

# Research Article

# The Impact of the Extraction Method on *Allanblackia floribunda* Butter's Physicochemical Properties as a Possible Pharmaceutical Excipient

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The extraction method of edible *Allanblackia floribunda* seed butter is crucial for preserving its constituents. The objective of the present study was to investigate the effects the extraction methods have on the physicochemical properties of *A. floribunda* butter regarding its potential use as a pharmaceutical excipient. Butter obtained from different extraction methods (including solvent/ hexane, cold press, and traditional/hot water) was analyzed for its physicochemical properties such as yield, melting point, relative density, refractive index, moisture content, pH, acid value, saponification value, percentage of free fatty acids, and iodine value as well as beneficial elements and pathogenic microorganisms. All physicochemical parameters were within the standard limits for edible and industrial oils/butter (Codex Stan 210-1999) and were free from pathogenic microorganisms. However, the pH value of all extracts was higher than that of olive oil. The moisture content (calcium, sodium, magnesium, potassium, and iron) than the cold press and hot water extracts. Extraction with hexane gave the highest yield. The identified fatty acids in all extracts are palmitic and stearic (saturated fatty acids), oleic, linoleic, and linolenic (polyunsaturated fatty acids) acids. Based on the physicochemical analysis, *A. floribunda* seed butter is edible and has the potential as a pharmaceutical excipient in drug delivery.

# 1. Introduction

Allanblackia floribunda (fam., Clusiaceae), a flowering plant commonly found in the rainforest of Africa, is traditionally used to treat hypertension, dysentery, toothache, stomachache, asthma, bronchitis, cough, and urethral discharge [1, 2]. The plant grows naturally in parts of Central Africa, East Africa, and West Africa. Countries including Nigeria, Ghana, the Democratic Republic of Congo, Uganda, and Tanzania have Allanblackia growing naturally in moist zones. In Ghana, it is found in Western, Central, Ashanti, Eastern, and parts of Brong Ahafo regions, where cocoa (*Theobroma cacao*) develops best [3]. The tree's fruits are not edible, but its seeds are a source of oil/butter. The oil from its seeds is a natural and potentially excellent substitute for other oils in domestic use and commercial production of food and nonfood products. It consists almost exclusively of triglycerides of stearic (45–58%) and oleic (40–51%) fatty acids, which are components that have always been part of the human diet [4].

A. floribunda oil is extracted from the dried seeds at the local and industrial levels. The butter (oil solidified at room temperature) is usually extracted locally using hot water. Commercially, expeller pressing and solvent extraction have been used to extract the oil. The modern process of vegetable oil is by chemical extraction, using solvent extracts such as hexane [5]. This produces higher oil yields and is quicker and less tedious. However, the vegetable oil industry needs some

suitable and environmentally friendly extraction methods due to the safety, toxicological, environmental, and potential health risks involved in using methods like hexane extraction [6]. Mechanized and semimechanized methods have also been used to extract the butter. A survey conducted by Jibreel et al. [7] in the Tamale Metropolis (Ghana) indicated that about 14% of people still preferred the traditional method to the modern or mechanized and semimechanized methods. The extraction process could affect the quality of the butter.

Recently, various novel extraction techniques such as pressurized hot water extraction (PHWE), supercritical fluid extraction (SFE), ultrasonication-assisted extraction (USE), and microwave-assisted extraction have been developed for the extraction of different compounds from natural sources [8–10]. Among these new methods, SFE has been extensively studied as an excellent alternative to conventional oil extraction techniques, including organic solvent extraction and hydrodistillation. Unfortunately, access to these newer techniques is limited and unavailable for this study.

This study, therefore, investigated the effect of using cold press (semimechanized), hot water (traditional), and hexane (chemical) extraction methods on the preservation of the quality of the oil (butter) extracted. Allanblackia seed oil has a high melting point, distinguishing it from alternatives such as palm oil [11]. It is solid at ambient temperature and is therefore used to manufacture soap. It is commercially valued in margarine production, requiring less chemical processing and refraction than palm oil. Recently, the international food industry has become interested in fat as a natural solid component for margarine and similar products [12]. These attributes of Allanblackia butter could make it a probable excipient for pharmaceutical use as it can be a good and cheap substitute for cocoa butter, which has been used in cosmetic and medicinal formulations. Even though some form of characterization has been done on the oil, this research provides an exhaustive characterization of butter as a potential pharmaceutical excipient. A pharmaceutical excipient must be inert, stable, and reproducible, have desired functionality, be cost-effective, and be free of disease-causing microorganisms [13].

