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Physico-chemical, GC-MS analysis and cold saponification of canary melon (*Cucumis melo*) seed oil

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ABSTRACT

The physicochemical analysis revealed that the hexane extract of Canary melon seed oil has acid value, iodine value, peroxide value, saponification value, relative density and refractive index of 0.35±0.01 mgKOH/g, 135.6±0.07 gI_2/100g, 1.80.00± 0.01 meq H_2O_2, 233.62±0.01 mgKOH/g, 0.82±0.01and 1.44±0.00 respectively. The percentage yield was 50.42±0.01%. The colour of the oil was light cream. The following fatty acids were identified form the GC-MS analysis; Palmitic acid, Stearic acid, 11-Octadecenoic acid,-5-Octadecenoic acid, Oleic acid, Octadecenoic acid, n-Hexadecanoic acid, Ricinoleic acid and Docosanoic acid (Behenic acid). The pH, foam ability (cm^3), total fatty matter, total alkali and percentage chloride of the Canary melon oil soap were 11.03± 0.02, 75.13±0.15 (cm^3), 36.66 ± 0.02 %, 0.92 ± 0.02% and 0.53 ± 0.15 % respectively. The texture was soft and the colour was lighter cream. The soap was slightly soluble in water. The results indicated that the hexane extract of the Canary melon seed oil has potential for pharmaceutical and cosmetic industries.

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1. INTRODUCTION

Canary melon is a bright-yellow melon with a white inner flesh. Melons generally belong to the cucurbit family (Cucurbitaceae), The scientific name for many melons is *Cucumis melo* (abbreviated; *C. melo*). A ripe Canary melon is nearly as sweet as other melons. There was no any literature report on cold saponification of Canary melon Seed Oil. This research work was aimed at quality evaluation, fatty acids identification using GC-MS and preparation of soap from hexane extract of the seed oil and potential applications in pharmaceutical and cosmetic industries.

2. MATERIALS AND METHODS

2.1. Sample collection and preparation

The Canary melon seed was procured from commercial producers at Jega town of Kebbi State, Nigeria. The dried seeds were crushed into powder using mortar and pestle and were stored in a plastic container for oil extraction.

2. 2 Oil extraction procedure

The hexane extract was obtained by complete extraction using the Soxhlet extractor (GG-17, SHUNIU). The 50 g of each powdered kernel sample was put into a porous thimble and placed in a Soxhlet extractor, using 150 cm^3 of n-hexane (with boiling point of 40- 60 °C) as extracting solvent for 6 hours repeatedly until required quantity was obtained. The oil was obtained after evaporation using Water bath at 70 ^oC to remove the excess solvent from the extracted oil. The oil was then stored in refrigerator for subsequent physicochemical analysis.

2.3. Percentage yield

The oil which was recovered by complete distilling of most of the solvent on a heating mantle was transferred to a beaker. The beaker

was then placed over water bath for complete evaporation of solvent for about 2 hours and volume of the oil was recorded and expressed as oil content (%) in line with literature report [1].

2.5. Determination of relative density

This was performed according to literature report [2].The 10ml of the oil was measured in a pre-weighed measuring cylinder. The weight of the cylinder and oil was measured; the weight of the oil was then obtained by subtracting the weight of the cylinder from the weight of the oil and cylinder. The density of the oil was obtained using equation below.

Density of oil = $\frac{W1-W\circ}{V\circ}$

Where *W1* = weight of empty measuring cylinder + oil.
Wo = weight of measuring cylinder, *Vo* = volume of oil used.

2.6. Physico-chemical analysis

The physico-chemical analysis of the Canary melon seed oil was carried out using the methods previously reported [3], [4], [5].

