

Chemical Composition GC/MS analysis, antioxidant and Antimicrobial Activity of Hibiscus, Baobab and Buckhorn Seed Oil's

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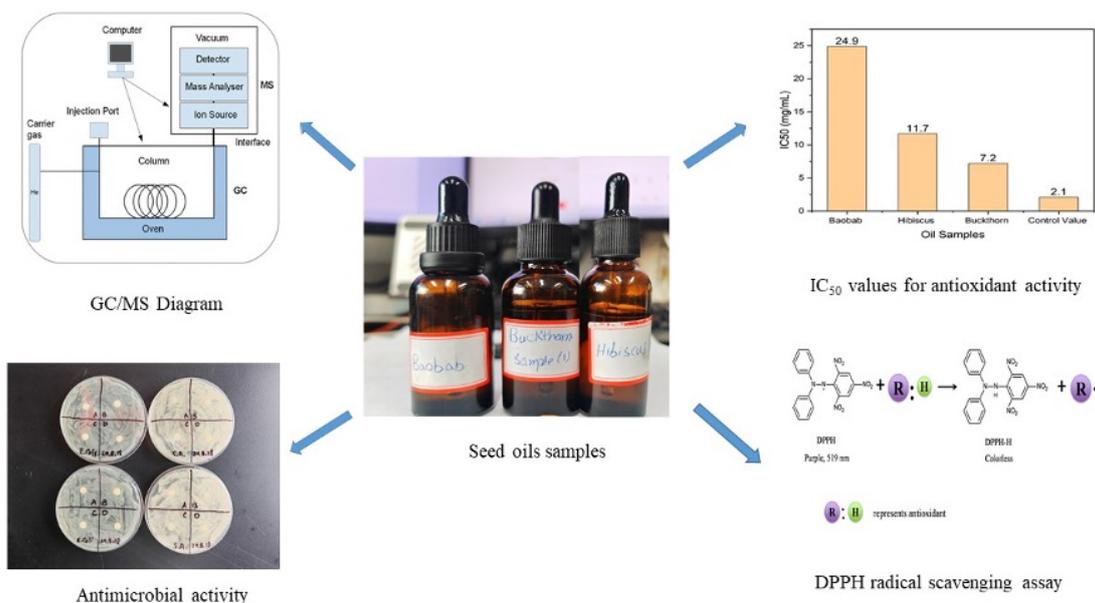
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GRAPHICAL ABSTRACT



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ABSTRACT

This study explores the chemical composition and bioactive potential of Baobab, Hibiscus, and Buckthorn seed oils, focusing on their antioxidant and antimicrobial activities for cosmetic and functional food applications. GC/MS analysis revealed distinct profiles: Hibiscus oil was rich in 10(*E*),12(*Z*)-conjugated linoleic acid (32.87%) and cis-13-Octadecenoic acid (31.54%), Baobab oil contained high levels of cis-Vaccenic acid (24.59%) and n-Hexadecenoic acid (17.57%), and Buckthorn oil was characterized by significant concentrations of gamma-Sitosterol (12.78%) and gamma-Tocopherol (11.5%). The antioxidant capacity, measured using the DPPH assay, showed Buckthorn oil as the most active (IC₅₀ = 7.2 mg/mL), followed by Hibiscus (IC₅₀ = 11.7 mg/mL) and Baobab (IC₅₀ = 24.9 mg/mL), highlighting their potential as natural antioxidants, with Buckthorn oil demonstrating superior

efficacy. Antimicrobial testing against *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans* revealed limited activity. Baobab and Buckthorn oils showed weak inhibition against *S. aureus* (9.3 mm and 9.0 mm, respectively). In contrast, Hibiscus oil had no effect, and none of the oils inhibited *E. coli* or *C. albicans*. One-way ANOVA confirmed no significant differences in inhibition zones against *S. aureus* ($p = 0.204$), indicating that the antimicrobial effects are weak and not statistically meaningful. These findings demonstrate that while the tested oils have strong antioxidant potential, particularly Buckthorn oil, their antimicrobial activity in crude form is limited. Future studies incorporating MIC determination and formulation optimization could enhance their practical applications in cosmetics, pharmaceuticals, and functional foods.

