

Exploring the Valorization of Vegetable Oil Extracted from *Ipomea muricata* Seeds in Food, Cosmetics, Pharmaceuticals, and Bioenergy

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Abstract: Many medicinal plants are veritable reservoirs of molecules that are highly sought after due to their multiple virtues in various fields of application, such as food, pharmaceuticals, cosmetics, and bioenergy. This is the case with *Ipomea muricata*, a species of Togolese flora that is not well known for its therapeutic properties. The main goal of this study is to promote the use of *I. muricata* seed oil in food, cosmetics, pharmaceuticals, or bioenergy. After extracting the oil from the kernels of this plant's seeds using the Soxhlet method, its physicochemical characteristics and mineral content were determined in accordance with AFNOR and Codex Alimentarius standards. The chemical profile of the fatty acids in the oil was established by GC-MS analysis. The oil was converted into biodiesel by transesterification process. The transesterification reaction was catalyzed in a homogeneous phase using KOH as a basic catalyst. The results revealed that the oil content in the kernel was approximately $13.16 \pm 0.87\%$. The physicochemical parameters of the extracted oil are: refractive index (1.46153 ± 0.00100); water and volatile matter content ($12.162 \pm 5.838\%$); acid value: (8.987 ± 0.202 mg KOH/g); saponification value (168.250 ± 4.220 mg KOH/g); ester index (159.26 ± 4.018 mg KOH/g); iodine index (38.065 ± 0.005 g I₂/100 g); peroxide index (7.320 ± 0.001 µg O₂/g); relative density at 29°C (0.90730 ± 0.001); volumic density at 29°C (0.9074 ± 0.0010 g/L, and calorific value (41.040 ± 0.162 MJ/kg). The results also showed that the oil is a significant source of: sodium (56.67 mg/L), iron (16.67 mg/L), potassium (11.33 mg/L), calcium (4.33 mg/L), and manganese (1.83 mg/L). The biodiesel produced is characterized by a lower refractive index and density, compared to the crude vegetable oil used in its production. Generally, the results confirmed that *I. muricata* seed oil owns promising characteristics for its use in food, cosmetics, or bioenergy. However, apart from the kernel, the other parts of the seed could also contain biomolecules of therapeutic interest that should be explored to optimize the value of this seed.

Keywords: *Ipomea muricata*, Soxhlet extraction, physicochemical parameters, mineral content, fatty acid profile, biodiesel production

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1. Introduction

For many centuries, human beings have been constantly seeking knowledge about their environment in order to gain benefits such as food, healthcare, clothing, and energy [1]. Their interest in acquiring this diverse knowledge continues to grow day by day as their needs become more complex over time [2]. To meet most of these needs, humans have been using plants for a very long time. For example, in the health domain, humans treat themselves using plants, through a combination of their instincts, observation, taste, and experience. Plants

are therefore a vital resource for ensuring their health and safety. Moreover, the majority of people in developing countries still rely on traditional medicine for treatment due to several factors that must be taken into account [3]. These factors include their very precarious living conditions, socio-cultural reasons, easy access to medicinal plants, their affordability, and the shortcomings of modern medicine due to a lack of adequate infrastructure and qualified healthcare staff. This is why, according to WHO estimations, more than 80% of the population in developing countries still use traditional medicine to treat illnesses [4,5,6]. African flora in general, and that of Togo in particular, is rich in plant resources with high nutritional value which are not yet used by local

populations in their diets [7]. In addition to the plants that humans commonly cultivate, there are also a large number of little-known wild plants that can offer them significant sociocultural and socioeconomic benefits. These latter plants have significant nutritional, therapeutic, and energy-giving properties [4]. This is exactly the case with *Ipomea muricata*, a plant found in the Togolese flora and which is largely unknown to the general public. Native to tropical America and introduced into Africa [8], *I. muricata* is a plant rarely known for its therapeutic benefits. In fact, this plant is used in the treatment of various diseases such as diabetes, high blood pressure, constipation, tiredness, and inflammatory diseases [9]. In Sri Lanka and India, young seeds, fruits, and thickened pedicels are consumed as vegetables. The mature seed is used as a laxative and purgative for its anthelmintic properties [10,11]. In Togo, the seed is traditionally used to treat insect and snake bites. Similarly, in Burundi, the plant, called “*Umurandaranda*,” is used in combination with other medicinal plants, specifically in the treatment of snake bites. Due to their wide range of chemical compositions, lipids are widely used in various applications, particularly in food, cosmetics, and bioenergy. Thus, lipids are used extensively in the bioenergy industry, where they are converted into biofuels; in cosmetics for their emollient and moisturizing qualities; and in food for their energy function and as a source of fat-soluble vitamins [12,13].

The vegetable oil contained in the seeds of *I. muricata* has several beneficial properties, as it can be used in food preparation and seasoning. It can also be used in several other applications, such as the formulation of cosmetic and therapeutic products [14].

To author knowledge, although this plant is used locally in Togo by traditional practitioners to treat insect and snake bites, its seeds also contain vegetable oil whose fatty acid profile has not yet been established. In addition, this oil could have nutritional and cosmetic properties, or alternatively, it could be used to produce biodiesel, an alternative fuel that is more environmentally friendly than petrodiesel. Therefore, a study of the chemical composition of *I. muricata* oil is necessary to determine what types of applications the oil can be used for. This study was conducted to contribute to the promotion of *I. muricata* seeds in the sectors of food, cosmetics, pharmaceuticals, and bioenergy.

2. Materials and Methods

2.1. Plant Material

The dry seeds of *I. muricata* used in the current study as plant material were harvested in a locality (Figure 1) situated in the Maritime region of Togo, specifically in Akoumape (VO Prefecture), a site with the following geographical coordinates: latitude: 6°23'14"N; longitude: 1°26'59"E.

The plant was identified and preserved at the National Herbarium of the University of Lomé under the number TOGO16015. In Figure 2, photos of the leaves, fruits, and seeds of *I. muricata* plant are shown. The seeds were harvested between July and September 2021, shelled, and

then dried. To prevent deterioration, they were ground into powder and stored in plastic bags pending extraction of the crude vegetable oil.

