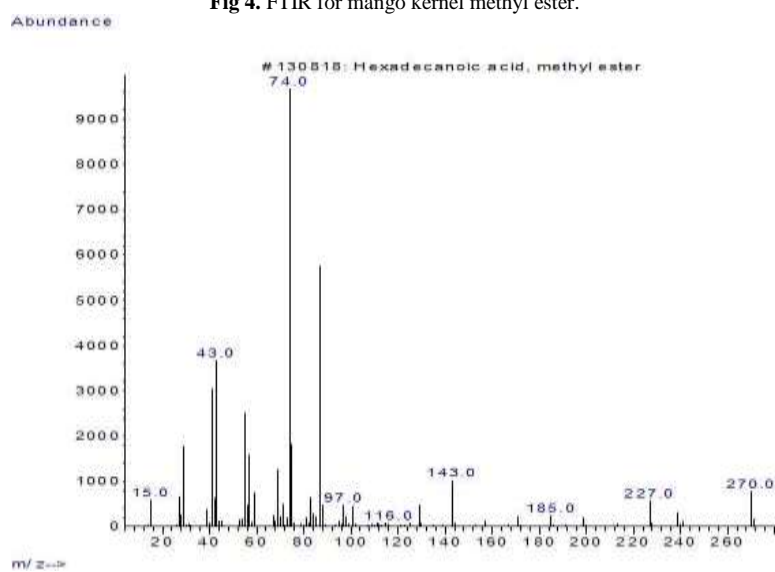


Figure 5a Hexadecanoic acid methyl ester (Instrument Library)

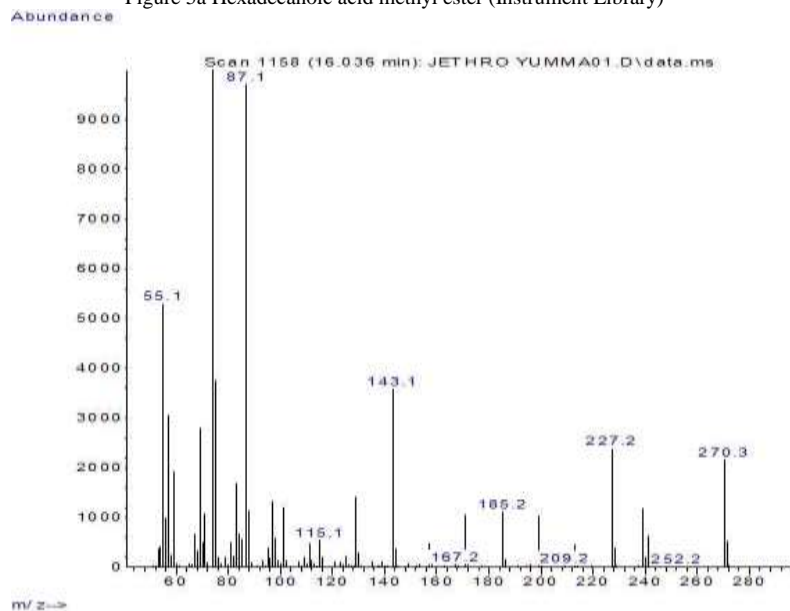


Figure 5. Showing spectra of Hexadecanoic acid methyl ester

These important fatty acids make it an essential oil for chemical and pharmaceutical applications. Dhinga and Kapoor 1985 reported in their journal “nutritive value of mango seed kernel” that oleic and stearic acid were the predominate fatty acid present in their Chausa and Dusheri Mango with 42 percent and 39 percent respectively. The fatty acid methyl esters presented in Table 3 for MKME also showed that 9, 12-octadecadienoic acid methyl ester was abundant with 47.83 % (Fig.6) followed by hexadecanoic acid methyl ester with 17.85 % (Fig.5) and methyl stearate with 17.75 % (Fig.7). Methyl 18-Methylnonadecanoate and Tetracosanoic acid methyl ester with relatively low abundance were captured in Figure 8 and 9 respectively. The FTIR analysis of the MKO and MKME (Figure 3 & 4) was characterized by some notable bands. The sp^3 hybridized C-H stretching appears around 2855 cm^{-1} and 2922 cm^{-1} , the sp^2 band of unsaturation (C-H) stretch also appears around 3000 cm^{-1} , while the strong band around 1744 cm^{-1} , signifies C=O stretch and C-O stretching around 1162 cm^{-1} . The weak band of unsaturation around 3000 cm^{-1} is further confirmed by the C=C weak band at 1528 cm^{-1} likely to be that of an aromatic ring since presence of m-cresyl acetate and 3-methylphenyl ester shown by GC-MS.