#### 2. Methods

2.1. Sample Collection and Preparation. A. floribunda fruits were collected from the Council for Scientific and Industrial Research (CSIR) forest at Fumesua in the Ashanti Region of Ghana (coordinates: 6.715495, -1.525865, or 6°42′55.8″N 1°31′33.1″W). Matured fruits, without surface blemishes, were plucked. The seeds were manually removed from the matured fruit by maceration and dried for three days in the sun. The seeds were dehulled and sun-dried until they reached a consistent weight (Figure 1). The dehulled dry seeds were milled using the disc miller an2d kept at room temperature.

#### 2.2. Oil Extraction

*2.2.1. Solvent (Hexane) Extraction.* Solvent extraction was performed using hexane as the solvent in a Soxhlet apparatus by the method described by AOAC 1990.

A 20 g portion of *A. floribunda* seed powder was weighed into a muslin fabric and put in a thimble with a Soxhlet device. The condenser was firmly secured at the bottom of the extractor, with a round bottom flask containing 250 ml of hexane inserted at the end of the device. The entire apparatus was preheated in a water bath at 60°C. After the extraction, the surplus solvent in the oil was recycled by heating it at  $60^{\circ}$ C in a heating mantle. The oil yield was calculated using the following equation:

% oil yield = 
$$\frac{\text{weight in gram of extracted oil}}{\text{weight in gram of milled seed}}$$
. (1)

2.2.2. Cold Press Extraction. A mass (1.5 kg) of A. floribunda milled seeds was weighed into three different metal containers and placed in an oven for about an hour. The heated sample was placed in a muslin cloth and put into the inlet of the screw press machine (IO 67, Bonagro screw press, China). By centrifugation, the oil was extracted from each sample through the outlet. The percentage oil yield was calculated using equation (1). The process was done in triplicate.

2.2.3. Traditional (Hot Water) Extraction. The milled seed (900 g) was roasted at 65°C for about four hours. Warm and cold water was added intermittently to make a paste. The paste was therefore transferred into boiling water and boiled while stirring periodically. The scum containing impurities was removed, and the released oil was skimmed off from the surface of the mixture using a calabash before being boiled to remove the residual moisture. The yield was calculated using equation (1).

2.3. Determination of the Melting Point of Butter. The Stuart melting point apparatus (SMP 10, United Kingdom) was used to determine the melting point of the extracted butter.

2.4. pH Determination of Butter. The pH electrode (Jenway 3510 bench pH/mV meter, UK) was calibrated using the appropriate buffer. The electrode was thoroughly rinsed with water and immersed into the hexane extract at room temperature. The pH meter indication was noted. The experiment was repeated twice, and the mean value was calculated. The above procedure was repeated for the hot water and cold press extracts.

2.5. Determination of Refractive Index of Butter. The *A. floribunda* butter (hexane extract) was melted and filtered to remove traces of moisture and impurities present in the butter. The Atago RX-5000 refractometer (Japan) was calibrated, and the temperature was set at 40°C. Two drops of filtered*A. floribunda* butter were put on the prism and covered, and the refractive index was read at 40°C. The test was carried out in triplicate. The procedure was repeated for the cold press and hot water extracts.



FIGURE 1: Pictures showing (a) A. floribunda tree, (b) matured A. floribunda fruits, and (c) dried A. floribunda seeds.

2.6. Determination of Relative Density of Oils/Butter. The A. floribunda butter (hexane extract) was melted at 40°C and filtered for relative density determination in a 10 ml Gay-Lussac bottle. The weight of a scrupulously cleaned and dried Gay-Lussac bottle was determined as a, followed by the weight of a dried Gay-Lussac bottle filled with distilled water (b). Finally, the weight of the dried Gay-Lussac bottle filled with the oil (hexane extract) was determined as c. The relative density was calculated using the following equation:

relative de nsity = 
$$\frac{\text{de nsity of the Allanblackia butter}}{\text{de nsity of water}} = \frac{c-a}{b-a}.$$
(2)

The same procedure was repeated for cold press and hot water extracts.