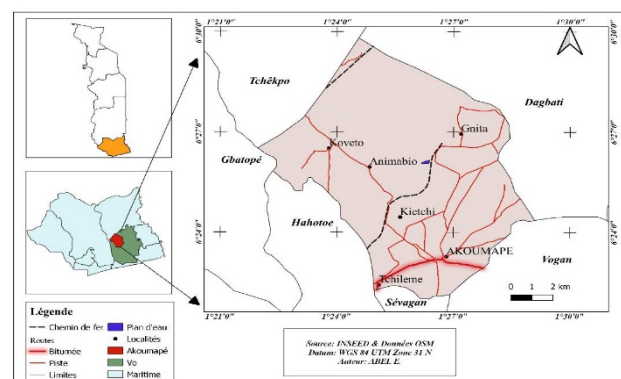


Figure 1. Map showing Akoumape, the site where *I. muricata* seeds were collected



Leaves



Fruits

Figure 2. Leaves and fruits of *I. muricata*

2.2. Method for Extracting Crude Vegetable Oil from Powder

To obtain a good yield of lipids in this study, the solid-liquid extraction method using a Soxhlet extractor was adopted and hexane was used as the extraction solvent. The Soxhlet extraction method is a popular choice in laboratories because it is simple, economical due to solvent recycling, and it allows for high yields. The process involves continuously circulating a solvent through a solid sample, which lets to the efficient extraction of the desired compounds [15]. The main advantages of extraction using the Soxhlet extractor are related to its continuous and efficient process, solvent recycling minimizing consumption, and its capacity for extracting various compounds (lipids, natural products, pollutants) contained in solid matter. The method also allows for high extraction yields due to repeated contact with the solvent, and it is applied in various fields such as

environmental analysis and phytochemistry [16].

2.2. Determination of Oil Extraction Yield

The vegetable oil extraction yield (**Y**: %) was calculated from a pair of tests using formula (F1).

$$Y(\%) = \frac{m_H}{m_p} \quad (F4)$$

Where:

m_p : Average mass of seed powder used for extraction

m_H : Mass of extracted oil.

2.3. Measurements of the Physicochemical Parameters of *I. Muricata* Oil

The main physicochemical parameters of the oil, such as: Water and volatile matter content, Density, Volumic mass of oil, Refractive index, Saponification index, Acid index, Iodine index, and Peroxide index, were determined according to AFNOR standards [14,15,16,17,18]. A pair of tests was performed for each parameter, and the mean of the two tests was taken as the final result.

2.3.1. Water and Volatile Matter Content of the Oil

To determine the water and volatile matter (**WVM**: %) content, 5 g of the oil was weighed into a Petri dish, which was then placed in an oven at $103 \pm 2^\circ\text{C}$ for two hours. It was weighed again after drying in a desiccator. This operation was repeated until a practically constant mass of less than 5 g was obtained. The formula (F2) was used to determine the **WVM** (%).

$$\text{WVM}(\%) = \frac{(M_1 - M_2) \times 100}{(M_1 - M_0)} \quad (F2)$$

Where:

M_0 : mass of the empty Petri dish

M_1 : mass of the loaded Petri dish

M_2 : mass of the Petri dish after drying.

2.3.2. Relative Density and Volumetric Density of the Oil

The relative density of oil at 20°C was determined according to AFNOR standard NF T 60-214 [19] with the modification made by [7]. According to this method, a 50 mL pycnometer was completely filled with distilled water and weighed at room temperature of 29°C . The pycnometer was emptied, rinsed with hexane, and dried with acetone. It was then filled with *I. muricata* oil and weighed again. The density of the oil was calculated using the formula (F3).

$$d^{29^\circ\text{C}} = \frac{m_2 - m_0}{m_1 - m_0} \quad (F3)$$

Where:

m_0 : Mass in g of the empty pycnometer;

m_1 : Mass in g of the pycnometer filled with distilled water;

m_2 : Mass in g of the pycnometer filled with *I. muricata* oil.

2.3.3. Volumetric Mass of the Oil

The volumetric density ($\rho^{29^\circ\text{C}}$) of crude vegetable oil from *I. muricata* was measured using the pycnometric method. The formula (F4) was used to calculate the value obtained.

$$\rho^{29^\circ\text{C}} = d_{29}^{29} \times \rho_e \quad (F4)$$

Where:

ρ_e : Volumetric mass of the water (1,000 g/L);

d_{29}^{29} : Relative density of oil at 29°C .

The arithmetic mean of the results of a pair of tests was used to make corrections based on the temperature at which the analysis was performed and the air pressure in accordance with AFNOR NF T 60-214 standard [19]. The values calculated using formula (F4) were converted to 20°C by applying formulas (F5) and (F6).

$$\rho^{20^\circ\text{C}} = \rho^{29^\circ\text{C}} + 0,00068(\rho^{29^\circ\text{C}} - 20) \quad (F5)$$

$$\rho^{29^\circ\text{C}} = \rho^{20^\circ\text{C}} + 0,0012(1 - \rho^{20^\circ\text{C}}) \quad (F6)$$

2.3.4. Refractive Index of Oil

The refractive index (n_D) of the oil was determined using the "AZZOTA" Abbe refractometer equipped with a thermometer with a temperature range of 20°C to 80°C . The process applied was that used by [7]. Based on this process, distilled water was used as a standard to calibrate the device. The sample was placed on the flat section of a glass prism whose refractive index was assumed to be higher than that of the sample.

The device was illuminated using a light source that emitted rays of light in different directions and focused on the center, allowing a clear area and a dark area to be observed. The refractive index of the oil was read, taking care to ensure that the boundary separating the two areas passed through the point of intersection of the oblique lines in the magnifying glass's viewfinder. The device and the sample were kept at the same constant temperature t_1 before the measurement. The reference temperature of the oils at which the result was expressed is $T_1 = 20^\circ\text{C}$.

The measures were carried out in accordance with the operating instructions provided by the manufacturer. The refractive index values were expressed to the nearest 1/10,000, as absolute values. The formulas (F7) and (F8) were used to determine the refractive index of the oil.

$$n_D^T = n_D^{T_1} + (T_1 - T) \times F \quad \text{if } T_1 < T \quad (F7)$$

Or :

$$n_D^T = n_D^{T_1} + (T - T_1) \times F \quad \text{if } T_1 > T \quad (F8)$$

Where:

T_1 : Measuring temperature;

T : Reference temperature

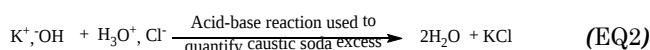
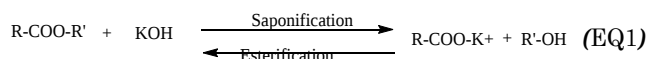
F : 0.00035

2.3.5. Saponification Index of Oil

The saponification index of the crude oil from *I. muricata* was determined in accordance with European standard NF EN ISO 6320 [20]. According to the principle used, the esters of the oil, after being treated in the presence of an excess of strong base (K^+ , OH^-) at a

temperature between 80°C and 100°C, produce a potassium salt of acids and alcohols (most often glycerol), which can lead back to the ester (EQ1). The excess KOH was then titrated with a HCl (0.4 N) solution. The equation for this acid-base titration reaction is shown below (EQ2).

For this purpose, 25 mL of ethanolic solution of potassium hydroxide KOH (0.4 N) was added to 2 g of *I. muricata* oil. The mixture was heated to reflux for 2 hours with continuous stirring using a magnetic stirrer, then hot-titrated with a hydrochloric acid HCl (0.4 N) solution in the presence of a few drops of phenolphthalein.