1. INTRODUCTION

Hibiscus (*Hibiscus sabdariffa*), baobab (*Adansonia digitata*), and buckthorn (*Hippophae rhamnoides*) seed oils have attracted increasing scientific interest due to their diverse chemical compositions and reported biological activities, particularly antioxidant and antibacterial properties. These oils are rich in bioactive constituents, including fatty acids, sterols, tocopherols, and other phytochemicals, which support their potential use in food, cosmetic, and pharmaceutical applications. GC/MS has been widely employed to characterize the chemical profiles of plant-derived oils, providing detailed insights into the compounds responsible for their functional properties.

Previous studies have individually reported the chemical composition of hibiscus, baobab, and buckthorn seed oils. Hibiscus seed oil has been shown to contain n-hexadecanoic acid, cis-13-octadecenoic acid, γ -sitosterol, and vitamin E, compounds associated with antioxidant and antimicrobial activities [1, 2]. Baobab seed oil is characterized by a high content of unsaturated fatty acids, particularly linoleic and oleic acids, along with phytosterols and tocopherols that contribute to its antioxidant and potential therapeutic effects [3, 4]. Buckthorn seed oil is distinguished by its unique combination of essential fatty acids, phytosterols, tocopherols, and phenolic compounds, which are linked to its strong antioxidant activity and reported antimicrobial effects [5, 6].

The antioxidant properties of these oils are primarily attributed to their ability to scavenge free radicals and reduce oxidative damage, a function largely associated with phenolic compounds, tocopherols, and unsaturated fatty acids. Hibiscus seed oil has been reported to exhibit notable antioxidant activity, contributing to the mitigation of oxidative stress and the prevention of oxidative-related disorders [7, 8]. Similarly, baobab seed oil has demonstrated significant free radical scavenging capacity, supporting its application in nutraceutical and functional food products [9, 10]. Buckthorn seed oil has demonstrated strong antioxidant activity, making it well-suited for use as a natural antioxidant in food preservation and cosmetic formulations [11, 12].

In addition to antioxidant effects, several studies have reported antibacterial activity for these oils. Hibiscus seed oil has been shown to inhibit the growth of certain pathogenic microorganisms, including *Staphylococcus aureus* and *Escherichia coli* [13]. Baobab seed oil exhibits antibacterial activity, though its effectiveness varies with the tested microorganism and extraction conditions [14]. Buckthorn seed oil has been reported to possess antimicrobial activity against a range of gram-positive and gram-negative bacteria, as well as fungal strains, attributed to its fatty acids and sterol content [15, 16].

Accordingly, the present study applies a comparative design in which the three oils were examined under the same GC/MS conditions and biological assays. This approach allows meaningful comparison of their chemical composition and bioactivity, while supporting clearer insight into composition-function relationships. Integrating chemical profiling with antioxidant and antimicrobial evaluation, the study builds on existing literature. It offers a coherent assessment of the potential use of these oils in food, cosmetic, and pharmaceutical applications.

2. EXPERIMENTAL METHODS

2.1. Plant material

Hibiscus, baobab, and buckthorn seeds were purchased from a local market in Omdurman, Khartoum State, Central Sudan, in March 2023, and were identified at the Department of Phytochemistry and Taxonomy (National Research Centre, Khartoum).

2.2. Extraction of oil's

2.2.1. Hibiscus seeds oil

Hibiscus seeds were first cleaned, air-dried, and finely ground to increase the surface area for oil recovery. A total of 300 g of crushed seeds was subjected to cold-press extraction at temperatures below 50 °C to minimize thermal degradation of heat-sensitive bioactive compounds. The obtained oil was filtered to remove residual solids, yielding approximately 50 mL, which was subsequently stored in a dark, cool environment until analysis.

Cold pressing was selected for hibiscus seeds to preserve thermolabile constituents such as unsaturated fatty acids, tocopherols, and phenolic compounds, which are known to contribute to antioxidant activity. Although this method generally yields lower oil yields than solvent-based extraction, it is widely regarded as suitable for maintaining the nutritional and bioactive qualities of seed oils intended for food and cosmetic applications [17].