GC-MS results for some spectrum of MKME: The GC-MS analysis for the MKME reveal the presence of thirty-two (32) compounds ranging from methyl esters, butyl ester, steroids, dihydric alcohols, naphthalene, and phenolic compounds. The wide range of compounds present in mango kernel oil including phytochemicals such as phytosterols, tocopherols, Carotenoids and Polyphenols are what gave the MKO its therapeutic properties. Mwaurah *et al.*, 2020 stressed that these phytochemicals

possess high antimicrobial, antioxidant, antiproliferation and anticancer properties and as such could be processed for pharmaceutical applications, cosmetics and food.

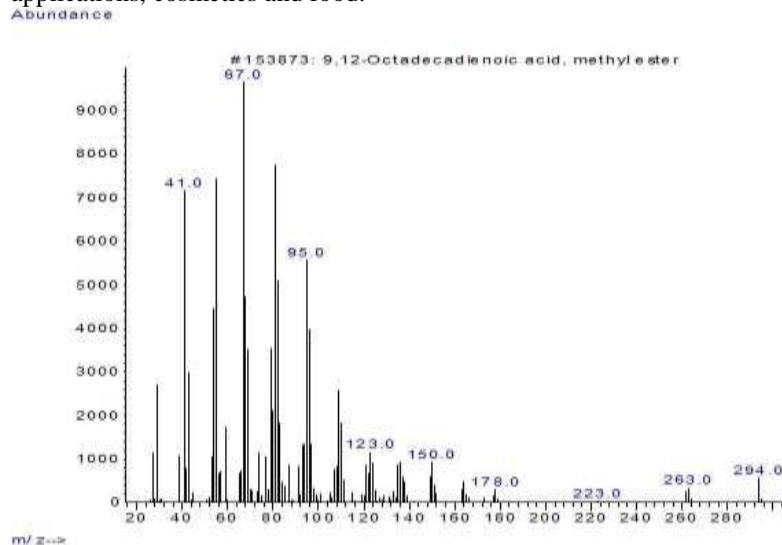


Figure 6a. 9, 12-Octadecadienoic acid methyl ester (Instrument Library)