A blank test was performed under the same conditions. The formula (F9) was used to calculate the saponification index (SI) of the oil.

$$\text{SI} = \frac{(V_0 - V_1)}{m} \times T \times 56.1 \quad (\text{F9})$$

V_0 : Volume in mL of 0.4 N hydrochloric acid used to perform the blank test;

V_1 : Volume in mL of 0.4 N hydrochloric acid used to determine the InS;

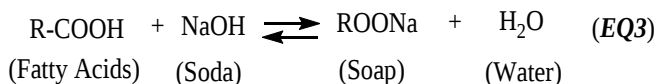
T: Exact titer of the hydrochloric acid solution;

M: Mass in g of the oil during the test.

2.3.6. Acid Index and Acidity of the Oil

The acid index (AI) and acidity (AC) were determined according to the AFNOR standard [19]. In this study, 5 g of *I. muricata* oil was weighed in an Erlenmeyer flask. Then, 50 mL of ethanol previously neutralized with an ethanolic KOH solution (0.9 N) was added to the Erlenmeyer flask containing the oil.

After homogenization of the mixture, the free fatty acids (FFA) were titrated with KOH solution (0.9 N). The end of the titration was detected by the presence of a pink coloration, used as a suitable color indicator of phenolphthalein. The balanced equation for this acid-base titration reaction is written as follows (EQ3).



The acidity index (AI) and acidity (AC) were calculated using the formulas (F10) and (F11).

$$\text{AI} = \frac{V \times T \times 56.1}{m} \quad (\text{F10})$$

$$\text{AC} = \frac{V \times T \times M}{10 \times m} \quad (\text{F11})$$

Where:

V: Volume of the ethanolic KOH solution used during the titration;

T: Titration factor of the ethanolic KOH solution;

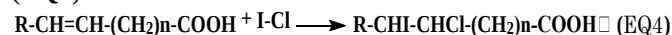
m: Mass (g) of the sample;

M: 282 g/mol, equal to the average molar mass of the fatty acids present in the oil.

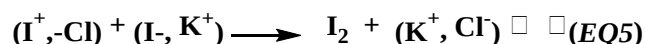
2.3.7. Iodine Value of Oil

The iodine index (Iod-V) of the crude vegetable oil was determined using AFNOR NF T60 – 220 standard [19]. According to the experimental protocol applied, the oil was introduced into chloroform containing an excess of iodine chloride (I-Cl), commonly known as Wijs' reagent. After a few minutes of reaction, a solution of potassium iodide and distilled water were added to the previous mixture. The released diiodine is titrated with a sodium thiosulfate solution (0.1 N) in the presence of starch paste acting as a color indicator [7].

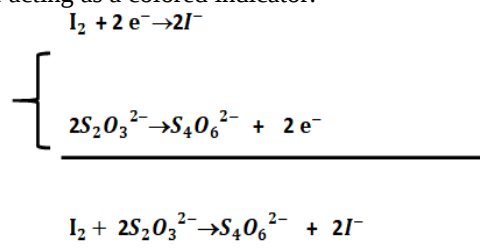
This method is based on the treatment of oil with excess iodine monochloride (Wijs' reagent) according to reaction (EQ4).



Then, by the action of potassium iodide (KI) on the excess of iodine monochloride (I-Cl), the iodine (I_2) is released according to the equation (EQ5).



The released iodine (I_2) was determined by oxidation-reduction reaction (EQ6) using a sodium thiosulfate Na_2SO_3 (0.1 N) titrant solution in the presence of starch paste acting as a colored indicator.



During this analysis, 7.5 mL of chloroform was added to 250 mL Erlenmeyer flask containing 1 g of oil. After vigorously shaking the mixture, 12.5 mL of Wijs' reagent was added. The mixture was then placed in the dark. After incubating for one hour, 10 mL of a potassium iodide solution and 75 mL of distilled water were added to the mixture. The final solution obtained was titrated with an aqueous solution of Na_2SO_3 (0.012 N) in the presence of starch paste. A blank test was performed under the same conditions. The iodine value of the oil was calculated using formulas (F12) and (F13).

$$\text{Iod} - \text{V} = \frac{(V_0 - V_1) \times T \times 12,69}{P} \quad (\text{F12})$$

$$\text{P} = \frac{m \times 2}{5} \quad (\text{F13})$$

Where:

V_0 : Volume in mL of Na_2SO_3 (0.2 N) solution used to perform the blank test;

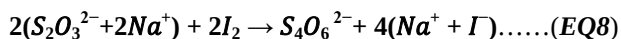
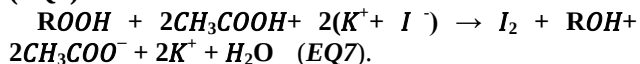
V_1 : Volume in mL of 0.2 N Na_2SO_3 solution used to perform the test;

T: Exact titer of Na_2SO_3 solution

M: Mass (g) of the test sample.

2.3.8. Peroxide Value of the Oil

The peroxide value (PV) of an oil was assessed by applying AFNOR NFT 60-220 standard [19]. The principle of this experimental analysis stipulated that the lipids, dissolved in a mixture of acetic acid and chloroform, were treated with a potassium iodide solution. This resulted in the reaction shown in the equation (EQ7). The iodine released is titrated with a sodium thiosulfate solution according to the following reaction equation (EQ8).



For this analysis, 2 g of oil and 10 mL of chloroform were placed in a conical flask with a ground glass stopper. Next, 15 mL of acetic acid and 1 mL of potassium iodide solution were added successively, and the flask was immediately sealed. The mixture was shaken and then placed in the dark for 5 min. A volume of 75 mL of distilled water was added to the previous mixture before titrating with $Na_2S_2O_3$ (0.012 N) solution in the presence of starch paste. A blank test was performed under the same conditions. The formula (F14) was used to determine the oil's peroxide value.

$$PV = \frac{(V_1 - V_0) \times N \times 1000}{M} \quad (F14)$$

Where:

V_0 : Volume (mL) of $Na_2S_2O_3$ (0.012 N) solution used to carry out the blank test;

V_1 : Volume (mL) of the $Na_2S_2O_3$ (0.012 N) solution used to carry out the test itself;

M: Mass (g) of the test sample;

N: Exact titer of the $Na_2S_2O_3$ solution used.

