Original Research Article

Journal of Biological Research

& Biotechnology

Bio-Research Vol. 20 No.3; pp. 1712-1720 (2022). ISSN (print):1596-7409; eISSN (online):2705-3822

Metal Analysis and Fuel Potentials of *Irvingia gabonensis* Seed

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Abstract

Irvingia gabonensis seeds yield a large amount of oil which can be evaluated for its fuel potential. In this study, *Irvingia gabonensis* seeds were analysed for their chemical composition. The analysis was carried out on the seeds using standard methods. Selected metals were determined in the seeds by dry ashing method using atomic absorption spectroscopy. Oil was extracted with n-hexane by sonication. Biodiesel (methyl esters) was produced from the oil by transesterification. The relative abundance of some fatty acid methyl esters were determined by gas chromatography-mass spectroscopy. The fuel properties of the oil methyl ester (biodiesel) were tested using standard methods. The results showed that the average moisture content was 3.21%, ash level was 1.73%, crude fat was 52.67%, while crude fibre, crude protein and carbohydrate contents were 2.63, 7.31 and 10.15% respectively. The seeds contained Na, K, Ca, Mg, Pb, Zn, Cu, and Cd which were in various amounts. Linoleic acid and methyl ester had the highest abundance at 48% respectively while oleic acid was of least abundance at 1.2%. The fuel properties showed that the properties of biodiesel from *Irvingia gabonensis* were within the acceptable quality standards for application in diesel engines.

Keywords: *Irvingia gabonensis*, biodiesel, concentration, proximate analysis, Atomic absorption spectroscopy, metals, transesterification

Received July 26,2022; **Revised** October 10, 2022; **Accepted** October 29, 2022

INTRODUCTION

Irvingia gabonensis commonly called African/bush/wild mango is a deciduous tree, grown in West and Central African countries (Mgbemena *et al*., 2019). The fruits have greenish mesocarp when unripe, and yellowish or light orange when ripe (Etebu and Tungbulu, 2015). The seeds have been the subject of research due to claims of their health benefits and nutritive content. Ngondi *et al*. (2005) opined that the seed is capable of reducing fasting blood glucose levels. Oben (2011) suggested that the seed extracts of *Irvingia gabonnsis* can be applied to manage metabolic syndrome with oxidative stress of different components and to significantly reduce the plasma levels of the inflammatory marker in overweight and obese patients. A research result revealed that ethanolic extract of *Irvingia gabonensis* at doses studied caused a reduction in the body weight in mice (Adesanya *et al.,* 2019).

Due to various claims on the health benefits, the trees are being exploited for economic reasons. Consequently, *Irvingia gabonensis is* in the list of endangered plant species (Koffi *et al*., 2018). Apart from the medicinal properties associated with *Irvingia gabonensis*, the oil content of the seed is known to be high, thereby making it useful for making various products like cosmetics and soap. It became imperative to produce biodiesel from seed oil and check its properties.

Because of these applications, searches were made on basis of the various assertions on *Irvingia gabonensis* . Literature search revealed scanty information on the analysis of *Irvingia gabonensis* seed for chemical composition. For example, Giami *et al* (1994) studied the chemical composition and functional properties of raw, heat-treated and partially proteolysed wild mango seed flour. Also, Ekpe *et al* (2007) studied the proximate composition and amino acid profile of bush mango seeds, while Ogunsina *et al* (2012) investigated the proximate composition of African bush mango kernels (*Irvingia gabonensis*) and characteristics of its oil. Therefore, this study investigated the chemical composition of *Irvingia gabonensis* The seed oil is assessed for its fuel potential for application in diesel engines.

MATERIALS AND METHODS

Sample collection and preparation of *Irvingia gabonensis* seed

Irvingia gabonensis fruits were picked up under the *Irvingia gabonensis* trees that were found at several locations in Nsukka town, Enugu State, Nigeria in 2020. The fruits were washed and identified by a botanist at the University of Nigeria, Nsukka, Nigeria. The mesocarp of the fruits and the seed coats were peeled off with a knife. The seeds were left to dry in the solar dryers at the National Center for Energy Research and Development, University of Nigeria, Nsukka, for two months. This was due to fluctuating temperatures at the time. The dried seeds from different fruits were ground and screened through a sieve of 250-micron pore size. The samples that passed through the sieve pores were mixed thoroughly and formed a composite mixture which was packaged in plastic bags. The labelled bags containing the samples were left in a cupboard which was dry and dark, to prevent degradation of the samples, before they will be used for pending analysis.

