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# Extraction and characterisation of the oil from *Moringa oleifera* seeds

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**Abstract**
An extract of *M.oleifera* oil was converted to biodiesel. The physicochemical properties of both oil and biodiesel were characterized using standard methods.
The result show that of the oil yield was 38%, Moisture was (0.22%), Density (0.9022 g/cm$^3$), Kinematic Viscosity at 40C (35.75 cp), PH (5.42), Fat content (38.3%), Acid value (9.89%), saponification value (210.13 mgKOH/g), Iodin value (66.50 g $I_2$/100 g), Peroxide value (6.50 Meq/kg), Refractive index (1.4791), Free fatty acid (4.22%), Flash point (185%).
FT-IR and GC-MS show successful conversion of oil into biodiesel.

**Keywords:** biodiesel, extraction, *M.oleifera*

## Introduction

Recently researcher was focused on *M.oleifera* plant due to its wide medicinal application, water-treatment and energy sector, etc. (Gupta *et al*, 2018) [1]. The native place of the plant is North-Western India, also tropical areas around the world (Pandey *et al*, 2011) [2], in Sudan the tree known as Rawag tree due to capability to purify turbid water specially in autumn season (Jahn S.A *et al*, 1986) [3]. The *M.oleifera* is rapidly growing tree even in poor soil and is little affected by drought. The leaves of *M.oleifera* is edible it used as food additive and rich source of potassium, calcium, vitamin A (Jahn, 1977) [4], (Kuete, 2017) [5].
Furthermore, *M.oleifera* tree grow to approximately 10 M in height, it often cut back annually 1-2 M to regrow. Renewable energy production is one of the main targets for countries policy measures to mitigate climate change and fuel price fluctuations. Among the energy produced from renewable resources (e.g., solar, wind), energy from biomass residues and vegetable oils in the form of solid fuels (e.g., briquettes, charcoal), gas fuel (e.g., biogas) and liquid fuels (e.g., bioethanol, biodiesel) could fulfill the local energy demand, generate economic development through agriculture activities and promote rural employment (Panwar *et al*, 2011) [6]. Moreover, *M.oleifera* oil present suitable viscosity and acidity index to be used as biodiesel feedstock (Mirhashemi *et al*, 2018) [7].
The term biodiesel defined as fatty acid alkyl esters of vegetable and animal oils; however, biodiesel have many properties such as friendly to environment, renewable, non-toxic, high flash point and low-cost price due to availability, abundant and high efficiency vegetable oil were used as main source of biodiesel production, however, many studies used sun flower, soybean and palm oil for biodiesel production, those oils competes with food chain series. Although, researchers focus on alternative feed stock like common and conventional oils include tobacco, rubber seed and moringa oils for biodiesel production. (Rashid *et al*, 2008) [8]. *M.oleifera* seed contains between 33-41% w/w oil. (Sengupta *et al*, 1970) [9]. there are three techniques to extract oil from the seeds: chemical Soxhlet extraction, cold chemical extraction, and mechanical pressing.

## Materials and Methods

### 1. Materials

The samples were supplied by Sudan University farms forestry collage. All chemical used were from Sigma Aldrich and SDFCL.

### 2. Methods

#### 2.1 Oil Extraction

*M.oleifera* seeds were dried in the oven for 12 hours. In the Soxhlet process, the solvent was evaporated; 1000 g from the seed samples by continuously washing the solid with volatile solvent such as (N-hexane) in Soxhlet extraction apparatus, heated to 60 °C for 8 hours.
The extracted oil and solvent were transferred to a rotatory evaporator to remove the n-Hexane (Hassan SA *et al*, 2016) [10]. In the Soxhlet round bottom flask, 1000 g of the seeds were added to 2000 ml of n-Hexane, heated at 60 ° C for 8 hours. The oil and solvent extracted was moved to a rotatory evaporator to remove N-Hexane (Dutta R*et al*, 2015) [11].
Eq.1 was used to calculate the oil yield from all methods of oil extraction.

$$Oil\ yield = (Weight\ of\ oil)/(Weight\ of\ seeds)\ \times\ 100 \quad (1)$$

#### 2.2 Fatty Acid Profile of *M.Oleifera* Seed Oil

0.3g of crude oil sample were weighted into a 250 ml round bottom flask, 6 ml of 0.5 M methanolic NaOH were add to the sample and boiled for 2.5 min. 1% Sulfuric acid, and methanol, was added to the mixture, shaken, and kept overnight at 50°C. 2 ml of hexane was added and well steered. 1 ml of the upper Hexane layer was transferred to a glass stoppered tube; anhydrous $Na_2SO_4$ was added to Hexane layer and kept in vial. The dried sample was subjected to GC analysis.