2.3.9. Ester Index of the Oil

The ester index (EI) of a vegetable oil cannot be measured experimentally, but it has been deduced by calculating the difference between the saponification index (SI) and the acid index (AI) according to formula (F15) [7].

$$EI = SI - AI \quad (F15)$$

2.3.10. Calorific Value of the Oil

The calorific value (CV: expressed in kJ/kg) used by was calculated using formula (F16) [7].

$$CV = 47645 - 4,187 \text{ Iod-I} - 38,31 \text{ SI} \quad (F16)$$

2.4. Quantification of Mineral Elements Contained in the Oil

After mineralization by wet destruction of organic matter using the combined action of hydrogen peroxide, nitric acid, and sulfuric acid, the mineral content of *I. muricata* vegetable oil such as potassium (K), sodium (Na), calcium (Ca), magnesium (Mg), and phosphorus (P) in *I. muricata* vegetable oil were measured by flame atomic absorption spectrophotometry.

2.5. Chemical Composition of *I. muricata* Oil in Fatty Acids

2.5.1. Preparation of the Sample for GC-MS Analysis

To prepare the methyl esters of fatty acids, 20 mg of crude vegetable oil (CVO) from *I. muricata* was dissolved in a methanolic solution of 2.5% H_2SO_4 . The mixture was stirred thoroughly, then heated in an oven at 80°C for 90 min. After cooling, 2.5% NaCl (0.9%) solution was added to the previous mixture, followed by vigorous stirring.

The final solution containing fatty acid methyl esters (FAME) was extracted with 1.5 mL of hexane. The hexane extract obtained was analyzed by GC-MS to determine the fatty acid composition of *I. muricata* oil.

2.5.2. Conditions for Analyzing FAME Prepared by GC-MS

The previously prepared fatty acid methyl esters (FAME) were analyzed using a chromatograph (TRACE 1300 Series GC) equipped with a DB5 capillary column - MS capillary column (length: 50 m, internal diameter: 0.25 mm, film thickness: 0.25 μ m) and an automatic injector (Auto sampler AIS/AS 1310) and coupled to an ISQ MS Tune mass spectrometer (MS).

The detector used was an electron impact detector equipped with XCalibur data acquisition software. The spectra were recorded at 70 eV. The analyses were performed under the following conditions: injector temperature: 200°C; furnace temperature was initially maintained at 120°C for 10 min; then it was adjusted from 120 to 210°C at a rate of 2°C/min and kept constant for 10 min; then it was raised again to 300°C at a rate of 5°C/min and kept isothermal for 2 min. The carrier gas was helium with a flow rate of 1 mL/min.

The identification of the different fatty acids in the hexane extract was performed by comparing their mass spectra with those stored in the XCalibur software databases.

2.6. Conversion into Biodiesel of the Oil

The conversion of *I. muricata* crude vegetable oil (CVO) into biodiesel was carried out in two steps, the first of which was pretreatment consisting of esterifying the FFA acids contained in the *I. muricata* CVO, while the second step was transesterification.

2.6.1. Pre-Treatment of the Oil by Esterification

Pre-treatment by esterification is a crucial step in biodiesel production when the FFA content of a fatty substance exceeds 2% of the recommended limit [21].

The principle of the esterification reaction consisted of reacting a carboxylic acid with an alcohol to obtain an ester. This reaction mechanism has the advantage of converting the fatty acids (FAs) contained in CVO of *I. muricata* into esters in order to optimize the transesterification reaction of the final product.

The experimental protocol adopted was similar to that described by [22] and [23]. A mass of 10 g of CVO from *I. muricata* was placed in a 250 mL Erlenmeyer flask, which had been washed and dried beforehand. The sample was preheated to a temperature of 60°C using a hot plate with automatic magnetic stirring for at least 30 min.

The stirring speed was maintained at 500 rpm, then methanol and sulfuric acid (catalyst) were added to the

previous mixture. The Erlenmeyer flask containing the mixture was closed with its stopper to prevent the methanol from evaporating from the reaction medium. At the end of the reaction, the mixture was left in a separating funnel and the acidity of the pretreated oil was reevaluated.

At the end of the esterification reaction, the Erlenmeyer flask containing the sample was removed from the hot plate and the content was transferred to a separating funnel. Two phases formed in the separating funnel: the upper phase consisted of methanol acidified with H_2SO_4 used as a catalyst, while the lower phase consisted of the pretreated oil. The latter phase was recovered and its acidity measured.

To determine the mass of methanol required to carry out the esterification, formula (F17) was used.

$$m_{MeOH} = \frac{N \times m_{FFA} \times M_{MeOH}}{M_{FFA}} \quad (F17)$$

Where:

m_{MeOH} : Mass (g) of methanol;

M_{MeOH} : Molar mass (g/mol) of methanol;

N : Number of moles of methanol;

M_{FFA} : Molar mass (g/mol) of free fatty acids.

Palmitic acid, with a molar mass of 256 g/mol, was considered to be the major fatty acid. The mass of methanol to be sampled corresponds to a given volume, which was calculated using the formula (F18).

$$V_{MeOH} = \frac{W_{MeOH}}{0,7915} \quad (F18)$$

With:

V_{MeOH} : volume (mL) of methanol

W_{MeOH} : Weight of methanol;

0.7915: Density (g/L) of methanol.

The formula (F19) was used to determine the mass of the predominant fatty acid. The mass of FAs is 0.8987 g.

$$W_{FFA} = \frac{W_s AI}{100} \quad (F19)$$

Where:

W_{FFA} : Weight (g) of FFA;

W_s : Mass of the sample;

AI: Acid Index of the oil.

2.6.2. Conversion the Oil into Biodiesel

The process used to convert the oil into biodiesel was similar to that used for pre-treatment, except that this time the transesterification reaction was catalyzed in a homogeneous phase using potassium hydroxide (KOH) as a basic catalyst.

According to the experimental protocol used, 10 g of fat was preheated in a 250 mL Erlenmeyer flask using a hot plate at 60°C for at least 30 min to ensure better dissolution of the fat. To optimize the transmethylation reaction, the KOH pellets used as a catalyst were first dissolved in methanol at 65°C, and the resulting potassium methanolate (CH_3O-Na^+) solution was added to the Erlenmeyer flask containing the previously esterified vegetable oil.

Next, the Erlenmeyer flask was immediately closed with its stopper and the mixture was stirred at 600 rpm

with a magnetic stirrer. After 1 hour, the Erlenmeyer flask was removed, the content was transferred to a separating funnel, and the mixture was left to settle. After 24 hours of settling, two distinct phases formed: the upper, clearer phase, consisting mainly of methyl esters, called biodiesel, and the lower phase, consisting mainly of glycerol and catalyst (Figure 3).

After separating the two phases, the biodiesel obtained was purified by washing with distilled water until the water from the last wash no longer colored the phenolphthalein purple. Finally, the washed biodiesel was dried before being stored in a hermetically sealed bottle.