This study was carried out at the Biomass Laboratory, National Center for Energy Research and Development, University of Nigeria, Nsukka, Nigeria, between 2020 and 2021.

Chemical composition of *Irvingia gabonensis* seed

Proximate analysis

The ground seed samples were analysed for moisture, ash, carbohydrate, protein and fat contents following AOAC methods (Mansouri *et al*., 2018). For the moisture determination, concisely, the ground and sieved samples (0.500 g) were weighed out and placed in the sample compartment of a moisture analyser, MB 35 Halogen moisture analyser. The equipment was operated according to the manufacturer's instructions and set at 110 °C. The results were recorded.Ash content determination involved using a muffle furnace (Vecstar, model LF3, Chesterfield, United Kingdom) in which the crucibles containing the sieved samples were after weighing. The furnace was set at 400 °C for 30 minutes, then at 850 °C for 1 h 45 minutes. After, the covered crucibles with the ash when cooled were brought out from the furnace and reweighed and then gravimetrically estimated. Crude protein was determined using the Kjeldahl method (Mæhre *et al*., 2018). Crude fat was determined by extraction method using a sonicator (Model SALD-BS2 Beaker type, Shimadzu Corporation). In this process, two grams of the dried sample were placed in the sonicator bath of the sonicator. The stirrer and the ultrasonic pulse were activated and the extraction was allowed to proceed for 25 minutes. Then the extracts were decanted. After the extraction by sonication, the extracts were placed in tubes at the same level and placed in different compartments in the centrifuge. The centrifuge was set at 3500 rpm for 30 minutes. The extract was thereafter decanted and filtered.

Determination of Selected Metals in *Irvingia gabonensis* Seed

The ash from the ash content determination was dissolved in concentrated HCL. The solution was decanted into the standard flask and made up to mark with distilled water.

The concentrations of Fe, Zn, Cd, Cu, Fe, and Pb in the sample solution were analysed with a Flame atomic absorption spectrophotometer (FAAS) (Model AA-6800, Shimadzu Corporation, Japan) using an air-acetylene flame which has a digital read-out system. The equipment was calibrated with standard solutions for the metals to be analysed. Each metal was determined using its lamp. The concentrations of the metals were obtained after subtracting from the blank solution. The analysis were done in duplicate as a quality assurance measure. A recovery experiment was carried out as outlined by Ugwu and Ofomatah (2021).

Determination of Sodium and Potassium by Flame Photometry

A flame photometer, the Gallenkamp flame photometer, was used for the determination of Na and K in the sample. The sample solution was prepared from the sample ash. The photometer was operated following the instructions of the equipment manufacturer. Calibration of the equipment was done with the standard solutions of the metals being determined. The meter of the instrument was set at 100% E (Emission) while the concentration of the standards is aspirated. The %T of all the intermediate standard solutions was recorded. The blank was used to reset the equipment. Each of the sample solutions was aspirated and the readings (T) recorded. The plot of the standard curve was made on linear graph paper. The concentration of each element in the sample solution is read from the standard curve which was obtained from the standard solution.

Determination of Calcium and Magnesium by Titrimetric Titration

Exactly 10 mL of sample solution obtained from the dry ashing digestion were pipetted into a separate 350 mL conical flask. About 25 mL of NH_3-NH_4Cl buffer solution was added. 25 mL of water and 2 drops of Erichrome Black-T indicator were added. The solution was titrated against 0.01 M EDTA solution until a very light blue colour was obtained as the endpoint. The volume of EDTA used is the volume equivalent of calcium and magnesium in the admixture. All the determinations were done in triplicate.

Transesterification of the seed oil of *Irvingia gabonensis*

The free fatty acid content in the oil was determined by the titrimetric method. The seed oil of *Irvingia gabonensis* was extracted by sonication using a sonicator. The transesterification process was used to produce the biodiesel following the method of Sokoto *et al*. (2018). The seed oil was pretreated with methanol using an H_2S0_4 acid catalyst to convert free fatty acid to ester. The reaction was conducted at a temperature of 65 °C for 60 minutes. After esterification, the alcohol layer was removed from the preheated oil before transesterification, in which methanolic sodium hydroxide was poured into a flat bottom flask containing the esterified oil. The mixture was refluxed at a constant stirring speed and was transferred into a separating funnel. The mixture was allowed to separate overnight due to the influence of gravity. The dark bottom layer (glycerol) was drained out and the light upper layer (biodiesel) was recovered and washed with a 20% volume of warm distilled water. The mixture was gently agitated for 5 min and allowed to settle such that two layers were formed, and the biodiesel was separated into another dry container (Meher *et al.,* 2010).