2.7. GC-MS analysis

The analysis of the fatty acids in the Canary melon seed oil sample was done at National Institute of Chemical Technology (NARICT), Zaria, Nigeria, a Shimadzu QP2010 plus series gas chromatography coupled with Shimadzu QP2010 plus mass spectroscopy detector (GC-MS) system was used. The temperature programmed was set up from 70°C to 280°C. Helium gas was used as carrier gas. The injection volume was 2 µL with injection temperature of 250 ^{o}C and a column flow of 1.80 mL/min for the GC. For the mass spectroscopy ACQ mode scanner with scan range of 30-700 amu at the speed of 1478 was used. The mass spectra were compared with the NIST05 mass spectral library [6].

2.8. Preparation and analysis of the canary melon seed oil soap

Saponification procedure. As reported in literature [7] 200 grams of sodium hydroxide pellets was dissolved in 1000cm3 volumetric flask and the volume made to the mark with distilled water. The required quantity of alkaline solution was mixed with Canary melon seed oil (ratio 1:1 v/v). The oil was warmed gently and poured into the beaker followed by the alkali solution to form an intimate mix and then stirred frequently for 7 minutes using stirring rod until reaction reached equilibrium. The saponification mixture was then poured into mould and allowed to dry (cure) for 24hours.

2.9. Determination of total fatty matter (TFM)

The TFM was determined by petroleum spirit extraction. Soap (5g) was dissolved in 50ml warm water and transferred to a separating funnel. Three to four drops of methyl orange indicator were added, followed by 4N sulphuric acid until the indicator colour changed from orange to pink. Petroleum spirit (10 ml) was added and the separating funnel shaken vigorously for 30s. The solution was then allowed to stand for a few minutes until the fatty acid liberated soap formed a clear layer on top. The soap was skimmed off, washed with distilled water and dried to constant weight in an oven at 60°C [8].

% TFM = $\frac{A-X}{W}$ x 100
Where *A*= weight of wax + oil, *X*= weight of wax, *W*= weight of soap.

2.10. Determination of total alkali

The total alkali was determined by titrating excess acid contained in the aqueous phase with standard volumetric NaOH solution. Procedure reported in [9] was modified and used. One grams of finished soap was weighed and 5ml of ethanol was added to it. 0.5 milliliters of 1N H_2SO_4 solution was added to the mixture and heated till the soap sample dissolved. Test solution was titrated against 1 N NaOH using phenolphthalein as indicator. The total alkali was obtained with the formula;

% Total alkali = $\frac{VA-VB}{W}$ ×3.1
Where *VA*= volume of acid *W*; *VB*= volume of base; *W*= weight of soap.

2.11. Determination of % Chloride

Five grams of finished soap was weighed and 50ml of distilled water added to it and heated to dissolve sample. The resulting solution was transferred into a 250ml volumetric flask and 10ml of 15 % Ca $(NO_3)_2$ was added to it and shaken to dissolve the soap. Distilled water was added to the on to the 150 ml mark. The solution was filtered and methyl red was added to 50 ml of the filtrate and was titrated against 10 N H_2SO_4 until a pink color was obtained. Resulting solution was titrated against 0.1 N $AgNO_3$ using $K_2Cr_2O_7$ as indicator, till a brick-red color was obtained [10].

% Cl = $\frac{\text{Titre volume X } 0.585}{\text{weight of soap}}$

2.12. pH determination

The pH was determined using pH meter (827PH Metronm Model). A 5g of the soap shavings were weighed and dissolved with distilled water in a 100ml volumetric flask. The electrode of the pH meter was inserted into the soap solution and the pH reading was recorded [11].

2.13. Foam ability test

A 2g of the soap was added to a 500 cm^3 measuring cylinder containing 100 cm^3 of distilled water. The mixture was shaken vigorously so as to generate foams. After shaking for some time, the cylinder was allowed to stand for 10 minutes. The height of the foam in the solution was measured and recorded [11].

Figure 1. (a) Canary melons; (b) Ground Canary melon seeds; (c) Hexane extract of Canary melon seed oil; (d) Canary melon oil soap.