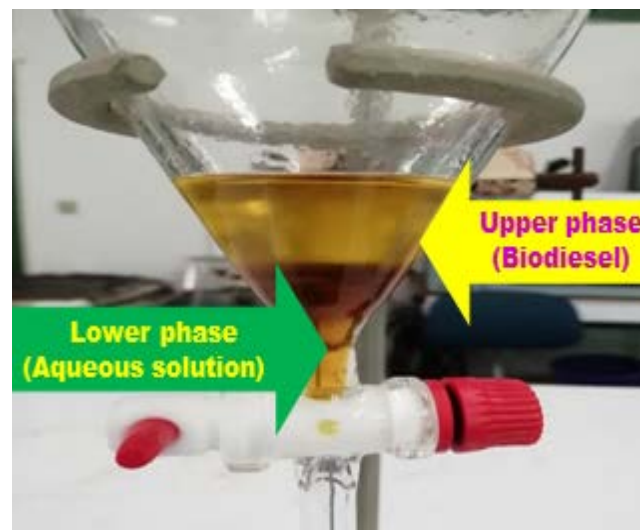


Figure 3. Photo showing the separation of the two phases formed after transmethylation

The biodiesel production yield (η) was calculated using the mass of the biodiesel produced ($m_{Biodiesel}$) and the mass of the oil used (m_{Oil}), as shown by formula (F20).

$$\eta = \frac{m_{Biodiesel}}{m_{Oil}} \times 100\% \quad (F20)$$

3. Results and Discussion

3.1. Extraction Yield of Crude Vegetable Oil from *I. muricata*

The extraction yield of crude vegetable oil (CVO) from *I. muricata* seeds is shown in Table 5.

Table 5. Vegetable oil extraction yield from *I. muricata* seeds

Parameters	Results
Extraction method	Solid-liquid extraction with the Soxhlet extractor
Extraction solvent	Hexane
Number of tests	2
Yield (%)	13.16 ± 0.87

The results presented in Table 5 show that solid-liquid extraction from *I. muricata* seed powder, performed using the Soxhlet method and hexane as the solvent, yields with CVO of $13.16 \pm 0.87\%$. This yield obtained in this study is higher than that obtained from the seeds of the same plant by [24], which was 8.7%.

The disparity between the two yields mentioned above

can be explained by several factors, including, for examples: the nature of the extraction solvent, the climate zone where the plant is harvested, the nature of the soil, the degree of seed maturity, crop genetic characteristics, the harvest period, and the type of pre-treatment carried out during extraction [25]. In the case of the current study, roasting the seeds could possibly improve the extraction yield of the oil contained in *I. muricata* seeds.

3.2. Physicochemical Characteristics of *I. muricata* Oil

The physicochemical parameters determined for HVB extracted from *I. muricata* seeds in this study are recorded in Table 6.

Table 6. Physicochemical parameters of the oil of *I. muricata*

Physicochemical parameters	Calculated values
Color	Yellow
Refractive index (n_D^T , T = 20°C)	1.46153 ± 0.00100
Water content volatile matter in oil	12.162 ± 5.838
Acid value (mg KOH/g oil)	8.987 ± 0.202
Acidity (g of oleic acid/100 g of oil)	4.670 ± 0.253
Saponification index (mg KOH/g oil)	168.250 ± 4.220
Ester index (mg KOH/g oil)	159.263 ± 4.018
Iodine value (g I ₂ /100 g oil)	38.065 ± 0.005
Peroxide value (µg O ₂ /g oil)	7.320 ± 0.001
Relative density at 29°C	90.730 ± 0.001
Volumetric mass (g/mL)	0.9074 ± 0.0010
Calorific value (MJ/kg)	41.040 ± 0.162

3.2.1. Color

The extracted oil is a liquid and yellowish in color at room temperature (25-32 °C) as shown in Figure 4.



Figure 4. Photo showing CVO extracted from *I. muricata* seeds

3.2.2. Refractive Index of *I. muricata* Oil

The refractive index value of the *I. muricata* oil, 1.461 ± 0.001, meets the standard established for edible oils by [18]. This value is less similar to that found for the same plant by [24], which was 1.465 at 30°C and 1.468 at 20°C for *Ipomea digitata* according to [26]. This value is also comparable to the refractive index of other vegetable oils, such as olive, palm, and avocado oils, which are 1.470, 1.458, and 1.474, respectively [27].

The refractive index is considered a guideline for the purity of an oil. Its value varies depending on the wavelength of the incident light used to determine it, as well as the temperature of the analysis [7]. This refractive index is also proportional to the molecular weight of fatty acids and their degree of unsaturation.

3.2.3. Water Content and Volatile Matter in the *I. muricata* Oil

According to this study, the water and volatile matter content (12.162 ± 5.837%) obtained from *I. muricata* oil is relatively high compared to the standard, which sets the maximum content at 0.2% at a temperature of 20°C for edible oils [18]. A high water and volatile matter content in vegetable oil may cause quickly the rancidity during storage. Therefore, to prevent premature rancidity of this oil, it must be dehydrated before being stored away from heat and light, because water activity > 0.3% promotes enzymatic oxidation of the oil [7].

3.2.4. Acid Index of the Oil

Generally, the acid index (AI) of an oil provides information about the level of FFA present in that oil. In this study, the acid value found for *I. muricata* oil is 8.987 ± 0.202 mg KOH/g of oil, equivalent to an acidity of 4.476 ± 0.253% (linoleic acid). This acid value is above the upper limit of the range (2.20-7.26 mg KOH/g) established by the AFNOR standard [19] for the proper preservation of vegetable oils.

This acidity (AC) is higher than the maximum value of 3% recommended for edible oil [18]. A low acidity value characterizes the purity and stability of a fat at room temperature. However, this is not the case of the CVO extracted from *I. muricata* seeds, explored in this study. Consequently, pre-refining and conditioning precautions must be taken to limit the likely subsequent denaturation of this oil during storage [7].

3.2.5. Saponification Index

The saponification index of fats or oils is an appropriate parameter for evaluating the length of the carbon chain of the acids contained in that fats and oils. Indeed, this index gives high values for short-chain fatty acids. The saponification index of the *I. muricata* oil studied in this work (168.590 ± 4.220 mg KOH/g oil) is lower than that found by [24], which was 200.00 mg KOH/g oil for the same plant. However, this index is still higher than that obtained by [25], which was 160 mg KOH/g oil for *Ipomea digitata*. This difference could be due to both the extraction method used and the type of solvent used, according to a study conducted by [28] on *Moringa oleifera* seed oil.

3.2.6. Iodine Value of the Oil

The iodine value provides information on the degree of fatty acid formation in an oil and is the most commonly used constant, because this value determines the classification of vegetable oils as drying, semi-drying, or non-drying oils. It is directly related to the degree of oxidation of an oil. Thus, to assess how an oil will easily go rancid, this value can be used as a basis, since the more unsaturated an oil is, the higher its iodine value; so, the

more instaurations it will contain, the more sensitive it will be to oxygen [7].