2.2.2. Baobab and buckthorn seed oil

Baobab and buckthorn seeds were thoroughly cleaned and dried to reduce moisture content, then ground into a fine powder to enhance solvent penetration. The powdered seeds (450 g for baobab and 600 g for buckthorn) were subjected to solvent extraction using n-hexane at a solid-to-solvent ratio of 1:3 (w/v). The extraction was carried out for 24 h under continuous stirring to maximize oil recovery. Following extraction, the mixtures were filtered to remove solid residues, and the solvent was removed using a rotary evaporator at 40–50 °C under reduced pressure to prevent oxidative or thermal degradation of the oils. Approximately 50 mL of oil from each seed type was recovered and stored in amber bottles at 4 °C until further analysis [18].

Solvent extraction was employed for baobab and buckthorn seeds due to their lower oil release efficiency under mechanical pressing, as reported in previous studies. This technique enables more thorough lipid recovery, particularly from dense or fibrous seed matrices, and ensures sufficient oil yield for comprehensive chemical and biological analyses. The use of different extraction techniques is therefore based on seed matrix characteristics and extraction efficiency considerations, and this methodological variation has been taken into account when interpreting compositional and bioactivity results.

2.3 GC/MS method: Chemical Composition Analysis

The chemical composition of the oil samples was analyzed using an Agilent 7890B-5977A GC/MS system with an HP-5ms column (30 m × 250 µm × 0.25 µm), operating at temperatures between 0°C and 350°C. Helium served as the carrier gas at a flow rate of 1 mL/min. The temperature treatment commenced at 50°C (maintained for 3 minutes), increased by 15°C/min to 150°C, then by 5°C/min to 180°C, and finally by 8°C/min to 325°C, where it was sustained for 10 minutes. The injection port was heated to 280°C, and 1 µL of each oil sample was injected in splitless mode. We performed mass spectrometry in electron ionization mode (70 eV) at 230°C, with a solvent delay of 3.5 minutes. Data acquisition was performed over a m/z range of 35–600 [19]. Compounds were identified by comparing their mass spectra with those in the NIST 17 mass spectral library. Identification was based solely on library matching; retention indices and authentic reference standards were not used in this study. The relative composition (%) was calculated by normalizing the peak areas so that the total identified compounds accounted for 100% of the oil composition. GC/MS analysis was performed in duplicate to ensure analytical reproducibility.

2.4 DPPH Radical Scavenging Assay

The DPPH radical scavenging assay was used to test the antioxidant activity of Baobab, Hibiscus, and Buckthorn oils. We followed Vinha's 2024 [20] description, albeit with some modifications. Scientists used this experiment to let test substances interact with a DPPH-stable free radical at 300 µM for 30 minutes at 37 °C. We solubilized the samples in ethanol, using ascorbic acid

as the standard control. We quantified the reduction in absorbance at 517 nm using a multiplate reader spectrophotometer after incubation. We determined the percentage of DPPH blocked by comparing the absorbance of the test samples to that of the ascorbic acid control. We determined the IC₅₀ values as the oil concentration required to achieve 50% inhibition of the DPPH radical. Antioxidant activity was evaluated in triplicate.

2.5 Antimicrobial assay

This test evaluated the efficacy of essential oils against *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 25923, and *Candida albicans*. It used the disc diffusion method. Sterile filter paper discs measuring 6 mm in diameter were prepared, each containing 10 μ L of the essential oil. A negative control disc containing 10 μ L of normal saline and a positive control disc with levofloxacin-sensitive tablets (5 μ g) were incorporated. It says that inhibition zones are divided into five groups based on their sensitivity: >20 mm means "severe sensitivity," 10-15 mm means "high sensitivity," 10–14 mm means "medium sensitivity," <10 mm means "low sensitivity," and 0 mm means "drug resistance." The bacterial and fungal strains were cultivated in nutrient broth until the optical density at 600 nm (OD₆₀₀) reached 0.5, corresponding to approximately 10⁸ CFU/mL [21]. Subsequently, 100 μ L of the bacterial and fungal suspensions were uniformly distributed on nutrient agar plates, left to rest for 10 minutes, and the produced discs were positioned on the agar surface. Following incubation, the widths of the inhibition zones were measured in millimeters to evaluate the antibacterial activity of the essential oils against each strain [22]. Antimicrobial activity assays were performed in triplicate.