Fuel properties of the biodiesel from *Irvingia gabonensis* seed oil

The fuel properties of the biodiesel produced from *Irvingia gabonensis* seed oil were determined using standard methods. These include specific gravity, viscosity and flash point (Ibeto *et al*., 2011). Viscosity was determined with an Oswald portable capillary viscometer. This investigation was done at the temperature of the laboratory (31ºC) by recording the time required for the biodiesel to pass between two marks in the viscometer (Ugwu and Eze, 2014).

Flashpoint was also determined for the biodiesel. It is used to assess the overall flammability hazard of a material. The flash point for the biodiesel produced in this study was measured by using a Pensky Martens semi-automatic multi-flash closed cup flash point tester (Made in Japan) ((Ugwu and Eze, 2014). The diesel index, cloud point, pour point, and cetane number were determined as described by Enweremadu *et al.* (2011).

GC–MS analysis of the biodiesel from *Irvingia gabonensis* seed oil

A gas chromatography/mass selective detector (GC/MSD) (GC model: 7890A; MSD model: 5975, Agilent Technologies, USA) in Selected Ion Monitoring (SIM) mode was used for the characterization of the fatty acid methyl esters in the biodiesel based on their boiling points and polarity. The analytes were separated in the capillary column of the machines. This was done after the injection of the derivatized sample into the equipment. A helium carrier gas was used for the analysis, and the oven program was initially at 65 °C for 1 min, up to 290 °C, for 11 min. The run time was 30 min, and at splitless injection, mode using an auto-sampler. The mass spectrometer quadrupole analyzer was used in electron ionization mode at 70 eV. Before the sample analysis, standards of fatty acids were analyzed with the equipment in SIM mode first to ascertain the fragmentation pattern. The target compounds and qualifier ions were determined after scanning the standard. The target compounds were identified by comparing the retention time of the compounds in the samples with the time of the standards used in the calibration of the equipment. The mass spectra of the target ions ratios were compared with the library database spectra of the National Institute of Standards and Technology (NIST). The concentrations were automatically read out from the instrument.

This study was conducted at the National Centre for Energy Research and Development, University of Nigeria, Nsukka.

RESULTS AND DISCUSSION

Proximate analysis is a determination of the moisture, ash, oil, fibre, protein and carbohydrate contents in a material. The mean proximate composition of *Irvingia gabonensis* seed, presented on Table 1, showed that the mean moisture content of *I. gabonensis* seed is 5.21%. This is relatively low and may be attributable to the drying in a solar dryer over a relatively long period due to fluctuations in weather. A matrice loses water as it stays longer in a dryer. It is advantageous for the moisture content to be low since it will reduce microbial growth and deterioration over a long time.

The mean results of proximate analysis of *Irvingia gabonensis* seeds are presented in Table 1

Table 1: Mean (%) results of proximate analysis of *Irvingia gabonensis* seeds

Parameters	%
Moisture content	5.21
Ash	2.73
Oil	57.97
Crude fibre	7.63
Crude protein	8.31
Carbohydrate	18.15

Ogunsina *et al.* (2012) reported a moisture content of 2.55%, while Mgbemena *et al.* (2019) recorded a moisture composition of 1.4%, and 5.20% was reported by Ekpe *et al.* (2007). The mean ash content in the present study was 2.73%. Ash content is an indication of mineral content. This indicated the low level of minerals in the seed. In comparison with other studies, ash content was 2.3% (Ogunsina *et al.,* 2012) 6.8% (Mgbemena *et al.*, 2019) and 9.50 (Ekpe *et al,* 2007). . At 57.97%, the oil content in the seed is very high. These implied that oil can be obtained easily from the seed of *I.gabonensis*. Other researchers reported oil content in the sample analysed as 68.39% (Ogunsina *et al.,* 2012) and 66.60% (Ekpe *et al,* 2007). The oil may be tested for application in domestic and industrial needs. With the mean % crude fibre at 7.63%, which is relatively low, the fibre level will barely help in bowel function during the consumption of the seed. The average crude protein content is 8.31% in the present study. This is close to 8.9% reported by Ogunsina *et al.* (2012) and 7.6% reported by Ekpe *et al.* (2007) and 5.6% recorded by Mgbemena *et al.* (2019). The protein level is reasonable for consumption, but I.*gabonensis* should be consumed with other foods for protein. The average level of carbohydrates in *I.gabonensis* is 18.15%. Ogunsina *et al.* (2012) reported a carbohydrate content of 18.67%. Carbohydrate level is an indication of energy content in food. This implied that *I.gabonensis* has a fair level of energy content.