The iodine value of our oil, 38.065 ± 0.005 g I₂/100 g of oil, is lower than that reported by [24] and [25], which are 68.67 g I₂/100 g and 81 g I₂/100 g, respectively. This value is lower than those of olive, peanut, and castor oil, which vary between 75 and 94 g I₂/100 g of oil [18]. This value indicates that the vegetable oil extracted from *I. muricata* seeds is less unsaturated than olive, peanut, and castor oil, and therefore, *I. muricata* oil could be considered as low food quality.

Given the relatively low iodine value of the oil, it can be stored without too much risk of auto-oxidation, as it has a low concentration of unsaturated fatty acids [7]. In addition, the vegetable oil from *I. muricata* seeds must be classified as non-drying because its iodine value is less than 100 [29].

3.2.7. Relative Density and Volumetric Density of the *I. muricata* Oil

The relative density of an oil is the ratio between the mass of a given volume of that oil at 20°C and the mass of the same volume of distilled water at the same temperature. Whether mineral or vegetable, the relative density of many vegetable oils ranges between 0.840 and 0.960. It is always lower than that of water (1.000), which explains why oils float on water and always remain above it. It depends on temperature and, of course, on the chemical composition of the oils in terms of fatty acids. It decreases as the temperature increases, hence the need to express the relative density of an oil as a function of its temperature [30].

In this study, the relative density of *I. muricata* oil is 0.9074 (29°C). This value is slightly higher than that reported by [24], who obtained a value of 0.916 at 30°C for the same plant. Furthermore, this value is very close to and meets the standard required by [18].

With regard to the volumetric density of the oil found in this investigation, is of 0.9074 ± 0.0010 g/mL, which is logically lower than that of water.

3.2.8. Peroxide Value of the Oil

The peroxide value of an oil is a very important factor in assessing the early stages of possible oxidative deterioration of the oil. The peroxide value found for the oil extracted from *I. muricata* seeds in this study is 7.320 ± 0.001 µg O₂/g of oil. This value is lower than 10 µg O₂/kg, which characterizes most conventional edible oils such as soybean, corn, and sunflower oils [18]. Consequently, *I. muricata* oil is likely to be rich in natural antioxidants, such as tocopherols, sterols, and carotenoids [28,31].

3.2.9. Ester Index of the Oil

The ester index value of the oil extracted from *I. muricata* seeds in this study is 159.263 ± 4.018 mg KOH/g of oil. The ester index of this oil is lower than its saponification index, which is 168.250 ± 4.220 mg KOH/g of oil. This implies that this oil contains a significant amount of FFA. Therefore, pre-refining and conditioning precautions must be taken beforehand to potentially limit subsequent denaturation that would lead to oil discoloration.

3.2.10. Calorific Value of the Oil

The calorific value (CV) of the crude vegetable oil from *I. muricata* seeds, which is the subject of this study, is 41.040 ± 0.162 MJ/kg, which is almost close to the calorific value of diesel, approximately 42-46 MJ/kg. The theoretical calorific value of this oil suggests that it has the potential to be used as an alternative fuel in compression ignition diesel engines.

This makes this oil a potential alternative fuel that could be used in its raw state or after conversion to biodiesel, as well as a bio-lubricant for diesel engines if it can remain liquid at ambient temperature. However, the use of this oil as a biofuel could raise ethical issues if further studies show that the oil is edible. If the oil is edible, this parameter would provide positive information on the calorific value of this fat in human nutrition, given the pharmacological usefulness of this species [7]. Another avenue to consider for adding value to this oil is its potential use in cosmetics.

3.3. Mineral Composition of the Oil of *I. muricata* Seed

In this study, the mineral content of *I. muricata* oil is presented in Table 7. The results included in this Table show that *I. muricata* seed oil is a significant source of sodium (56.67 mg/L), iron (16.67 mg/L), potassium (11.33 mg/L), calcium (4.33 mg/L), and manganese (1.83 mg/L). Zinc (0.50 mg/L), which is also present in the oil in trace amounts, is a "trace element."

For example, sodium and potassium play a role in regulating the water content of the human body and actively participate in maintaining acid-base balance [7].

Table 7. Mineral content of *I. muricata* oil

Mineral elements	Concentration (mg/L)
Na	56.67
K	11.33
Ca	4.33
Mg	1.83
Fe	16.67
Zn	0.50

Potassium is one of the elements that controls various metabolic reactions. It is also necessary for carbohydrate and protein metabolisms and cell growth. In fact, potassium deficiency causes hypokalemia. However, when the potassium content in the blood becomes very high, it induces cardiac arrhythmia and can even be fatal in cases of ventricular fibrillation [32]. Similarly, high sodium levels in the human body can cause edema and high blood pressure [7].

Table 8 shows the Na/K and Ca/Mg ratios, taking into account the mineral content of *I. muricata* seed oil as shown in Table 7.

Table 8. Na/K and Ca/Mg ratios

Ratios	Values obtained
Na/K	5.02
Ca/Mg	2.36

The Na/K and Ca/Mg ratios are very interesting because they exceed 1, indicating that this oil should be used cautiously in food if it does not contain any toxic substances. It is well known that calcium is necessary for bone growth. Calcium deficiency in the human body causes growth retardation and rickets in children, but also spasmophilia in adults and osteoporosis in the elderly.

The presence of magnesium in this *I. muricata* seed oil is particularly beneficial for consumers, as magnesium is vital for bones and the nervous system [7]. It plays an active role in energy metabolism and the synthesis of proteins and nucleic acids in humans. It also helps stabilize cell membranes. Finally, it also helps maintain sodium, calcium, and potassium homeostasis [33]. As for zinc, it is important for cell growth and wound healing [7].

The body needs minerals to sustain bone construction, regulate nerve and muscle function, participate in blood coagulation, maintain water balance, and act as cofactors in a variety of biochemical activities. Lipids, often known as fats or oils, are structural components and sources of energy. They interact significantly with minerals, especially when it comes to mineral balance and the absorption of fat-soluble vitamins. Serious health issues including iron deficiency-induced anemia or magnesium deficiency-induced neuromuscular diseases might result from mineral deficiencies [34].

In fact, the therapeutic properties of a vegetable oil depend not only on its chemical composition in fatty acids but also on other minor constituents, notably vitamins, phytosterols, carotenoids, tocopherols, tocotrienols, as well as terpenic alcohols, squalene, phenolic compounds, certain minerals, etc. [12].

3.4. Fatty Acid Profile of *I. muricata* Oil

The results of GC-MS analysis of fatty acid methyl esters prepared from CVO from *I. muricata* seeds helped

to determine the fatty acid composition of the oil using the chromatogram shown in Figure 5.

The predominant fatty acids that make up the oil from *I. muricata* seeds are as follows: (C16:1 ω^7): (Z)-hexadec-9-enoic acid (0.1611%); (C16:0): Palmitic acid (55.09%); (C18:0): Stearic acid (13.74%); (C18:1 ω^9): Oleic acid 14.60%; (C18:2 ω^6): Linoleic acid (12.89%) and (C20:0): Arachidic acid (2.35%) (Table 9). The chemical structures of these fatty acids identified in this oil are illustrated in Figure 6.