3. RESULTS AND DISCUSSION

The oil contentmof the Canary melon seed was 50.42±0.01 % lower than 56.30 ± 2.35% reported for *Terminalia Catappa L* "Congo-Brazzaville" seeds [12] higher than 38.74% reported for shea nut [13] and 48.19 ± 0.20% reported for wild castor seed [14] almost similar to 50.28±0.01 % reported for onion (*Allium cepa* L.) seed oil [15] which indicated that the seed has high oil content. The colour of the oil was light cream.

Physico- chemical analysis results showed that the Canary melon Seed Oil has acid value of 0.35±0.01 mgKOH/g lower than the value obtained for *Hyptis spicigera* seed oil 2.5mgKOH/g [16]. Lower acid value signifies a maximum purity and made it suitable for soap production.

Iodine value of 135.60±0.07 gI_2/100g was obtained having lower value than iodine value (mg/100g) of 152.3, reported for wild *Corchorusolitorius* seed oil [17]and higher than Iodine value of 50.50 ± 8.023 I2/100g reported for *Jatropha curcas* L. seed oil [18] recommended for cosmetics and medicinal purposes.

Table 1: Physicochemical properties of Canary melon Seed Oil*

Parameters	Values
Oil yield (%)	50.42±0.01
Colour	Light cream
Acid value mgKOH/g	0.35±0.01
Iodine value gI_2/100g	135.6±0.07
Peroxide value meq H_2O_2	1.80± 0.00
Saponification value mgKOH/g	233.62±0.01
Relative density (g/cm3)	0.82±0.01
Refractive index	1.44±0.00

* Values are expressed as mean and ± standard deviation of triplicate determinations