2.7. Determination of Moisture Content of Butter. The moisture content of A. floribunda butter was determined using the gravimetric method on unfiltered Allanblackia butter (hexane extract). A 10 g portion of Allanblackia butter was weighed into a dried and tarred evaporating dish. The extract was then dried at a temperature of  $105 \pm 1^{\circ}$ C for about 5 hours and cooled in a desiccator, and the weight was recorded. Drying and weighing at one-hour intervals were done until the variance between two consecutive weights corresponded to 0.25% w/w or less [14]. The determination was carried out in triplicate. The above procedure was repeated for hot water and cold press extracts. The percentage moisture content was calculated using the following equation:

moisture content (%) = 
$$\frac{\text{loss in weight of the butter (g)}}{\text{initial weight of the butter (g)}} \times 100.$$
(3)

2.8. Determination of Acid Value of Extracted Butter. The acid value was determined using the British Pharmacopoeia method. In brief, 10 g of A. *floribunda* butter (hexane extract) was weighed into a 250 ml Erlenmeyer flask. The butter was dissolved in a 50 ml combination of diethyl ether and ethanol (96%) that had previously been neutralized with phenolphthalein solution R1 using 0.1 M KOH. The mixture was titrated with 0.1 M KOH using 0.5 ml of phenolphthalein solution R1 as an indicator until the solution

remained slightly pink after shaking for at least 15 seconds [11]. The same procedure was repeated for hot water and cold press extracts. The acid value was calculated using the following equation:

acid value (mg/g) = 
$$\frac{5.610 \times \text{volume of titrant (ml)}}{\text{quantity of the Allanblackia butter (g)}}$$
. (4)

The percentage of free fatty acids was further calculated using the following equation:

free fatty acid % = 
$$\frac{\text{acid value}}{1.99} \times 100.$$
 (5)

2.9. Determination of Saponification Value of Butter. The method specified in the British Pharmacopoeia was followed. The hexane-extracted butter was melted at 40°C and filtered. An aliquot (2 g) of the melted butter was accurately weighed into a 250 ml flask. 25 ml of 0.5 N alcoholic KOH was added to the butter and heated in a steam bath under an appropriate condenser. The content was frequently rotated while maintaining reflux for 30 minutes. With 0.5 N HCl and 1 ml of phenolphthalein as an indicator, the excess KOH was titrated. A blank determination was performed using the same amount of 0.5 N alcoholic KOH [11, 12]. The above procedure was repeated for hot water and cold press extracts. Equation (6) was used to compute the saponification value of the butter.

Saponification value = 
$$\frac{56.1(B-S)N}{W}$$
, (6)

where B = volume of standard HCl required for blank (ml), S = volume of standard HCl required for the sample (ml), N = normality of standard HCl, and W = weight (g) of *Allanblackia* butter taken for the test.

2.10. Determination of Iodine Value of Butter. The procedure by Aberimah et al. [15] was adopted with slight modification. In a 250 ml conical flask, 0.25 g of the hexane extract was weighed and dissolved in 10 ml chloroform. 30 ml Hanus solution was added, and the container was sealed with parafilm. The mixture was left to stand in the dark for 30 minutes while constantly shaken. 10 ml of 15% KI solution was added and agitated. After that, 100 ml of distilled water was added. The solution was titrated with 0.1 N Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> until a yellow colour appeared. About 2-3 drops of the starch solution were added to the solution where the blue solution developed, and the titration was maintained until the blue colour vanished. The method was repeated, but this time without using a sample (blank). Equation (7) was used to compute the iodine value.

Iodine value = 
$$\frac{(B-S) \times N \text{ of } Na_2S_2O_3 \times 0.127g/meq}{\text{weight of sample}} \times 100,$$
(7)

where B = volume (ml) of Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> used for the blank and S = volume of Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> used for the sample.

The above procedure was repeated for the hot water and cold press extracts.