3. RESULTS AND DISCUSSIONS

3.1 Chemical Composition Analysis GC/MS

3.1.1 Chemical Composition Analysis for hibiscus oil

GC/MS analysis of Hibiscus seed oil revealed 10 compounds, accounting for 100% of the total identified fraction. The oil was predominantly composed of 10(*E*), 12(*Z*)-conjugated linoleic acid (32.87%) and *cis*-13-octadecenoic acid (31.54%), both of which are well recognized for their antioxidant, anti-inflammatory, and skin-conditioning properties. These fatty acids play a crucial role in protecting skin cells against oxidative stress and improving skin barrier function.

Other major constituents included *n*-hexadecanoic acid (palmitic acid) (12.56%), 9-octadecenoic acid (*Z*-), 2-hydroxy-3-[(1-oxohexadecyl) oxy] propyl ester (10.11%), and oleic acid (2.96%). These components are known to enhance emolliency, skin softness, and moisturizing performance, making the oil suitable for cosmetic applications [23]. Minor bioactive compounds, such as γ -sitosterol, vitamin E, and various triterpenoids, further enhance the oil's biological functionality, contributing to its antioxidant capacity and potential therapeutic value in cosmetic and dermatological formulations [24].

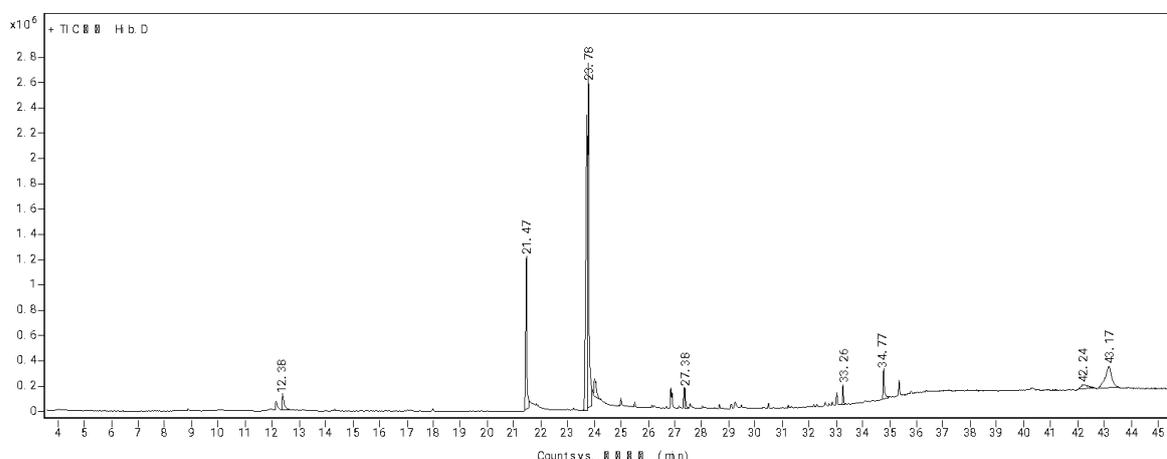


Figure 1. GC/MS chromatogram of Hibiscus oil

TABLE I. Chemical constituents of Hibiscus oil

No.	Retention time	Compound	Molecular Formula	M.W g/mol	Relative composition (%)	Area (%)
1	12.38	2,4-Decadienal (<i>E,E</i>)	C ₁₀ H ₁₆ O	152.23	2.34	7.13
2	21.47	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	12.56	38.21
3	23.72	10(<i>E</i>),12(<i>Z</i>)-Conjugated linoleic acid	C ₁₈ H ₃₂ O ₂	280.45	32.87	100
4	23.78	cis-13-Octadecenoic acid	C ₁₈ H ₃₄ O ₂	282.46	31.54	95.95