#### 2.3 Transesterification Reaction

500 ml of acid treated oil were placed into a 1 L beaker and heated up to 60 °C, 100 ml of fresh methanolic Sodium Hydroxide were added under stirring at 3000 rpm for two hours. The mixture was transferred to a separating funnel

and kept for 24 hours. Then the lower, glycerol layer was drained, and the upper biodiesel layer was washed, three times, with warm distilled water to remove soap, methanol, and remaining glycerol (Okullo A *et al*, 2006) [12].

### 2.4 Physicochemical properties of *M.Oleifera* oil and Biodiesel

#### 2.4.1 Determination of Chemical parameters

Acidity, iodine number and saponification values were determined according to the MPOB (Malaysian Palm Oil Board) methods (Kuntom *et al*., 2005) [13].

#### 2.4.1.2 Acid value of oil

Standard solution of alcoholic potassium hydroxide (0.1N) was prepared and standardized by using standard solution of potassium hydrogen phthalate (0.1N). Phenolphthalein (1g) was dissolved in 95% ethanol to prepare (1%w/v) solution and used as indictor. The sample (2 – 3g) was dissolved in a 50 ml mixture of ethanol and diethyl ether (1:1) solution. The solvent mixture was notarized the 0.01 N KOH. The sample was titrated against the standard solution of the alcoholic potassium hydroxide (0.1N) to the end point of the phenolphthalein indictor.

Eq.2 Acidity (mg KOH/g) was expressed as free fatty acids percentage (FFAs %) as oleic according to the following equation:

$$\text{FFAs} = \frac{8.2 \times N \times V}{M} \quad (2)$$

Where, N is the exact normality of the standard alcoholic KOH, V is the volume of the standard KOH to neutralize the sample.

Acid value (mg KOH/g) was obtained by multiplying FFAs% by the factor of 1.99 (Kuntom *et al*., 2005) [13].

#### 2.4.1.3 Iodine value of oil

O.4 g of crude oil was weighed in a conical flask (500ml) and dissolved in 15 ml mixture of cyclohexane and acetic acid (1:1). Solution (25 ml) was dispended in the flask. The flask was closed, swirled to intimate mixture, and kept in the dark for 30 min. At that, 20 ml of 10% potassium iodide (KI) were added followed by addition of 100 ml distill water. The mixture was then titrated against standard solution of Na2 S2O3.5H2O (0.1N) until very faint yellow iodide color persisted. At that point, few drops of starch were added, and the titration was continued until the blue color disappeared. The procedure was repeated for the blank.

Eq.3 Iodine value was calculated by the following formula:

$$\text{Iodine value} = (B - S)\ N\ 12.69 / M \quad (3)$$

Where, B is the titrant volume of the blank, S is the titrant volume of the sample, N the normality of the potassium thioslfate penta hydrate and M the weight of the sample.

#### 2.4.1.4 Saponification value of oil

2.0 grams of the crude oil were weighed in round bottom flask (500 ml) and dissolved in a 50 ml of ethanolic potassium hydroxide solution (0.5N). The mixture was heated under reflux for 30 min and cooled to room temperature. Excess potassium hydroxide was titrated against standard 0.5N hydrochloric acid solution using phenolphthalein as an indicator. A blank determination was carried out simultaneously under the same set of experimental condition. The standard solution of hydrochloric acid was standardized by using standard sodium bicarbonate solution 0.1N.

Eq.4 The saponification value was determined as follow:

$$\text{Saponification value (mg KOH/g)} = (B - S)\ 56.11 / M \quad (4)$$

Where, B theis titrant volume of the blank, S is the titrant volume of the sample, N the normality of the hydrochloric acid and M the weight of the sample.

#### 2.4.2 Determination of Physical parameters

Some physical properties Such as kinematic viscosity, density, cloud points were determined in accordance with the ASTM D6751 standard. Automatic SVM 3000 Stabinger Viscometer (Antoon Par, UK) was used to determine viscosity (D445) and density (D1298) simultaneously at 40 C. A fully automated cloud point (CP) tester (model NTE 450. REF 60300 (Normalab, France) was used to determine CP (D2500).

### Results and Discussions

Table 1 shows the percentages yields of *M.Oleifera* seed oil was 38% w/w. Which is to that reported by (Anwar *et al*, 2005) [14].

**Table 1:** Oil yield from *M.Oleifera* Seeds Using Soxhlet extraction method.

| No. | Extraction method | Oil yield, wt% |
|---|---|---|
| 1 | Soxhlet solvent Extraction | 38.0 |

Table 2 shows the physicochemical properties of *M.Oleifera* oil with high Kinematic Viscosity 35.75 $mm^2$ /s, while low amount of Acid Value 9.98%.