2.11. Microbial Quality Assessment of A. floribunda Butter. One gram (1 g) of the hexane extract of A. floribunda was dissolved with 3 ml of Tween 80 and topped up with peptone water to a final volume of 10 ml. About 0.5 ml of the prepared sample solution was transferred into different sterile Petri dishes. Twenty millilitres each of sterile molten nutrient agar (Aerobic count), Sabouraud dextrose agar (mold and yeast), mannitol salt agar (Staphylococcus aureus), MacConkey agar (Escherichia coli), cetrimide agar (Pseudomonas aeruginosa), and bismuth sulphite agar (Salmonella typhi) stabilized at 45°C for 15 minutes was poured into their respective Petri dishes and swirled to mix with extract solutions. The plates were allowed to stand for 30 minutes in the laminar floor hood to solidify and incubated at  $37^\circ\!\mathrm{C}$  for 48 hours for bacteria growth and at 25°C for 5 days for fungi growth [16]. The cell colony counter was then used to enumerate the number of viable cells. The respective agars were used as negative controls. The procedure was performed in triplicate and repeated for the cold press and hot water extracts.

2.12. Determination of Mineral Content in Butter. The mineral content in the butter (hexane) was determined using an atomic absorption spectrophotometer (Analytic Jena Nov AA-400P, Germany). One gram of butter was weighed into a vessel, and 10 ml of concentrated nitric acid was added. The mixture was heated at a temperature of  $150^{\circ}$ C to near dryness. It was then cooled, and 4 ml of a mixture of concentrated nitric acid was added. Concentrated H<sub>2</sub>SO<sub>4</sub> (5 ml) was added to the mixture and heated at 150°C until the solution became clear. For each mineral, a triplicate determination was done. The procedure was repeated for the cold press and hot water extracts.

#### 2.13. Determination of Fatty Acids Present in the Butter

2.13.1. *Methylation.* 0.6 g of extracted butter was placed in a round bottom flask. 18 ml of ethanolic NaOH solution was added to the sample and refluxed for 10 minutes under constant temperature. 30 ml of an ethanol-hydrochloric acid mixture (2:3) was added and refluxed for another

10 minutes, after which 30 ml of hexane was added to the resulting solution. Refluxing was continued for another 2-3 minutes. 50 ml of distilled water was added to the mixture to dissolve the inorganic salt and separated with a separatory funnel. The top layer was taken for GC-MS analysis.

2.13.2. Gas Chromatography-Mass Spectrophotometric (GC-MS) Analysis. The GC-MS analysis of the fatty acids in the samples was performed using Perkin-Elmer GC Clarus 580 Gas Chromatograph interfaced to a Perkin-Elmer Mass Spectrometer (Clarus SQ 8 S, USA) equipped with ZB-5HTMS (5% diphenyl/95% dimethylpolysiloxane) capillary column (30 mm, 0.25 m ID, 0.25 m DF). The oven temperature was set to 80°C (isothermal for 2 minutes), then increased from 15°C/min to 150°C, then 3°C/min to 250°C, and held at 250°C for 4 minutes.

For the MS detection, an electron ionization system was operated in electron impact mode with an ionization energy of 70 eV. Helium gas (99.999%) was used as the carrier gas at a constant flow rate of 1.6 ml/min, with an injection volume of 1  $\mu$ l. The injector temperature was kept at 250°C, while the ion source temperature was kept at 220°C. Mass spectra were measured with a scanning interval of 0.5 s and fragment sizes ranging from 45 to 4500 da. The solvent delay ranged from 0 to 3 minutes, and the overall GC/MS run duration was 34.5 minutes. TurboMass version 6.1.0 was used to analyze the mass spectra and chromatograms. The National Institute of Standards and Technology (NIST) database, which contains over 62,000 patterns, was used to interpret the mass spectra.

#### 3. Results and Discussion

This study assessed the sensory and physicochemical properties of *A. floribunda* oil/butter sampled from Fumesua in the Ashanti Region of Ghana. Evaluation of physical parameters like colour, smell, refractive index, and relative density helped identify the butter. Some evaluated quality parameters included moisture content, microbial quality, acid value, iodine value, and saponification value.

3.1. Sensory Evaluation of Allanblackia floribunda Oil/Butter. The extracted A. floribunda oil/butter had no occasional variance in colour. The colour of the hexane and cold press extracts was whitish-yellow or pale yellow, while the hot water extract was yellow. The colour of the butter is a result of the presence of carotenoids in the butter, and the extraction method influenced the extent of the extraction of these pigments. Carotenoids are insoluble in water but soluble in organic solvents such as acetone, chloroform, ethyl acetate, ethyl ether, and tetrahydrofuran (THF) [17]. The light colour of the butter in the hexane extract indicates that the carotenoids are not readily soluble in hexane. The hot water extract's yellowish colour indicates more carotenoid extraction due to the boiling. Koa et al. [18] investigated the effect of water boiling on the total carotenoid content in vegetables and found that boiling increased the carotenoid content. The colour of the respective butter did not change throughout the assessment period.