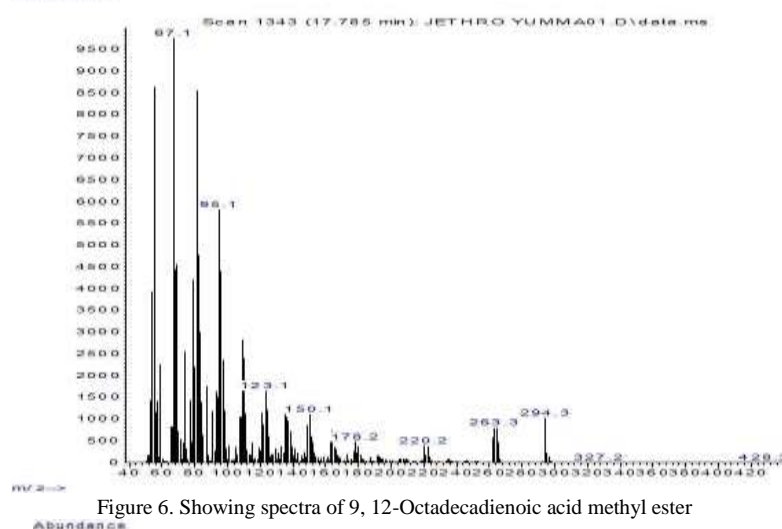


Figure 6. Showing spectra of 9, 12-Octadecadienoic acid methyl ester

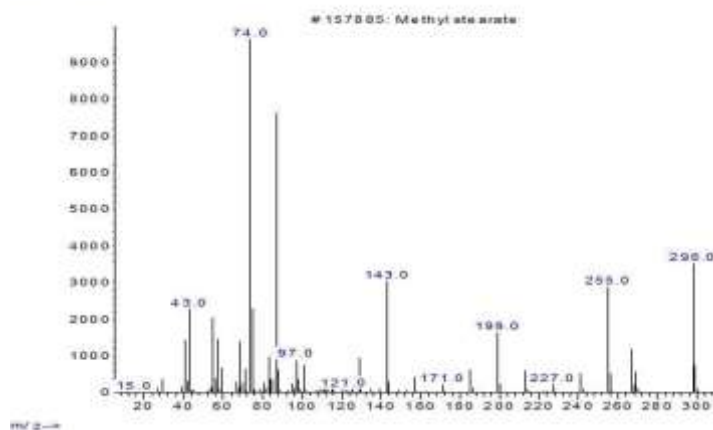


Figure 7a. Methyl stearate (Instrument Library)

The GC-MS result (Table 3) shows varying amount of both saturated and unsaturated fatty acid methyl esters. However, Linoleic acid methyl ester was still the most abundant with relative percentage of 47.84, followed by Palmitic acid methyl ester with 17.86 percent and Stearic acid methyl ester with 17.75 percent. Considering these three major constituent of MKME the ratio of unsaturated to saturated fatty acid is about 47:35, the report of MKME having relatively high percentage composition in Oleic and Stearic acid methyl ester abound in literature (Chakraborty & Das, 2017; Dhinga and Kapoor, 1985 and Mwaurah *et al.*, 2020) in contrast to the mango kernel used in this work which agrees with what was reported by (Diarra, 2014 and Lieb *et al.*, 2019). The variation could be as a result of mango variety and the method of extraction (Al-Saadi, 2017). The Palmitic and Stearic acid, methyl ester are saturated and could account for the relative oxidative and thermal stability possessed by MKME, although polyunsaturated fatty acid methyl esters are prone to auto-oxidation but MKME is more stable relative to other polyunsaturated oils such as mustard (94.27 %) and sunflower (88.39 %) (Dorni *et al.*, 2018 and Ramadan, 2019). The synthesis of MOTE was monitored by FTIR in which 0.5 cm³ was taken out at two separate intervals of 75 mins each with help of 2 cm³ syringe and analyzed for completion of reaction, the two products in each case was Mango oil based trimethylolpropane ester 1 (MOTE1) and Mango oil based trimethylolpropane ester 2 (MOTE2). The third was the acetylation of MOTE2 to produce the Mango oil based trimethylolpropane ester 3 (MOTE3). The presence of broad OH band at the TMP spectra (Figure 10) which disappears as the reaction progressed was used to monitor reaction completion.

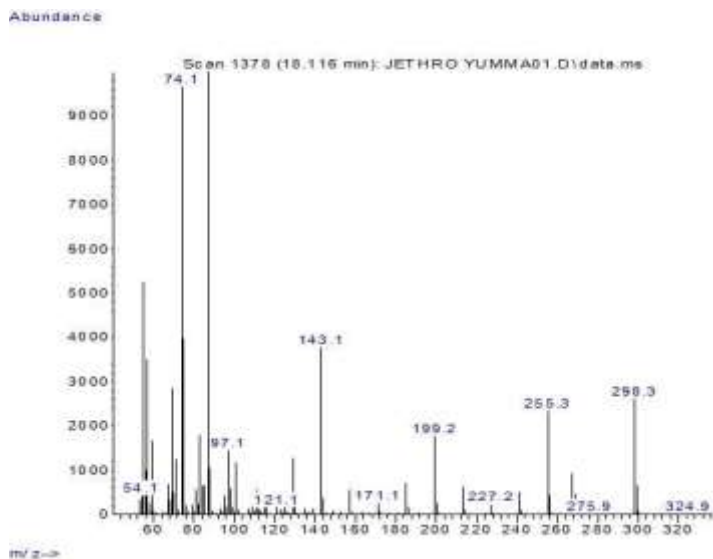


Fig 7. Showing spectra of Methyl stearate.