5	24.01	Oleic Acid	C ₁₈ H ₃₄ O ₂	284.48	2.96	8.99
6	27.38	9-Octadecenoic acid (<i>Z</i>)-oxiranylmethylester	C ₂₁ H ₃₈ O ₃	338.53	1.5	4.57
7	33.26	Vitamin E	C ₂₉ H ₅₀ O ₂	430.71	1.26	3.83
8	34.77	Gamma Sitosterol	C ₂₉ H ₅₀ O	414.71	2.63	8.01
9	42.24	9,19-Cyclolanost-24-en-3-ol, (3.β)	C ₃₀ H ₅₀ O	426.72	2.22	6.76
10	43.17	9-Octadecenoic acid (<i>Z</i>)-, 2-hydroxy-3-[(1-oxohexadecyl)oxy]propyl ester	C ₃₇ H ₇₀ O ₅	594.95	10.11	30.75

3.1.2 Chemical Composition Analysis for Baobab oil

GC/MS profiling of Baobab seed oil demonstrated a rich composition of bioactive compounds, with *cis*-vaccenic acid (24.59%) identified as the predominant constituent. This monounsaturated fatty acid has been associated with anti-inflammatory effects and cardiovascular health benefits, and these effects also translate into protective roles in skin health.

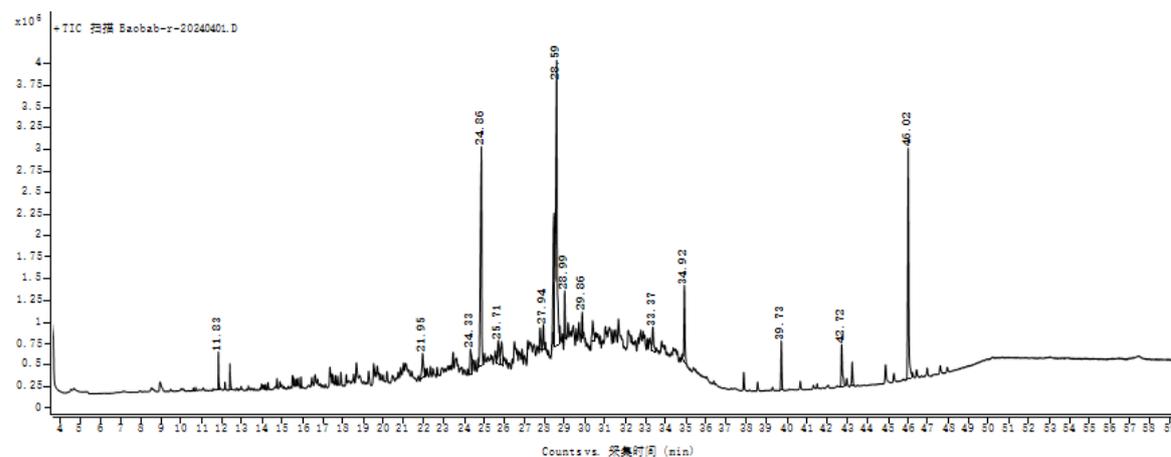


Figure 2. GC/MS chromatogram of Baobab oil

n-Hexadecanoic acid (palmitic acid) (17.57%) was detected as a major saturated fatty acid, commonly employed in skincare formulations for its emollient and moisturizing characteristics. The oil also contained a notable proportion of γ -sitosterol (14.13%), a phytosterol known for its cholesterol-lowering activity and anti-inflammatory potential.

Furthermore, 9, 12-octadecadienoic acid (*Z, Z*) (linoleic acid) (9.34%) plays an essential role in maintaining skin barrier integrity and regulating lipid metabolism [25]. Octan-2-yl palmitate (4.43%) contributes to improved texture, spreadability, and formulation stability, while γ -tocopherol (3.11%) provides potent antioxidant protection against oxidative damage. Collectively, these constituents underscore the suitability of baobab oil for cosmetic and health-promoting applications [26].