Table 9. Fatty acid composition of *I. muricata* seed oil

Fatty acids	Composition (%)
Palmitoleic acid (C16: 1 ω^7)	0.16
Palmitic acid (C16: 0)	55.09
Stearic acid (C18: 0)	13.74
Oleic acid (C18: 1 ω^9)	14.60
Linoleic acid (C18: 2 ω^6)	12.89
Arachidic acid (C20: 0)	2.35
Total saturated fatty acids (SFAs)	71.18
Total unsaturated fatty acids (UFAs)	27.65
Total identified	9
AGI/AGS ratio	0.39

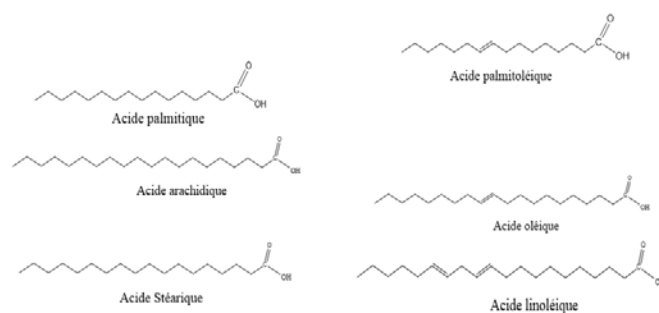


Figure 6. Fatty acid structures of *I. muricata* oil

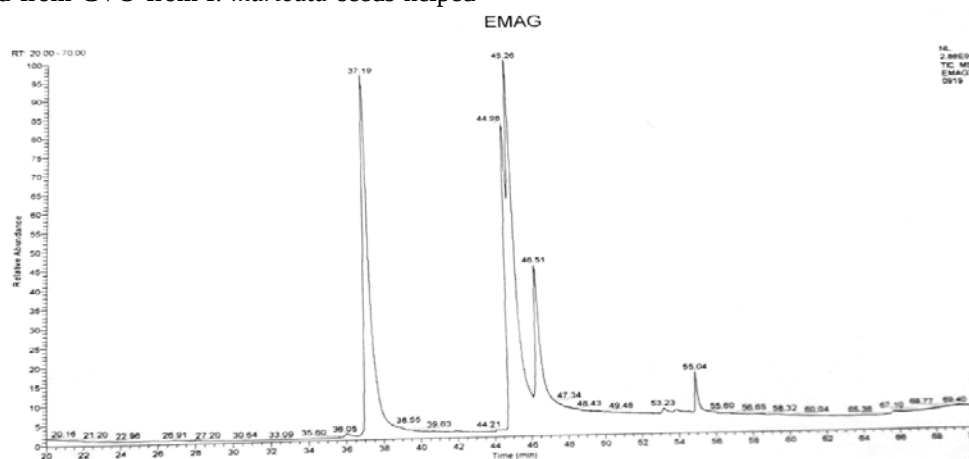


Figure 5. Chromatogram of vegetable oil extracted from *I. muricata*

The results of this analysis show that the oil contains mainly saturated fatty acids (SFAs) with an overall level of 71.18% compared to 27.49% for unsaturated fatty acids (UFAs). These results also indicate that *I. muricata* seed oil is a good source of palmitic acid with a proportion of 55.09%.

The fatty acid composition of this oil presented in this study differs only in the presence of arachidic acid and palmitoleic acid, which were absent in the oil composition

reported by [24] for the same plant. The absence of behenic acid in the current fatty acid composition, but reported by [35] in the oil of this plant, is a remarkable feature of the oil studied here. With the exception of missing fatty acids such as heptadecanoic acid and heptadecenoic acid, the oil analyzed in this study has almost the similar fatty acid composition as peanut oil, but with different levels [30].

In this study, the presence of omega-6 fatty acids

(linoleic acid) and omega-9 fatty acids (oleic acid) gives this oil a definite advantage, as it can also be seen in some fats such as peanut oil. In fact, fatty acids from the omega-6 and omega-9 series are both naturally considered essential fatty acids for the proper functioning of the human body, but the body itself is unable to synthesize them.

Therefore, the only way for the body to benefit from these essential nutrients is through dietary intake. When consumed in the right proportions, omega-6 fatty acids help lower cholesterol (LDL cholesterol, considered “bad” cholesterol) and thus reduce the risk of cardiovascular disease. As a result, they help lower blood pressure and contribute to the synthesis of several molecules (prostaglandin E2, thromboxane A2, and leukotriene B4) that play a mediating role in inflammatory and immune responses. They help maintain the skin’s “barrier” function against toxins and facilitate the passage of nutrients into the epidermis [36].

The data presented in Table 10 compare the SFA, MUFA, and PUFA lipid profile of *I. muricata* oil with that of some of the world’s major edible oils [37].

An overall analysis of the results in Table 10 reveals that the SFA profile (71.18%) of *I. muricata* oil lies between that of palm oil (37-54%) and coconut oil (80-93%), which are considered as the oils with total SFA contents exceeding those of total PUFA.

Table 10. Comparison of the lipid profile of SFA, MUFA, and PUFA of *I. muricata* BV compared to some of the main consumable oils in the world [37]

Types of lipids	SFAs	MUFAs	PUFAs	ω^3
<i>I. muricata</i> oil	71.18%	14.76%	12.89%	---
Palm oil	37-54%	36-44%	6-12%	---
Coconut oil	80-93%	5-10%	2%	---
CVO rich in MUFAs- ω^9				
<i>I. muricata</i> oil (ω^9 : 14.60%)				
Olive oil	12-22%	63-82%	---	---
Rapeseed oil	4-9%	52-67%	26-35%	10%
Peanut oil	13-27%	33-67%	---	---
Hazelnut oil	6-10%	72-85%	6-22%	---
Avocado oil	9-21%	58-81%	10-14%	---
CVO rich in PUFAs- ω^6				
<i>I. muricata</i> oil (ω^6 : 12.89%)				
Sunflower oil	6-20%	13-41%	17-85%	---
Soybean oil	12-22%	18-29%	59-65%	5-11%
Grape seed oil	4-20%	12-29%	40-78%	---
Corn oil	8-24%	20-42%	45-46%	2%
Cottonseed oil	21-42%	14-40%	53-62%	> 1%
Walnut oil	4-12%	17-22%	50-80%	3-16%
CVO rich in PUFAs- ω^3				
<i>I. muricata</i> oil (ω^3 : 0.00%)				
Linseed oil	6-25%	18-20%	51-80%	35-66%
Camelina oil	6-13%	12-24%	45-66%	33-43%

CVO: Crude vegetable oil; SFA: Saturated fatty acids; UFA: Unsaturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: Polyunsaturated fatty acids.