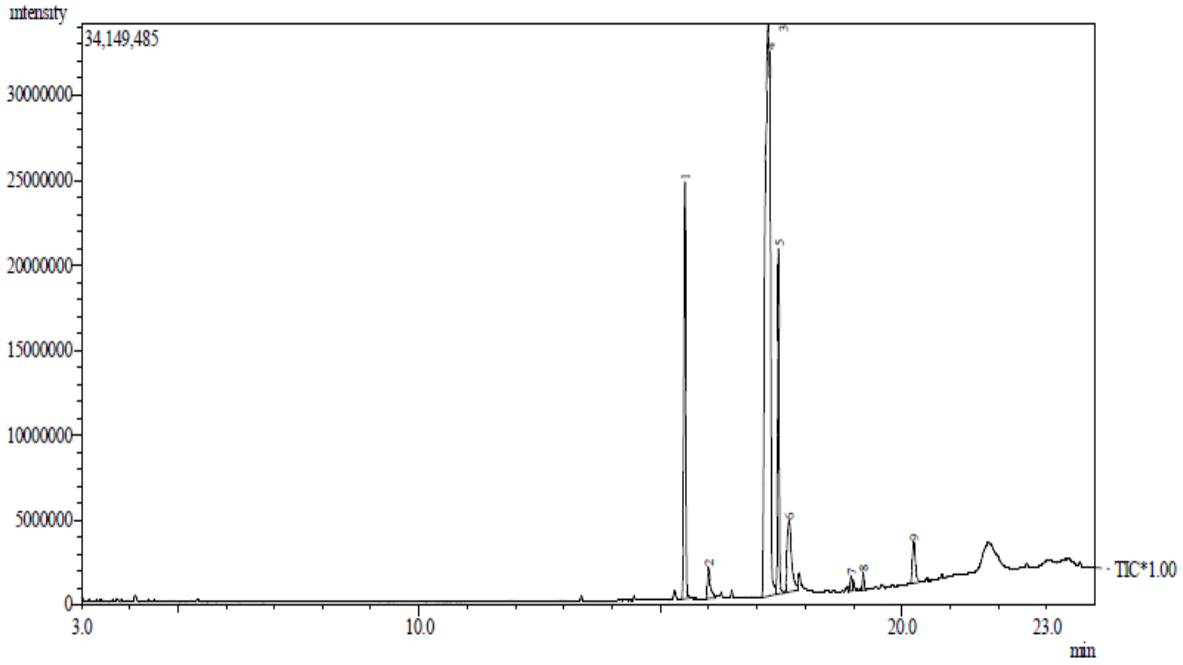
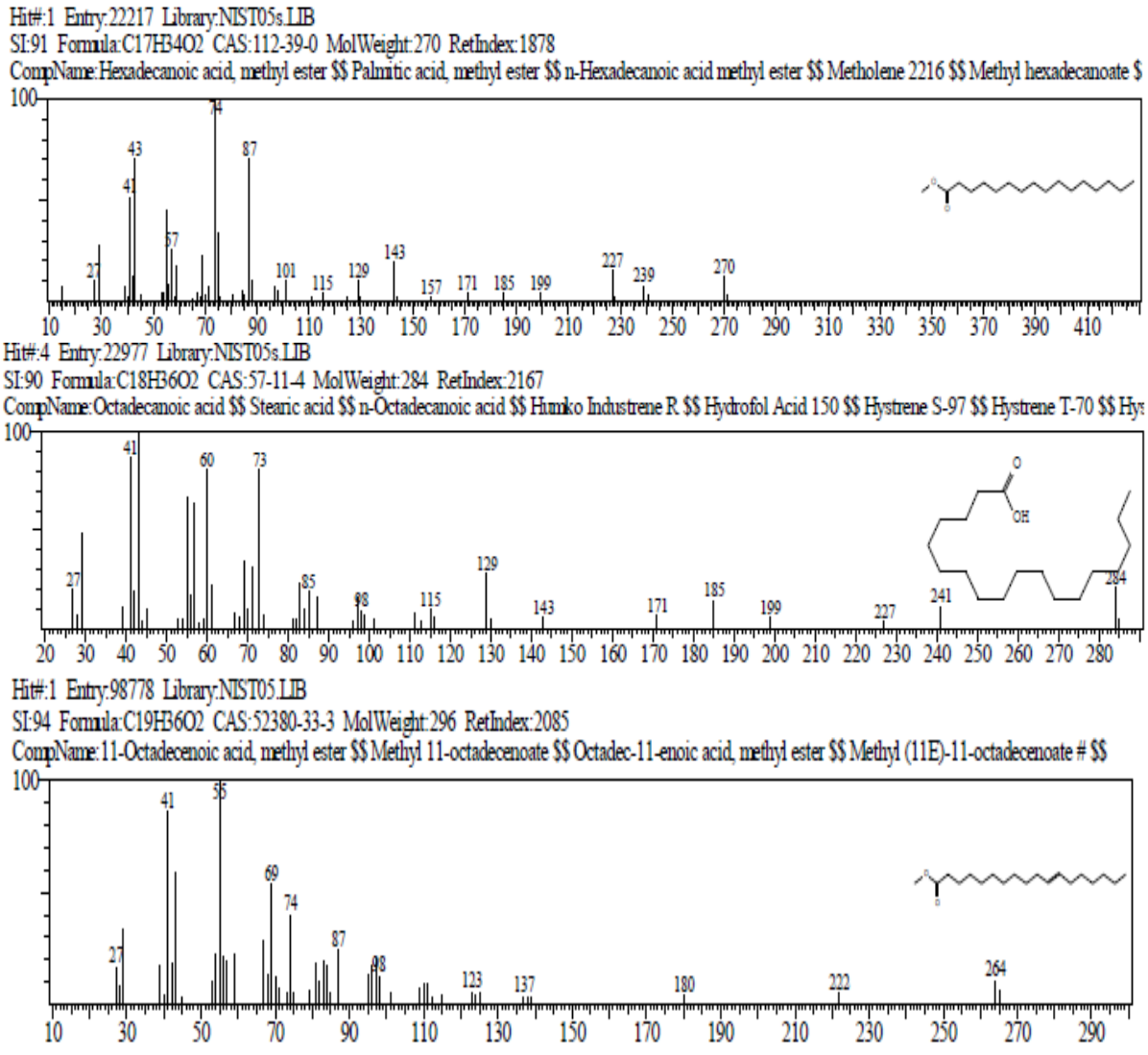