TABLE II. Chemical constituents of Baobab oil

No.	Retention time	Compound	Molecular Formula	M.W g/mol	Relative composition (%)	Area (%)
1	11.83	Nonanoic acid	C ₉ H ₁₈ O ₂	158.24	1.33	5.4
2	12.4	2,4-Decadienal, (E,E)	C ₁₀ H ₁₆ O	152.23	1.03	4.2
3	21.95	Octadecane, 1-chloro	C ₁₈ H ₃₇ Cl	288.93	2.25	9.14
4	24.33	Eicosane, 10-methyl	C ₂₁ H ₄₄	296.58	2.82	11.47
5	24.86	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	17.57	71.45
6	25.71	1-Chloroeicosane	C ₂₀ H ₄₁ Cl	322.00	2.16	8.8
7	25.86	Heptadecane, 9-hexyl	C ₂₃ H ₄₈	338.67	2.69	10.93
8	27.76	Heneicosane	C ₂₁ H ₄₄	296.56	1.33	5.42
9	27.94	Heptadecane, 9-hexyl	C ₂₃ H ₄₈	338.67	1.63	6.64
10	28.46	9,12-Octadecadienoic acid (Z,Z)	C ₁₈ H ₃₂ O ₂	280.45	9.34	37.99
11	28.59	cis-Vaccenic acid	C ₁₈ H ₃₄ O ₂	282.46	24.59	100
12	28.99	Octadecanoic acid	C ₁₈ H ₃₆ O ₂	284.48	2.24	9.12
13	29.86	Pentacosane	C ₂₅ H ₅₂	352.67	1.27	5.17
14	30.38	Pentacosane	C ₂₅ H ₅₂	352.67	1.73	7.05
15	33.37	Octadecane, 3-ethyl-5-(2-ethylbutyl)	C ₂₆ H ₅₄	340.66	2.15	8.76
16	34.92	Octan-2-yl palmitate	C ₂₄ H ₄₈ O ₂	384.66	4.43	18.02
17	39.73	Squalene	C ₃₀ H ₅₀	410.72	2.68	10.88
18	42.72	Gamma Tocopherol	C ₂₈ H ₄₈ O ₂	416.68	3.11	12.64
19	43.24	Stigmasta-3,5-diene	C ₂₉ H ₄₈	398.70	1.51	6.13
20	46.02	Gamma Sitosterol	C ₂₉ H ₅₀ O	414.71	14.13	57.48

3.1.3 Chemical Composition Analysis for buckthorn oil

GC/MS analysis of Buckthorn oil identified twelve components contributing to its pronounced bioactive profile. The dominant compound was cis-vaccenic acid (20.39%), a monounsaturated fatty acid recognized for its cardioprotective and anti-inflammatory effects. 9, 12-octadecadienoic acid (Z, Z) (linoleic acid) (18.71%) was also present in substantial amounts, highlighting the oil's importance in skin regeneration and lipid metabolism.