**Table 2:** Physical and Chemical properties of *M.Oleifera* oil

| Parameter | Value of raw oil |
|---|---|
| Moisture content % | 0.22 |
| pH | 5.42 |
| Kinematic Viscosity at 40 °C | 35.75 |
| Refractive Index | 1.4791 |
| Fat Content % | 38.3 |
| Density at 40 °C | 0.9022 |
| Flash point | 158.0 |
| Acid Value % | 9.89 |
| Free Fatty Acid % | 9.22 |
| Peroxide Value meg/Kg | 6.00 |
| Saponification Value mg KOH/g oil | 210.13 |
| Iodine Value $I_2$ /100g oil | 66.5 |

Table 3 shows to analysis of *M.Oleifera* methyl ester, the result showed that there are 21 compounds in the sample, however, the identified chemical compounds contain 82.2% probability and 65% area in methyl ester sample, while fatty acid determined in methyl ester sample.

**Table 3:** Table 3 and Fig 1 display the fatty acids composition of *M.Oleifera* crude oil.

| No | Fatty Acid | Area % |
|---|---|---|
| 1 | Hexadecanoic acid methyl | 22.300 |
| 2 | Docosanoic acid methyl ester | 19.180 |
| 3 | 10-Octadecenoic acid methyl ester | 17.230 |
| 4 | Octadecanoic acid methyl ester | 15.090 |
| 5 | Eicosanoic acid methyl ester | 14.590 |
| 6 | Tetracosanoic acid methyl ester | 3.880 |
| 7 | 9-Octadecenoic acid (Z) methyl ester | 3.490 |
| 8 | 9-Hexadecenoic acid methyl ester | 1.880 |
| 9 | Octadecanoic acid methyl ester | 0.560 |
| 10 | Heptadecanoic acid methyl ester | 0.340 |
| 11 | 9-Octadecenal | 0.290 |
| 12 | Tricosanoic acid methyl ester | 0.230 |
| 13 | Ricinoleic acid methyl ester | 0.230 |
| 14 | Triacontanoic acid methyl ester | 0.190 |
| 15 | 13-Docosenoic acid methyl ester | 0.170 |
| 16 | Di-(Octadecenoyl)-Glycerol | 0.090 |
| 17 | Cyclopropaneoctanoic acid | 0.080 |
| 18 | 2-Methylene-1,5-pentanediol | 0.070 |
| 19 | (E)-9-Octadecenoic acid methyl ester | 0.040 |
| 20 | 10-Nonadecenoic acidmethyl ester | 0.040 |
| 21 | Stigmasterol | 0.030 |
| | Total | 100 |