These results show that this oil would be more suitable for frying than for seasoning, since it is known that SFA are more heat-resistant than PUFA. On the other hand, PUFA are more suitable for food consumption, as they are more biodegradable than SFA.

The MUFA content belonging to the ω^9 (14.60%) of the CVO from *I. muricata* is almost similar to that of PUFA

belonging to the ω^9 series of edible vegetable oils such as hazelnut oil (6-22%) and avocado oil (10-14%). Similarly, the content of PUFA belonging to the ω^6 series (12.89%) of CVO oil from *I. muricata* is roughly similar to that of MUFA belonging to the ω^6 series of CVO from sunflower (13-41%), soybean (18-29%), grape seed (12-29%), and cottonseed oil (14-40%).

However, the CVO from *I. muricata* is noticeable by its UFA content belonging to the ω^3 series which is zero; which makes it closer to many of the food CVO, notably those from: palm, coconut, olive, peanut, hazelnut, avocado, sunflower and grape seeds [37].

The results presented in their entirety on the lipid profile of SFA, MUFA, and PUFA of *I. muricata* CVO suggest that it could be used in frying when it is not toxic to humans. Otherwise, it could be used for cosmetic applications because the fatty acids belonging to the ω^6 series which this oil contains are very good for maintaining the skin. In addition, it could also be used for other purposes, including the formulation of pharmaceutical products, bio-lubricants or the production of alternative fuels from renewable sources.

3.5. Characterization of the Biodiesel Produced from *I. muricata* oil

The results of the yield, the refractive index and the density of biodiesel produced from *I. muricata* seed oil compared to those of CVO of this plant are presented in Table 11.

Table 11. Yield, refractive index, and density of biodiesel produced from *I. muricata* seed oil compared with the oil

Parameters	Determined values	
	<i>I. muricata</i> CVO	Biodiesel of <i>I. muricata</i>
Production yield (%)	13.16 ± 0.87	22.50 ± 1.45
Refractive index	1.461 ± 0.001	1.441 ± 0.001
Relative density (29°C)	0.907 ± 0.001	0.807 ± 0.001

3.5.1. Yield of Biodiesel Produced

The yield of biodiesel produced in this work is 22.50 ± 1.45% relatively to the mass of the oil used. This yield expresses that the losses are more or less considerable during the transesterification reaction. Consequently, the transesterification requires a possible optimization of the overall process in order to minimize possible losses.

3.5.2. Refractive Index of the Biodiesel Produced

The refractive index is one of the most important characteristics of biodiesel. In the current study, this parameter was measured using an Abbe refractometer and the value found is 1.441 ± 0.001. This value is much lower than that corresponding to the *I. muricata* CVO which was used as raw material for the biodiesel production, which is 1.461 ± 0.001. However, this result found is still low compared to the value found by [38] for palm oil unfit for human consumption, which was 1.4495 ± 0.0007.

This decrease in the refractive index of the biodiesel produced compared to the vegetable oil used for its production, could be due to the elimination of the glycerol in the biodiesel, thus leading to the fluidity of the biodiesel, thus to the reduction of the biodiesel density.

3.5.3. Relative Density

The conversion of CVO into biodiesel resulted in a decrease in relative density (29°C) from 0.907 ± 0.001 g/mL for CVO to 0.807 ± 0.001 g/mL for the biodiesel produced from *I. muricata* CVO used (Table 10). This indicates that the process of converting crude vegetable oil from *I. muricata* into biodiesel was indeed successful. This result is in agreement with the work carried reported by [38] who also noted that the relative density of the crude vegetable oil which was 0.9005 ± 0.0007 had become 0.8595 ± 0.0007 for the biodiesel produced, demonstrating the interest of this technology for obtaining an alternative fuel whose physicochemical properties would be much more compatible for the proper functioning of a diesel engine.

The valorization of CVO into biodiesel as done in the current study is a viable alternative that lowers greenhouse gas emissions because it is biodegradable and renewable. However, it can also pose technical difficulties, like oxidation-related shelf-life limitations and the need to prevent water contamination, which can lead to filtration issues [39].

This energy transition is only economically viable if the raw materials are exploited with negligible market value, such as food waste, like vegetable oils unfit for human consumption. On the other hand, the use of food products in energy production can cause specific issues such as starvation, increased food prices, confiscation of agricultural land for the growth of plants dedicated to bioenergy production, with dramatic consequences on all different terrestrial ecosystems [40].

4. Conclusion

The main objective of this work was to contribute to the promotion of *I. muricata* seeds in food, cosmetics, pharmaceuticals, and bioenergy.

Solid-liquid extraction using the Soxhlet method yielded an oil content of at least one-tenth of the dry weight of *I. muricata* seed powder.

The results showed that the CVO of *I. muricata* has physicochemical characteristics similar to those of fats in general, particularly density, specific gravity, and refractive index. These physicochemical characteristics complied with [18] standards, with some exceptions. Most of the physicochemical characteristics measured were more or less similar to those of certain edible oils commonly sold around the world.

The results revealed also that *I. muricata* seeds are an excellent source of various minerals, the most important of which are sodium, iron, potassium, calcium, and manganese. The Na/K and Ca/Mg ratios are very significant for this oil, as their values exceed one (01). However, this oil should be used sparingly in food.

The results of this GC-MS analysis identified an unmatched predominance of saturated fatty acids over their unsaturated counterparts in this vegetable oil. Taking these results into account, it is noted that this oil is distinguished by a chemical profile of fatty acids that lies between that of crude palm vegetable oil and that of crude coconut vegetable oil.

The transmethylation reaction using homogeneous base catalysis resulted in the formation of FAME, with a yield

exceedingly at least one-fifth of the mass of the crude vegetable oil used as raw material.

Overall, the results presented in the current study on the CVO from *I. muricata* seeds suggest that it could be used as a frying oil if it is not toxic to humans. Unless otherwise specified, the oil could be used in cosmetics, pharmaceuticals, bio-lubricants, or biodiesel production, at least. In order to guarantee the availability of the seed oil of this plant for applications in the industrial scales, suitable growing techniques of the raw material must be developed.

For better operationalization of the results, further studies are possible to ensure better utilization of the oil from this plant. These include research focusing mainly on the toxicity of the oil, its antioxidant capacity, and its resistance to oxidation. The complete characterization of the biodiesel produced and its effects on a diesel engine also appear to be essential for evaluating the energy properties of the biodiesel produced. The influence of seed maturity on the lipid profile of the oil extracted from the seeds seems very relevant in determining when to harvest the seeds in order to improve the quality of their vegetable oil. Other parts of the seed may also contain biomolecules of therapeutic interest that could be explored for optimal use of this seed.

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Conflicts of Interest

The authors declare no conflicts of interest regarding this manuscript publication.

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