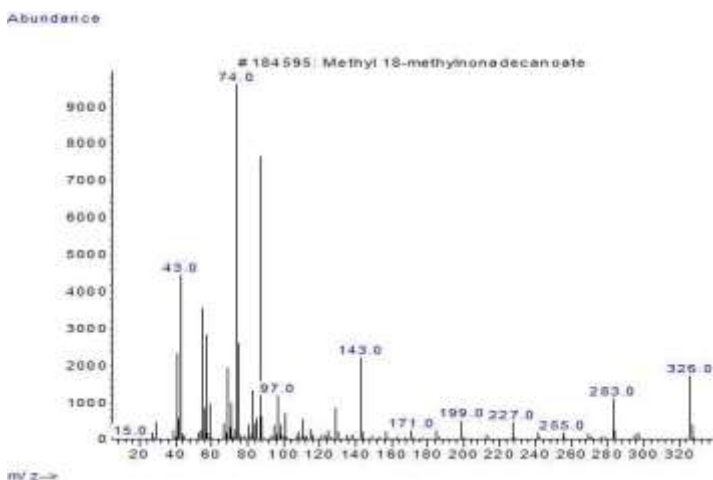


Figure 8a. Methyl 18-Methylnonadecanoate (Instrument Library)

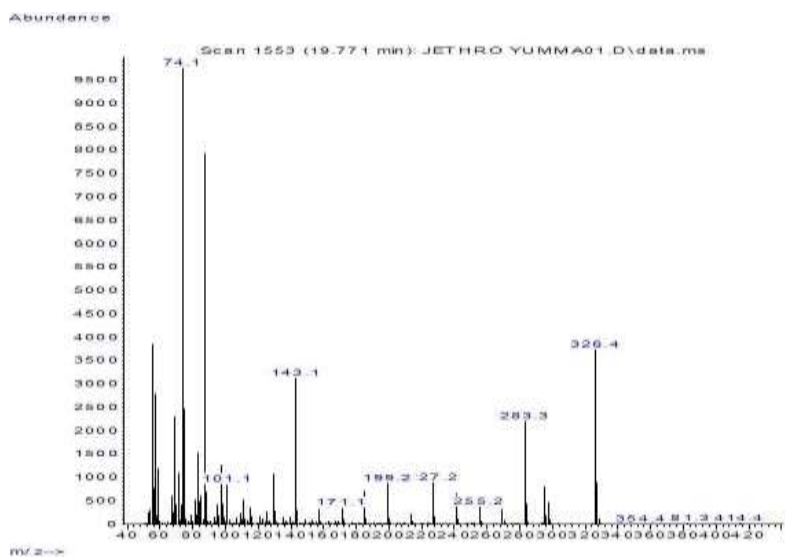


Figure 8. Showing spectra of Methyl 18-Methylnonadecanoate

The TMP spectra with broad O-H absorption at about 3,300 cm⁻¹ which is the functionality along with its branched network that made it suitable for transesterification with fatty acid methyl ester in order to improve upon its hydrolytic and thermal properties a phenomenon associated with natural oils (Erhan *et al.*, 2006; Menezes *et al.*, 2012 & Saboya *et al.*, 2017).

The reaction of the fatty acid methyl ester with TMP progressed with decrease in O-H band (MOTE1) as seen in (Fig. 10), at MOTE2 the reaction has been nearly completed. Acetylation of the final product completely removed any trace of O-H in the biolubricant produced as can be seen in the spectrum for MOTE3. The FTIR analysis of the MEB-L synthesis via epoxidation route present only little significant variation from the spectra obtain from MKME precursor. There was no trace of unwanted functional group in the final product MEB-L2 as seen in (Figure 11).