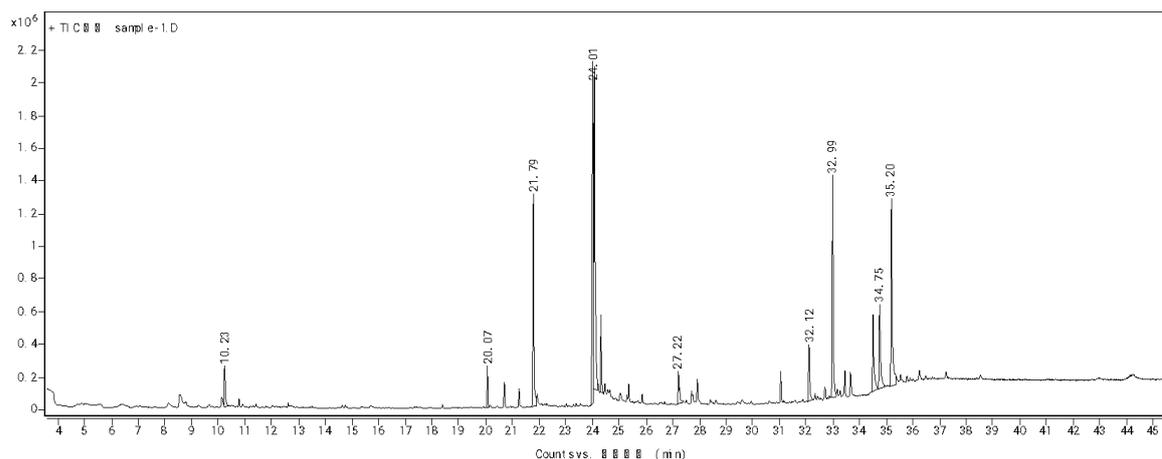


Figure 3. GC/MS chromatogram of buckthorn oil

The oil contained significant levels of γ -sitosterol (12.78%), a phytosterol associated with cholesterol-lowering and anti-inflammatory activity, and γ -tocopherol (11.50%), a powerful antioxidant that protects cells from oxidative stress. n-Hexadecanoic acid (palmitic acid) (11.45%) further contributes to the oil's emollient properties, enhancing skin softness and hydration.

Additional phytosterols such as stigmasterol (6.03%) and campesterol (5.82%) support anti-inflammatory activity and cholesterol regulation. δ -Tocopherol (3.41%) enhances antioxidant performance, while octadecanoic acid (stearic acid) (3.41%) provides formulation stability. Minor constituents, including nonanal, neophytadiene, and fatty acid esters, further contribute to the oil's fragrance, protective functions, and cosmetic appeal [27, 28].

TABLE III. Chemical constituents of Buckthorn oil

No.	Retention time	Compound	Molecular Formula	M.W g/mol	Relative composition (%)	Area (%)
1	10.23	Nonanal	C ₉ H ₁₈ O	142.24	2.57	12.58
2	20.07	Neophytadiene	C ₂₀ H ₃₈	278.48	1.67	8.17
3	21.79	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	11.45	56.13
4	24.01	9,12-Octadecadienoic acid (Z,Z)	C ₁₈ H ₃₂ O ₂	280.45	18.71	91.74
5	24.08	cis-Vaccenic acid	C ₁₈ H ₃₄ O ₂	282.46	20.39	100
6	24.32	Octadecanoic acid	C ₁₈ H ₃₆ O ₂	284.48	3.36	16.47
7	27.22	9,12-Octadecadienoic acid (Z,Z), 2-hydroxy-1-(hydroxymethyl)ethyl ester	C ₂₁ H ₃₈ O ₄	354.50	2.33	11.41
8	32.12	Delta Tocopherol	C ₂₇ H ₄₆ O ₂	402.65	3.41	16.73
9	32.99	Gamma Tocopherol	C ₂₈ H ₄₈ O ₂	416.68	11.5	56.41
10	34.51	Campesterol	C ₂₈ H ₄₈ O	400.68	5.82	28.54
11	34.75	Stigmasterol	C ₂₉ H ₄₈ O	412.69	6.03	29.56
12	35.2	Gamma Sitosterol	C ₂₉ H ₅₀ O	414.71	12.78	62.67

3.1.4. Chemical Composition and Industrial Applications of Three Oil Samples:

The GC/MS results revealed distinct chemical profiles across the three oil samples, each characterized by varying proportions of fatty acids, sterols, and other bioactive constituents with diverse industrial relevance. Sample 1, dominated by 10(*E*),12(*Z*)-conjugated linoleic acid (32.87%) and cis-13-octadecenoic acid (31.54%), is a rich source of unsaturated fatty acids. These compounds are associated with potential cardiovascular and metabolic health benefits, which may support their inclusion in functional food ingredients and nutraceutical applications [29].

Sample 2 exhibited comparatively high levels of cis-vaccenic acid (24.59%) and n-hexadecanoic acid (17.57%), both of which contribute to nutritional and skin-conditioning properties valued in personal care formulations. In contrast, Sample 3 contained particularly elevated amounts of γ -sitosterol (12.78%) and γ -tocopherol (11.50%), molecules well recognized for their antioxidant and anti-inflammatory activities [30, 31]. The presence of these sterols and tocopherols makes Samples 2 and 3 especially attractive for cosmetic applications, such as anti-aging, moisturizing, and skin barrier repair products.