Figure 2. Typical GC-MS total ionic chromatogram (TIC) of Canary Seed Oil



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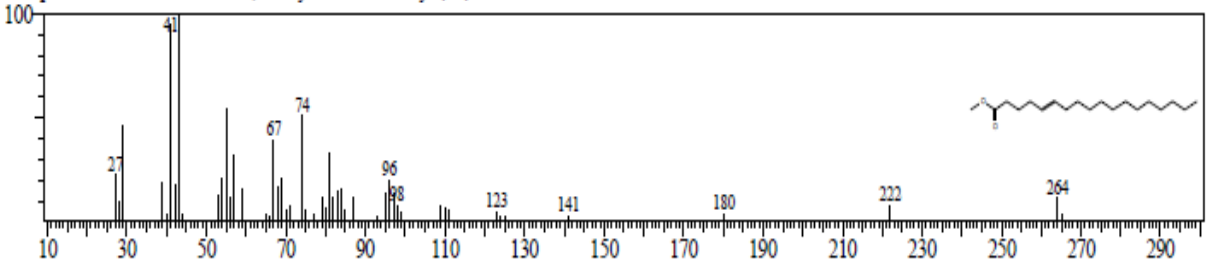

Hit#:4 Entry:22869 Library:NIST05s.LIB
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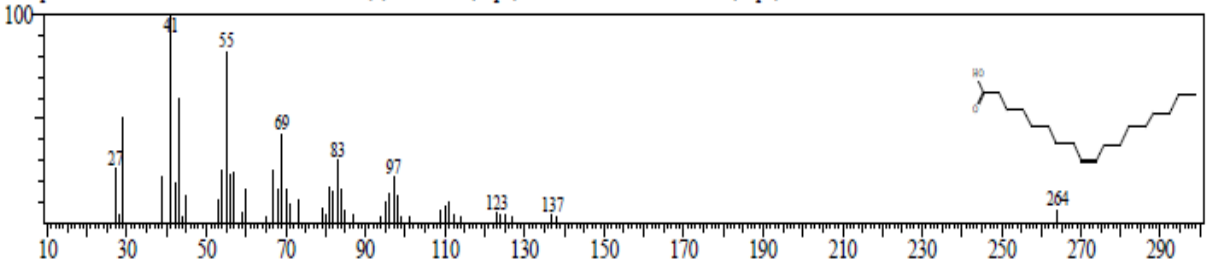

Hit#:5 Entry:23567 Library:NIST05s.LIB
SI:89 Formula:C19H36O2 CAS:2777-58-4 MolWeight:296 RetIndex:2085
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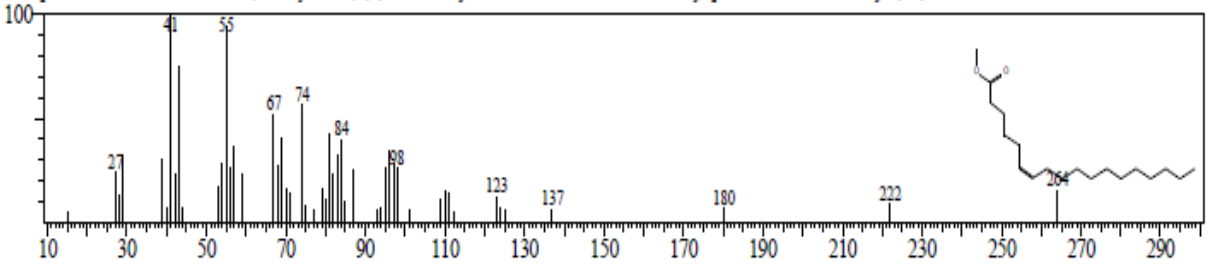
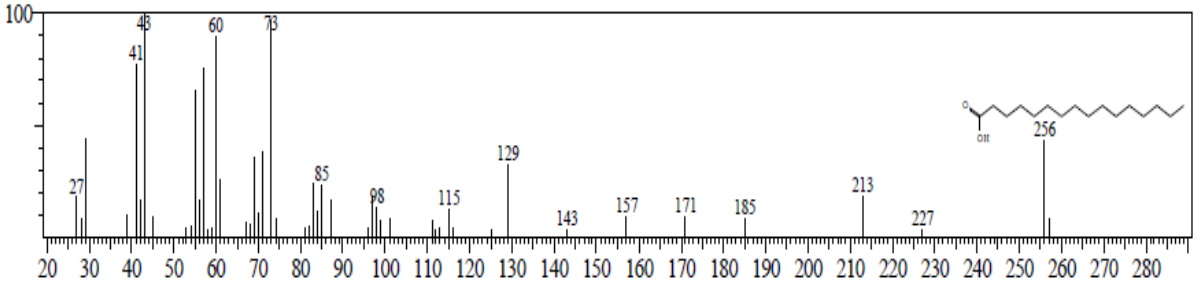