The concentrations of some metals in *I.gabonensis* seed are shown in Table 2. Fe, Cu and Zn were essential elements required in small quantities by living organisms, including humans, to ensure good health (Briffa *et al*., 2020). These were found in the *I.gabonensis* samples. Iron is required for the normal functioning of the central nervous system and blood formation. Other elements found include Na, K, Ca and Mg. These were detected in various quantities as shown in Table 2. These elements all play vital roles in human health. While Na is an important component of sodium chloride, Ca and Mg are necessary for bone development. Magnesium activates many enzyme systems and maintains the electrical potential in nerves. The levels of Cd and Pb were low. However, Cd is a known heavy metal that can cause adverse effects on human health if present above a certain limit. Sodium, potassium and chlorine are important in the maintenance of osmotic balance between cells and the interstitial fluid (Soetan *et al.,* 2010). The results of the metal analysis in the present study were compared to the results from a similar study by Mgbemena *et al.* (2019) and presented in Table 2.

The relative abundance of various fatty acid methyl esters in the *Irvingia gabonensis* biodiesel presented in Table 3, shows varying compositions of the methyl esters. At 48%, linoleic acid methyl ester was most abundant. Next in abundance was myristic acid and then stearic acid at 30.54 and 10% respectively. These values show that *Irvingia gabonensis* oil is appropriate for use in biodiesel production.

Some of the fuel properties of biodiesel produced from *Irvingia gabonensis* seed oil were presented in Table 4. From Table 4, It is obvious that the fuel parameters were within the specifications of ASTM. A comparison of the cetane number of the *Irvingia gabonensis* biodiesel with biodiesel made from castor seed oil (68.55-71.16) (Auwal *et al*., 2022). indicates that *Irvingia gabonensis* biodiesel has a more favourable cetane number for use as biodiesel. Also, Neem oil biodiesel has Flash point, Pour point and Cloud point of 150 °C, 3 °C and 6 °C respectively (Aransiola *et al*., 2012), while this study has 101 °C, 4 °C and 1.9 °C for Flashpoint, Point and Cloud point respectively. This reinforces the suitability of *Irvingia gabonensis* biodiesel for fuel application in engines. The results of tests for fuel properties of the *Irvingia gabonensis* biodiesel are presented on Table 4. The variations in the composition of the *Irvingia gabonensis* samples from different locations are normal because of differences in varieties, types of soil where they were grown, and climatic changes (Ibeto *et al*., 2011). Also, the different sample preparation methods and processing techniques will result in differences in the final results obtained by different researchers.

This research is inconclusive at this stage. Therefore, the *Irvingia gabonensis* biodiesel cannot be used in engines. The biodiesel requires further purification, optimization of inputs and testing for certification before it may be confirmed for engine application. The results of the GCMS analysis of one of the samples showing relevant fatty acid methyl esters are presented graphically in Figure 1.

Table 2: Mean concentrations (in ppm) of selected metals in *Irvingia gabonensis* seeds

Metals	Fe	Zn	Na	K	Cd	Cu	Pb	Ca	Mg	References
Concen-trations	36.30 ±4.10	0.03 ±0.01	4.26 ±1.01	82.50 ±7.30	0.02	0.02	0.01	132.24 ±38.52	53.53 ±7.93	This study
Concen-trations	0.04	2.94	7.30	NT	NT	NT	NT	3.28	27.94	Mgbemena *et al* (2019)

NT = Not tested

Table 3: Relative abundance of fatty acids in the *Irvingia gabonensis* biodiesel

Fatty Acid Methyl Ester	Relative Abundance (%)
Linoleic acid	48.00
Palmitic acid	6.72
Stearic acid	10.00
Linolenate acid	7.20
Oleic acid	1.20
Myristic acid	30.53
lauric acid	8.23