Analysis of Tribometer Results: The coefficient of friction (COF) with symbol (μ) represents a measure of the amount of friction existing between two surfaces, it is the ratio between frictional force and normal force. Materials with COF less than 0.1 are considered lubricous materials. The COF for MOTE and MEB-L reveal 0.083 μ and 0.08 μ (Figure 12 & 13) respectively pointing to the fact that MEB-L has superior lubricity properties than MOTE. The lower COF value of MEB-L shows its tribological effectiveness Arumugam *et al.*, 2020, however the standard deviation for both MOTE and MEB-L stand at 0.008 and 0.013 respectively meaning MOTE is relatively more stable than MEB-L. The presence of longer side chain ester in MOTE resulted to the relative stability of MOTE to MEB-L.

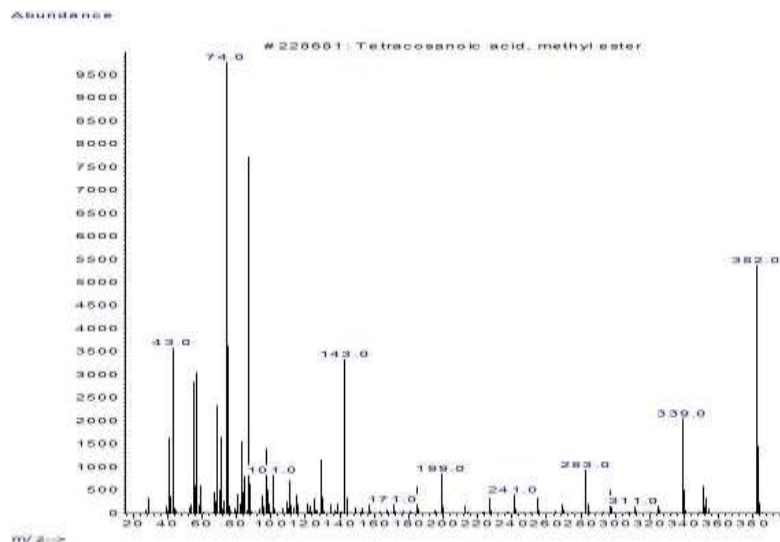


Figure 9a. Tetracosanoic acid, methyl ester (Instrument Library)