Hit#:5 Entry:21331 Library:NIST05s.LIB
SI:89 Formula:C16H32O2 CAS:57-10-3 MolWeight:256 RetIndex:1968
CompName:n-Hexadecanoic acid $$ Hexadecanoic acid $$ n-Hexadecoic acid $$ Palmitic acid $$ Pentadecanecarboxylic acid $$ 1-Pentadecanecarboxylic acid



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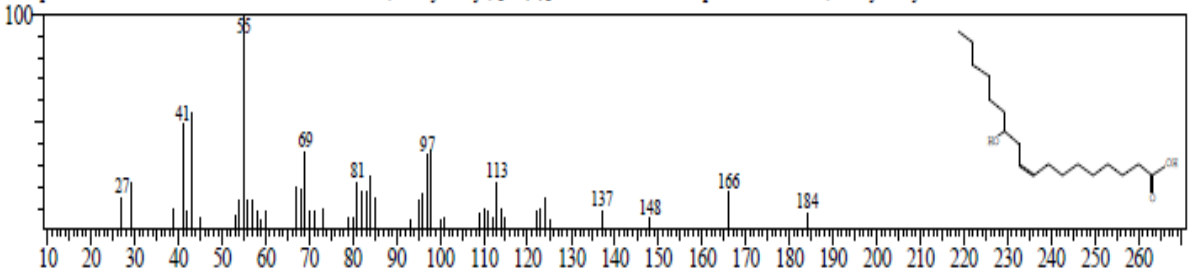
SI:80 Formula:C18H34O3 CAS:141-22-0 MolWeight:298 RetIndex:2337
CompName:Ricinoleic acid $$ 9-Octadecenoic acid, 12-hydroxy-, [R-(Z)]- $$ l'acide ricinoleique $$ Oleic acid, 12-hydroxy- $$ Ricinic acid $$ Ricinolic acid $