There was consistency in the surface appearance of the *A. floribunda* butter samples. When the butter was rubbed between the fingers, it produced a smooth and soft feel, but upon continuous rubbing, all samples melted, giving a smooth feel to the skin. All extracted butter gave a pleasant smell indicating no observable rancidity.

3.2. Physicochemical Characterization of A. floribunda Butter. Table 1 gives the physicochemical parameters of all three extracts of A. floribunda butter. These parameters were compared to a standard reference for vegetable oils (Codex Stan 210-1999) [19]. The physical constants of the oil extracted by the different methods, such as melting point, relative density, and refractive index, conformed to official standards. However, the pH of the extracts was relatively high compared to olive oil as edible vegetable oil. Most oils are weakly acidic, so the extracted oils are considered acceptable.

As expected, the moisture content was higher in the water and hexane extracts compared to the cold-pressed ones. However, this result is lower than the moisture content of *A. floribunda* seed oil reported by Sefah et al. [20], which ranged from 3.1% to 23%. High moisture content leads to the decomposition of *A. floribunda* butter through hydrolysis, enzyme action, and microbial growth [21]. There was no significant difference ( $P^{>}0.05$ ) between the extraction methods for the physical parameters assessed.

The hexane extraction gave the oil a higher yield than the hot water and cold-pressed methods (hexane <sup>></sup> hot water <sup>></sup> cold press). This result conforms to similar studies by Sefah et al. [20] and Alenyorege et al. [22], which investigated the yield of oil extracted by the cold press and hexane and hot water extraction, respectively. The hexane yield was, however, higher in this study (50.40%) than that of Sefah et al.'s study [20].

The chemical parameters that indicate oil quality include acid value, iodine value, saponification value, and percentage of free fatty acids. Except for saponification value, the others are indicators of the rancidity of fats and oils. The assessed parameters were within the recommended standards by the codex. However, the acid value and percentage of free fatty acids were higher in the cold press than in the hexane and hot water extracts. The saponification values were comparable with the different methods of extraction. The values were, however, higher than those of olive oil. This indicates that *A. floribunda* oil could find better applications in soapmaking and cosmetics. The oils are less rancid, and no pathogenic microorganisms were identified. The oils are, therefore, suitable for human consumption and can serve as a promising drug delivery excipient.

3.3. Microbial Quality Assessment of A. floribunda Butter. Oils for pharmaceutical applications must be devoid of the presence of pathogenic organisms such as *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Salmonella typhi*. Table 2 shows the absence of these pathogenic organisms. 3.4. Mineral Analysis of A. floribunda Butter Extracted by Hexane, Cold Press, and Hot Water. Calcium (Ca), sodium (Na), magnesium (Mg), potassium (K), and iron (Fe) were the major minerals present in A. floribunda butter. No heavy metals were present in the oil, making it suitable and healthy for consumption (Table 3). The presence of Ca, Mg, and K adds to the nutritional value of A. floribunda butter extracts. Ca, Mg, and K are responsible for bone formation and maintaining the body's blood. Ca helps the brain and other areas of the body to communicate properly and assists muscular movement and cardiovascular function.

On the other hand, Mg has been reported to help in essential enzyme system activation [23]. Thus, magnesium may be useful for oral lipid-based formulations with medicaments that do not complex with metals [24]. K-rich foods have been linked to various health advantages such as lowering blood pressure and water retention. Potassium also protects against stroke, osteoporosis, and kidney stones [25]. The mineral content was higher in the cold press and hexane extracts than in hot water.

3.5. Fatty Acid Analysis. The identified fatty acids in A. floribunda butter extracted by hexane, hot water, and cold press consisted of saturated fatty acids (C8-C16), monounsaturated fatty acids (C16:1 and C18:1), polyunsaturated fatty acids (C18:2, C18:3, C18:4, and C20:3) and other unclassified fatty acids (Table 4). The prominent fatty acids are palmitic and stearic (saturated fatty acids), oleic, linoleic, and linolenic (polyunsaturated fatty acids) acids. The hardness of butter depends on the proportion of stearic and oleic acids. The higher the proportion of stearic acid, the harder the butter [21]. The extracted A. floribunda butter had a desirable texture and hardness, making it ideal for use as a moisturizer and topical drug delivery agent. Hot water and hexane extracts had a higher composition of saturated fatty acids (52.01% and 50.59%, respectively) and were hence harder in texture than the cold press extract. The saturated fatty acids accounted for more than 50% of the total fatty acids present in the extracts except for the cold press extract, which contained about 20.24% of saturated fatty acids. Cold press extract contained a high amount of monounsaturated fatty acids.