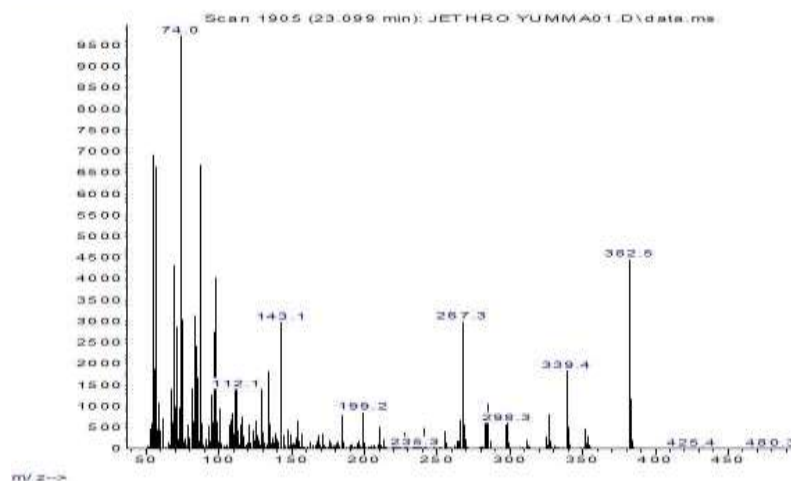


Figure 9. Showing spectra of Tetracosanoic acid, methyl ester

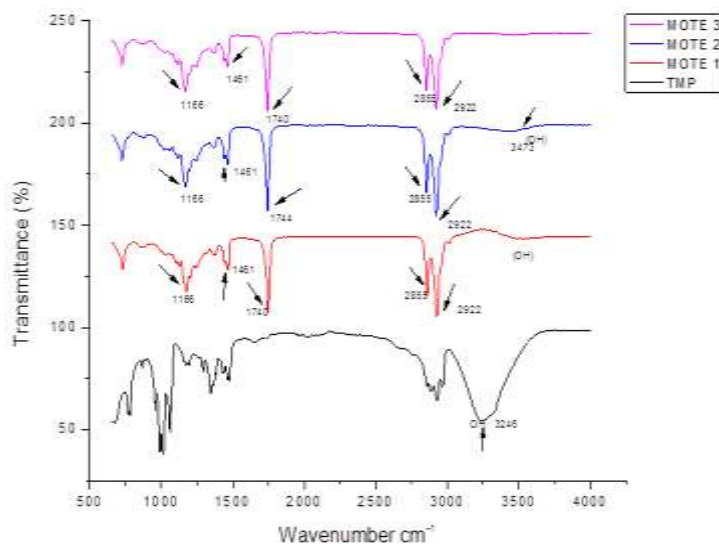


Fig 10. FTIR showing progress of synthesis of MOTE

However, the coefficient of friction reported in this work for MOTE and MEB-L are better than those reported for Palm biodiesel and Diesel fuel (Mofijur *et al.*, 2017).

Conclusion: The physicochemical parameters of Mango (*Mangifera indica* L.) kernel oil look good for a typical biolubricant formulation (density, viscosity), the fatty acid composition which is rich in linoleic, oleic and palmitic acids presents its use for cosmetic and pharmaceutical industries. MOTE and MEB-L are lubricious because their coefficient of friction is below established standard. However, thermal and oxidative studies as well as the low temperature fluidity is recommended for MOTE and MEB-L.

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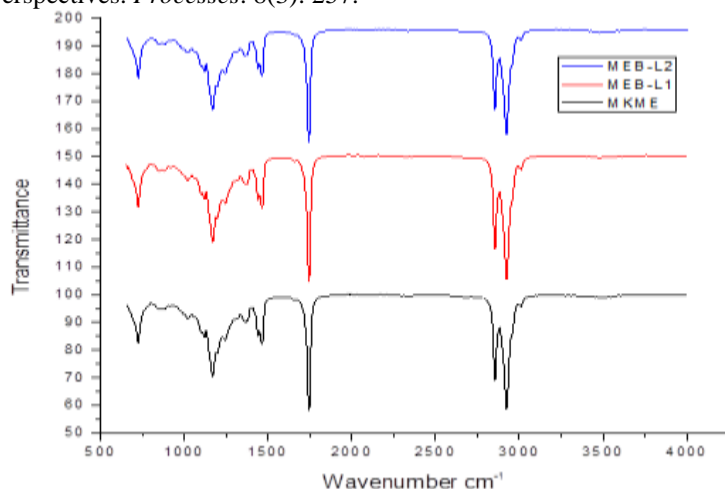


Fig 11. Showing the progress of synthesis MEB-L

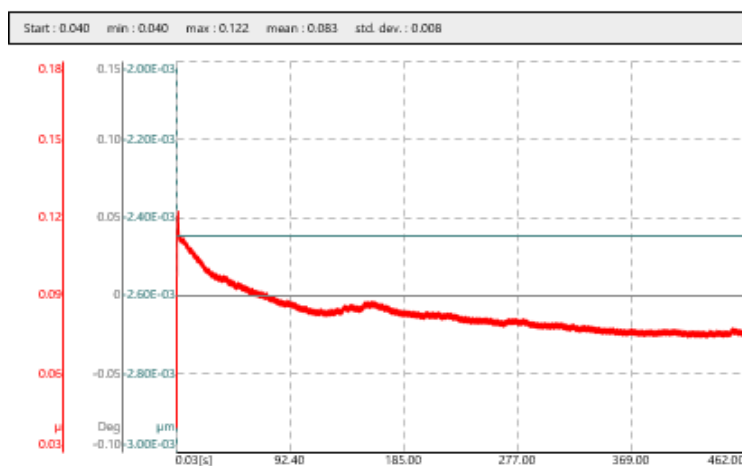


Fig 12. Tribological result of MOTE



Fig 13. Tribological result of MEB-L

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