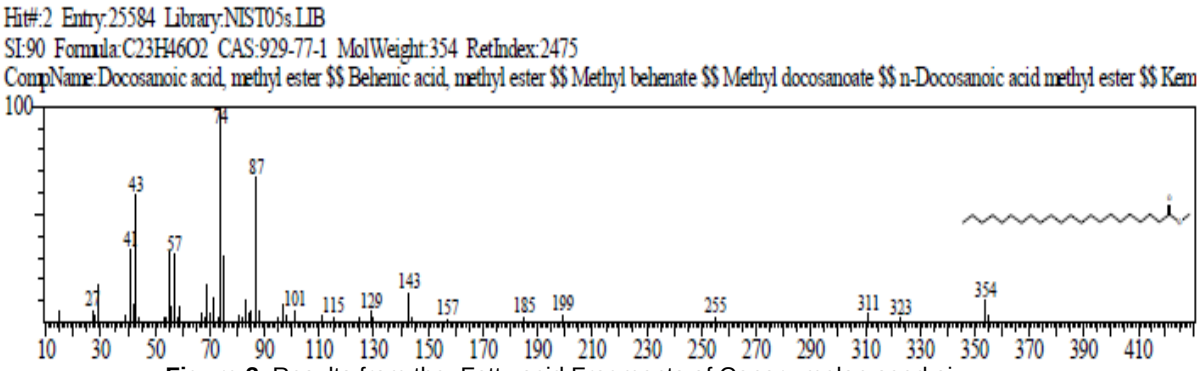


Figure 3. Results from the Fatty acid Fragments of Canary melon seed oi

Table 2. Major fatty acids derived from hexane extract of Canary melon L. seed oil

S/N	Name of fatty acid	MF	MW	RI	SI% to T.C.
1.	Palmitic acid	$C_{17}H_{34}O_2$	270	1878	91
2.	Stearic acid	$C_{18}H_{36}O_2$	284	2167	90
3.	11-Octadecenoic acid	$C_{19}H_{36}O_2$	296	2085	94
4.	Octadecenoic acid	$C_{19}H_{36}O_2$	296	2085	90
5.	Oleic acid	$C_{18}H_{34}O_2$	282	2175	90
6.	Octadecenoic acid	$C_{18}H_{34}O_2$	296	2085	90
7.	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	1968	89
8.	Ricinoleic acid	$C_{18}H_{34}O_3$	298	2337	80
9.	Docosanoic acid	$C_{23}H_{46}O_2$	354	2475	90

Note: S/N = Serial number, M.F.=Molecular formula, M.W. = Molecular weight, RI= Retention index SI% = Similarity index, T.C. = Target compound.

Table 3. Physicochemical characteristics of the Canary melon seed oil soap*

Parameters	**Values/Observation**
pH	11.03± 0.02
Foam ability (cm^3)	75.13±0.15
Solubility in water	Slightly soluble
Texture	Soft
Color	Lighter cream
%Total fatty matter	36.66 ± 0.02
%Total alkali	0.92 ± 0.02
% Chloride	0.53 ± 0.15

* Values are expressed as mean ± standard deviation of triplicate determinations

The peroxide value was found to be 1.80± 0.01 meq H_2O_2 which is lower than 37.79± 0.02 reported for *Moringa oliefera* Lam seed oil [19] Higher eroxide value indicate deterioration or not of seed oils. Fresh oils have values less than 10 mEq kg-1 values between 20 and 40 mEq kg-1 results to rancid taste [20]. Saponification value of 233.62±0.01(mgKOH/g) was obtained which is lower than that of *Elaeis guineensis* seed oil 246.60 mg KOH/g [21] but higher than that of African pear oil 143.76 mgKOH/g [22] recommended for soap making.

The relative density of 0.82±0.01 was obtained, the value is lower than 0.97±0.03 reported for *Dennettia tripetala* Fruit Oil [23], 0.94 reported for *Cucumis melo* Linn Seed oil [24] and 0.93± 0.00 reported for *Blighia sapida* fruit oil [25]. The refractive index was 1.44±0.00 lower than refractive indexes of Gundelia seed oil at 25 ºC and 40 ºC which were 1.4715 and 1.4675, respectively [26]. Refractive index measures the purity of oil. The results indicate that the oils are of high purity [27].