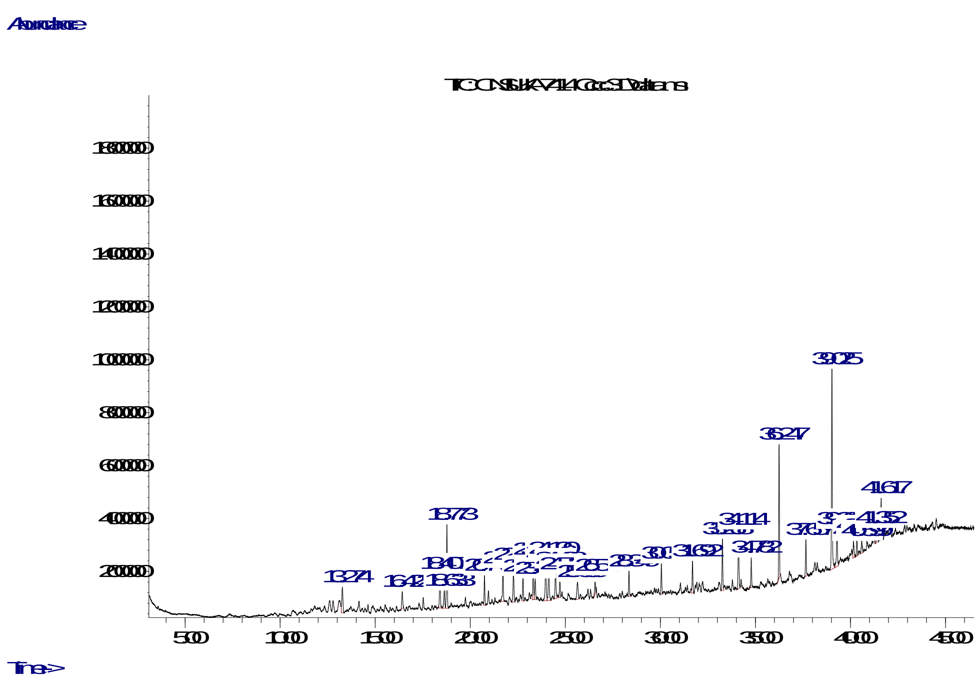

Figure 1: GCMS assay showing different peaks

Table 4: Fuel properties of *Irvingia gabonensis* biodiesel

Parameter	Values	ASTM standard (Sokoto *et al.*, 2018)
Specific gravity (g/cm^3) at 15 °C		
Viscosity	0.87	0.90
Flash point	5.64	1.90 – 6.00
Cloud point	101.00	100.00 – 170.00
Pour point	1.90	-3 – 2.00
Diesel index	4.00	-15 – 10.00
API Gravity	33.51	NA
Colour comparator	27. 12	NA
Aniline point	1.00	NA
Cetane number	49.00	NA
	34.72	48.00 – 65.00

NA = Not available

CONCLUSION

This study determined the chemical composition of *Irvingia gabonensis* from the proximate analysis. This revealed that *Irvingia gabonensis* has high oil content, low ash (mineral), low moisture, and moderate fibre and carbohydrate contents. *Irvingia gabonensis* seed was studied for some metals. It was found that Na, K, Ca, Mg, Cu, Fe, Zn, Pb and Cd were detected at varying amounts. Some of these elements are essential to human health. The potential of *Irvingia gabonensis* seed oil for producing biodiesel was investigated using the transesterification process. It was found that *Irvingia gabonensis* biodiesel had qualities that complied with specifications for biodiesel applications. Therefore, this study confirmed *Irvingia gabonensis* as a viable source of biodiesel that can complement energy supplies from fossil fuels. However, biodiesel requires further testing and optimization of inputs.

Conflict of interest:

The authors have no conflict of interest to declare.

Acknowledgements

National Center for Energy Research and Development, University of Nigeria, Nsukka, is hereby acknowledged for providing the resources including equipment and staff for this project. Dr. Anthony Ofomatah, head of Laboratory, NCERD, is appreciated for carrying out the required analysis in the Biomass Laboratory, NCERD.

Authors contribution:

UKE conceptualized the project, designed the experiment, collected the samples, supervised the project, prepared the draft manuscript, and contributed personal funds to the project. ECG made input during project meetings, proofread the manuscript, and contributed personal funds to the project

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