The saponification degree is inversely related to the fatty acids' average molecular weight or chain length. Therefore, the shorter the average chain length ( $C_4$ – $C_{12}$ ), the higher the saponification value [25]. From the study, the average molecular weight of the compounds present in the three extracts was 316.3, 298.3, and 319.6 for hot water, hexane, and cold press, respectively (Table 4). Moreover, long-chain compounds like 1-heptatriacotanol ( $C_{37}H_{76}O$ ), stigmasterol ( $C_{29}H_{48}O_2$ ), and 2-(7-heptadecynyloxy)tetrahydro-2H-pyran ( $C_{22}H_{40}O_2$ ) could account for the relatively low saponification values for the hot water and cold press extracts as their quantity was low [26].

The cold press extract had a higher iodine value than the hexane and hot water extracts, but the value was less than that of the codex standard. The fatty acid analysis revealed that the cold press extract had a high degree of unsaturation compared to the hot water and hexane extracts. Oils with

Devoice chamical parameter	Extraction methods			Reference oil	Standard
Physicochemical parameter	Hexane	Hot water	Cold press	Olive oil	ranges for oils
Refractive index	$1.467\pm0.00$	$1.465\pm0.00$	$1.465\pm0.00$	$1.471\pm0.00$	1.468-1.471
Relative density	$0.90\pm0.00$	$0.89 \pm 0.00$	$0.86 \pm 0.00$	$0.92\pm0.00$	0.92-0.93
pH	$5.03\pm0.08$	$4.28\pm0.01$	$4.84\pm0.01$	$0.8 \pm 0.01$	0.8-2
Acid value (mg KOH/g)	$1.16\pm0.07$	$3.48\pm0.06$	$4.43\pm0.06$	$0.48 \pm 0.00$	0.6-4
Iodine value (100 g/g)	$49.88 \pm 0.69$	$49.04\pm0.76$	$49.96 \pm 0.29$	$16.37\pm0.18$	75-94
Moisture content (%)	$1.93 \pm 0.20$	$2.00\pm0.51$	$0.70 \pm 0.26$	$0.10 \pm 0.00$	0.10-0.12
Saponification value (mg KOH/g)	$208.51 \pm 8.45$	$204.67 \pm 2.86$	$192.05 \pm 1.56$	$192\pm1.60$	184-196
Free fatty acid (%)	$0.58\pm0.03$	$1.75 \pm 0.03$	$2.23 \pm 0.03$	$0.24 \pm 0.00$	0.30
Percentage yield (%)	$50.40\pm0.00$	$37.36 \pm 0.00$	$20.48\pm0.00$	_	_
Melting range (°C)	$4043\pm0.00$	$4143\pm0.00$	$4044\pm0.00$	_	_

TABLE 1: The physicochemical parameters assessed on *A. floribunda* butter extracted by hexane, cold press, and hot water compared to a reference standard (olive oil).

Data are presented as mean  $\pm$  SD.

 TABLE 2: Microbial assessment of A. floribunda butter.

Organism	Hot water	Hexane	Cold press	Control
*Total aerobic count (cfu/ml)	$1.2 \times 10^{2}$	$5.6 \times 10^{2}$	$1.05 \times 10^{3}$	No growth
*Mold and yeast (cfu/ml)	$3.0 \times 10^{1}$	$2.52 \times 10^{2}$	$2.92 \times 10^{3}$	No growth
E. coli	None detected	None detected	None detected	No growth
P. aeruginosa	None detected	None detected	None detected	No growth
S. aureus	None detected	None detected	None detected	No growth
S. typhi	None detected	None detected	None detected	No growth

\*BP 2020 specification for the total aerobic count is  $1.0 \times 10^5$  cfu/ml and that for mold and yeast is  $1.0 \times 10^4$  cfu/ml [16].

TABLE 3: Mineral content of the A. floribunda butter extracted by hexane, cold press, and hot water.