The following fatty acids were identified form the GC-MS analysis; Palmitic acid which is one of the the most abundant and widespread natural saturated acid, present in plants, animals, and microorganisms was identified [28] It is among the fatty acids that is used in concentration in cosmetics [29] Stearic acid a saturated fatty acid with an 18-carbon chain and the IUPAC name octadecanoic acid was found, Stearic acid is mainly used in the production of detergents, soaps, and cosmetics such as shampoos and shaving cream products. Soaps are not made directly from stearic acid, but indirectly by saponification of triglycerides consisting of stearic acid esters. Esters of stearic acid with ethylene glycol, glycol stearate, and glycol

distearate are used to produce a pearly effect in shampoos, soaps, and other cosmetic products. They are added to the product in molten form and allowed to crystallize under controlled conditions. Detergents are obtained from amides and quaternary alkylammonium derivatives of stearic acid. Surfactants, cosmetics and personal hygiene products are in fact prospects of stearic acid [30]. 11-Octadecenoic acid, reported for the first time in a seed oil of *Asclepias syriaca* L. (common milkweed) was also found. 11-Octadecenoic acid differs from most natural unsaturated fatty acids in having the double bond at the seventh carbon atom from the methyl end of the chain [31]. 5-Octadecenoic acid a highly unusual fatty acid and a novel type of fatty acid modification in reported in schistosomes (Brouwers et al., 1998) was identified. *Oleic acid was identified, o*n the basis of available data from studies using animals and humans, it was concluded that Oleic acid among others are safe in present practices of use and concentration in cosmetics [29]. Octadecenoic acid was also iedetified which is any of several unsaturated fatty acids $C_{18}H_{34}O_2$ of which some (as oleic acid and vaccenic acid) occur in fats and oils. n-Hexadecanoic acid was identified. Anti-Inflammatory Property of n–Hexadecanoic Acid: Structural Evidence and Kinetic Assessment was reported [32] *Ricinoleic acid was identified* The hydroxylated fatty acid product ricinoleic acid has a wide-range of useful industrial properties. These include uses in lubricants, hydraulic fluids, surfactants, cosmetics, and nylon production. The hydroxyl group of ricinoleic acid may also increase the lubricity of fatty acid methyl esters in biodiesel applications [33]. *Docosanoic acid (Behenic acid).* A major component of Ben oil which is extracted from the seeds of the *Moringa oleifera* tree [34] was also identified. Behenic acid is often used to give hair conditioners and moisturizers their smoothing properties. Also used as anti-foam in the manufacturing of detergents [35].

The pH of the soap was found to be 11.03± 0.02 higher than pH of 8.6 reported for *Jatropha* oil soap [36] and 10.4 ± 0.04 reported for neem oil soap [37]. Even though the pH was higher than the skin friendly pH, it can be regulated by superfatting. The Foam ability (cm^3) was 75.13±0.15 higher than 2.5cm reported for animal fat soap [38]and 4.2 cm reported for sheanut oil soap [39].Foam ability has little to do with cleansing ability [40] it is of interesting importance to the consumer and is therefore considered a parameter in evaluating soaps and detergents. The percentage Total fatty matter was 36.66 ± 0.02%, the value is lower than 63.75 ± 0.07% reported for neem oil soap[37] and 60.20% reported for Jatropha oil soap [36] Total Fatty Matter (TFM) tells how much fat substance the soap has. The quality of a soap can be indicated with the amount of fatty matters. Soap with 70% to 80% fatty matters is considered best one in quality. If the TFM lowers then it would not give you the softness that is expected out of a soap along with the other qualities of a beauty soap [19]. The percentage total alkali was 0.92 ± 0.02% higher than 0.76% reported for *Jatropha* oil soap [36] NaOH and NaCl contribute to the total alkali of sheanut fat soap The percentage chloride was 0.53 ± 0.15% lower value than 1.15 ± 0.02 reported for neem oil soap [37] and 1.26±0.01 reported for sheanut fat soap [13].

4. CONCLUSION

From the results of the physicochemical, GC/MS and Canary melon oil soap produced, it can be concluded that seed oil has potential in the production of soap, perfumery and pharmaceuticals.

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AUTHOR CONTRIBUTIONS

AAW designed the study, wrote the protocol, and wrote the first draft of the manuscript and the literature searches. AAW, FS, HSA managed the analyses of the study. Interpretation and write up of the manuscript were done by authors AAW and AA. All authors read and approved the final manuscript.

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