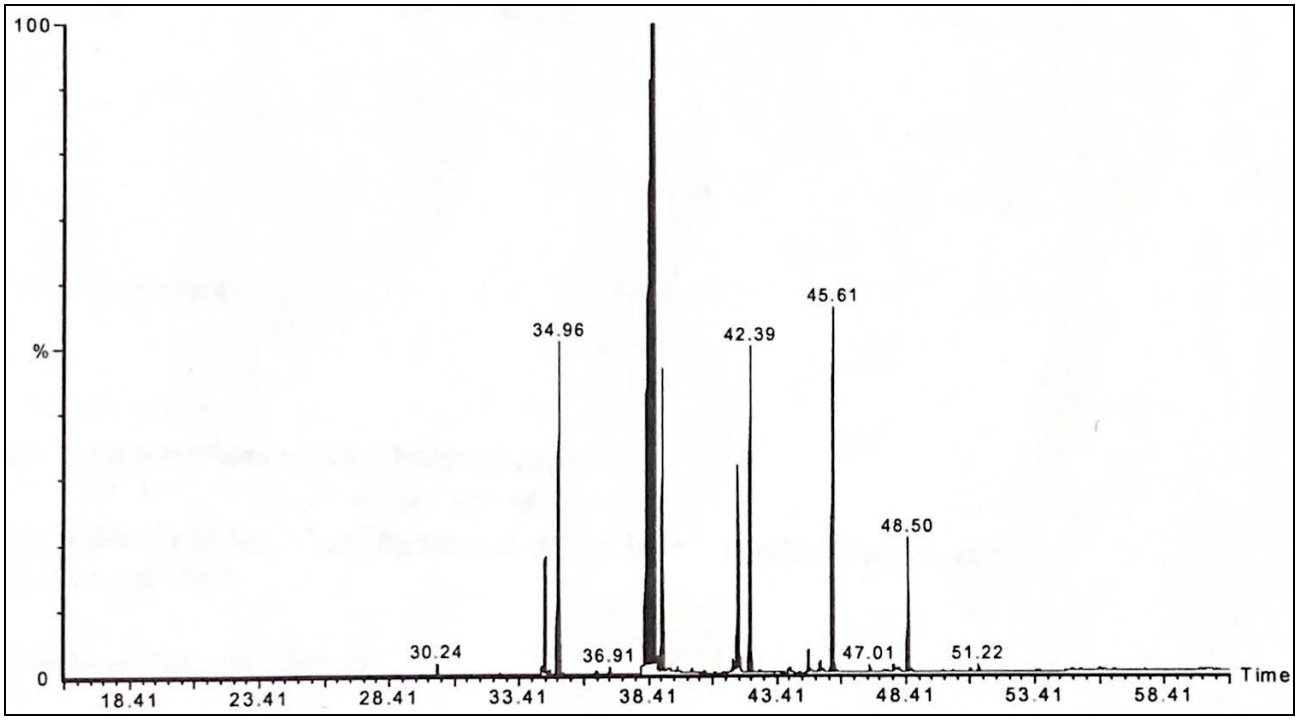


**Fig 1:** GC-MS Spectrum of *M.Oleifera* seed oil.

Fig 2 shows the FTIR spectrum of *M.Oleifera* methyl ester, the range of spectrum start 4000 and end in 355 $cm^{-1}$ to determine main functional groups. Hydroxide group normally showed in wavelength 300 to 3500 $cm^{-1}$ in this graph exanimated in 3002 $cm^{-1}$ with stretch of vibration, while alkane group C-H absorbed in range 2852 to 2923 $cm^{-1}$. The wavelength around 1745$cm^{-1}$ sign to carbonyl group.

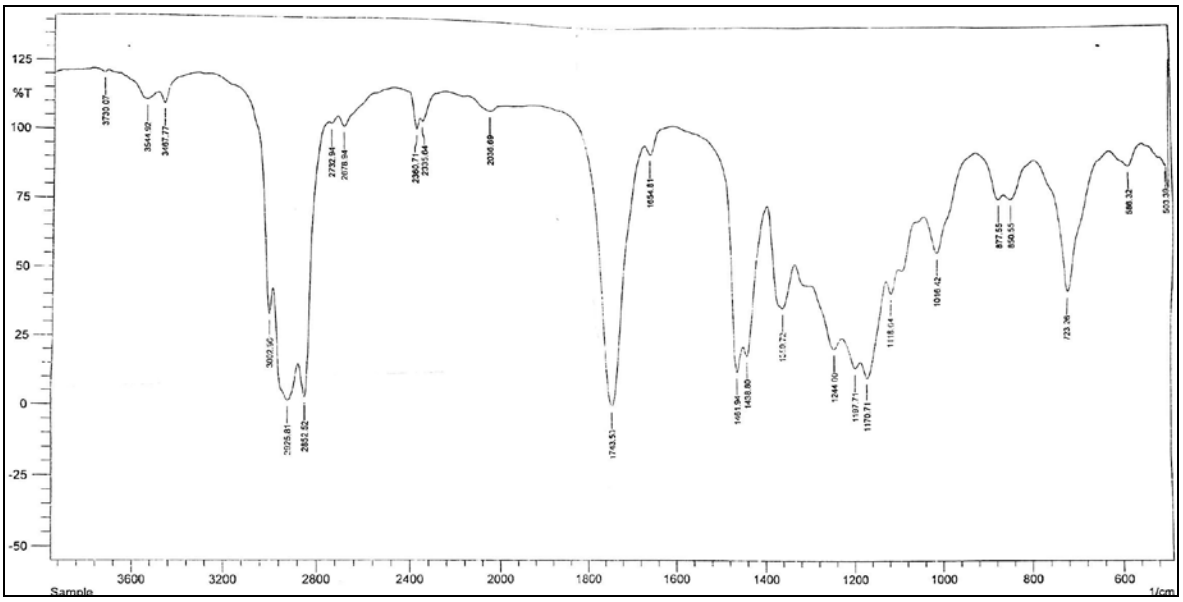


**Fig 2:** FTIR Spectrum of *M.Oleifera* methyl ester.

## Conclusions

- High yield of oil was gained from Chemical extraction using Soxhlet.
- *M.Oleifera* oil contains 82.2 % of saturated fatty acids and 17.8% of unsaturated fatty acid.
- *M.Oleifera* bio diesel was, successfully, synthesized using Alkali transesterification and proven using FTIR.
- The Physical and Chemical properties of *M.Oleifera* oil show high Kinematic Viscosity and low amount of Acid Value and meets of both ASTM D6751 and EN14214 standards.

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