	Sample	es	
Minerals (mg/L)	Cold press	Hexane	Hot water
Ca	$1.56 \pm 0.03$	$1.31 \pm 0.02$	$0.45\pm0.09$
Na	$0.17 \pm 0.01$	$0.20 \pm 0.00$	$0.06 \pm 0.01$
Mg	$1.04 \pm 0.05$	$1.66 \pm 0.08$	$0.60 \pm 0.04$
K	$0.54 \pm 0.01$	$1.07 \pm 0.02$	$0.34 \pm 0.01$
Fe	$0.43 \pm 0.01$	$0.60 \pm 0.05$	$0.14\pm0.01$

Data are presented as mean  $\pm$  SD.

TABLE 4: Fatty acid	analysis of A.	floribunda butter	extracted by hexane,	, cold press, and hot water.

Commound name		Percentage composition	
Compound name	Hot water	Hexane	Cold press
Hexadecanoic acid, methyl ester	$1.189 \pm 0.010^{a}$	$1.602 \pm 0.020^{\rm b}$	$1.137 \pm 0.010^{\circ}$
Heptadecanoic acid, methyl ester		$0.083 \pm 0.010$	
9,12-Octadecadienoic acid, methyl ester	$0.090\pm0.010$		
n-Hexadecanoic acid			$0.053\pm0.010$
9-Octadecenoic acid, methyl ester, (E)		$42.852 \pm 0.020^{\rm d}$	$35.280 \pm 0.020^{e}$
Linolenic acid, methyl ester	$37.94 \pm 0.010$		
9,12-Octadecadienoic acid, methyl ester, (E, E)			$0.086\pm0.020$
Methyl stearate	$40.337 \pm 0.020^{\rm f}$	$41.063 \pm 0.010^{\rm g}$	
Oleic acid	$6.919 \pm 0.010^{ m h}$	$5.602 \pm 0.010^{i}$	
Methyl elaidate			$35.401 \pm 0.020$
Octadecanoic acid	$10.375 \pm 0.020^{j}$	$7.626 \pm 0.010^{ m k}$	$18.970 \pm 0.020^{ m l}$
9-Octadecenoic acid, (E)			$7.913 \pm 0.020$
cis-11-Eicosenoic acid, methyl ester		$0.071 \pm 0.010$	
Eicosanoic acid, methyl ester	$0.112 \pm 0.010^{\rm m}$	$0.096 \pm 0.010^{\rm n}$	$0.083 \pm 0.020^{\circ}$
1-Heptatriacotanol	$0.257 \pm 0.010^{\rm p}$		$0.053 \pm 0.010^{ m q}$
Cyclopropaneoctanoic acid, 2-octyl-, methyl ester		$0.071 \pm 0.010$	
Campesterol	$0.537 \pm 0.030$		

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TABLE 4: Continued.

Compound name		Percentage composition	
Compound name	Hot water	Hexane	Cold press
2-(7-Heptadecynyloxy)tetrahydro-2H-			$0.055\pm0.010$
Octadecanoic acid, 9,10-dihydroxy-methyl ester		$0.049\pm0.020$	
Stigmasterol	$0.606\pm0.010$		

Data are presented as mean  $\pm$  SD. Different alphabets in rows represent statistical significance after using either Student's two-tailed test or one-way ANOVA (p < 0.0001).

lower iodine values are less susceptible to oxidative rancidity. Due to the lower iodine values of the three extracts, *A. floribunda* butter will not be susceptible to oxidative rancidity, an ideal property for lipid pharmaceutical excipients. Thus, the butter extracted from *A. floribunda* by any of the three methods is ideal as a drug delivery excipient for formulations such as suppositories, ointments, creams, emulsions, solid-lipid nanoparticles, and liposomes.

## 4. Conclusion

*A. floribunda* butter was successfully extracted using three different extraction methods. The hexane extraction method gave the highest percentage yield, followed by the hot water and cold press methods. The physicochemical properties and the identified fatty acids of *A. floribunda* butter indicated that the butter is edible and suitable as a pharmaceutical excipient with no disease-causing microorganisms. All extracts contained essential minerals such as Ca, K, Mg, and Fe which were found to be high in hexane and cold press extracts.

#### **Data Availability**

The data used to support the findings of this study are included within the article and also available from the corresponding author upon request.

### **Conflicts of Interest**

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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