Beyond cosmetic uses, the coexistence of conjugated linoleic acid and biologically active sterols in these oils suggests potential pharmaceutical relevance, particularly in anti-inflammatory and lipid-modulating formulations [32]. Additionally, their fatty acid profiles indicate suitability for industrial applications, including biodiesel synthesis, lubricant production, and surfactant manufacture, with Sample 1's elevated linoleic acid content offering advantages for oxidative stability and combustion performance in biodiesel [33].

Extraction methods influenced both composition and bioactivity: cold-pressing (used for hibiscus) better preserves unsaturated fatty acids and antioxidants, whereas solvent extraction (used for baobab and buckthorn) enhances oil yield but may slightly alter bioactive content. This factor should be considered when comparing functional properties and potential applications of the oils. Together, the three oils provide a complementary mix of fatty acids, antioxidants, and sterols, thereby maximizing their applicability across the food, cosmetic, pharmaceutical, and industrial sectors.

TABLE IV. Comparison of Chemical Composition in Three Oil Samples Based on GC/MS Analysis

Compound	Sample 1 Hibiscus	Sample 2 Baobab	Sample 3 Buckhorn	Note
10(<i>E</i>),12(<i>Z</i>)-Conjugated linoleic acid	32.87	-	-	Predominant compound in Sample 1
cis-13-Octadecenoic acid	31.54	-	-	High concentration only in Sample 1
n-Hexadecanoic acid	12.56	17.57	11.45	Present in all samples, higher in Sample 2
9-Octadecenoic acid (<i>Z</i>)-, 2-hydroxy-3-[(1-oxohexadecyl)oxy]propyl ester	10.11	-	-	Unique to Sample 1
Oleic acid	2.96	-	-	Detected only in Sample 1
Gamma Sitosterol	2.63	14.13	12.78	Significant in Samples 2&3
2,4-Decadienal, (<i>E,E</i> -)	2.34	-	-	Found only in Sample 1
9, 19-Cyclolanost-24-en-3-ol, (3.β.)	2.22	-	-	Detected only in Sample 1
9-Octadecenoic acid (<i>Z</i>)-oxiranylmethyl ester	1.50	-	-	Present exclusively in Sample 1
Vitamin E	1.26	-	-	Present only in Sample 1
cis-Vaccenic acid	-	24.59	20.39	Major compound in Samples 2 and 3
9,12-Octadecadienoic acid (<i>Z,Z</i> -)	-	9.34	18.71	Higher in Sample 3
Octan-2-yl palmitate	-	4.43	-	Detected only in Sample 2
Gamma Tocopherol	-	3.11	11.5	High in Sample 3
Stigmasterol	-	-	6.03	Present only in Sample 3
Campesterol	-	-	5.82	Unique to Sample 3

3.2 Antioxidant activity of oil samples (DPPH)

The antioxidant activities of Baobab, Hibiscus, and Buckthorn oils were assessed using the DPPH radical scavenging assay, with ascorbic acid ($IC_{50} = 2.1$ mg/mL) serving as a reference control. Among the three oils, Buckthorn oil demonstrated the strongest free radical scavenging capacity, with an IC_{50} value of 7.2 mg/mL, indicating high antioxidant effectiveness. Hibiscus oil followed with an IC_{50} of 11.7 mg/mL, reflecting moderate antioxidant potential. In contrast, Baobab oil exhibited the weakest activity, with an IC_{50} of 24.9 mg/mL, suggesting comparatively lower efficacy in neutralizing free radicals.

These results reveal a clear gradient in antioxidant potential among the oils, with Buckthorn oil showing the most pronounced activity, followed by Hibiscus and Baobab oils. This variation in antioxidant capacity emphasizes the differing suitability of each oil for potential incorporation into food, cosmetic, or pharmaceutical products where oxidative stability and radical scavenging are critical functional properties [34, 35].

TABLE V. DPPH antioxidant activity values

Oil Samples	IC_{50} (mg/mL)	Control Value (Ascorbic Acid)
Baobab	24.9 mg/mL	IC_{50} : 2.1 mg/mL
Hibiscus	11.7 mg/mL	IC_{50} : 2.1 mg/mL
Buckthorn	7.2 mg/mL	IC_{50} : 2.1 mg/mL

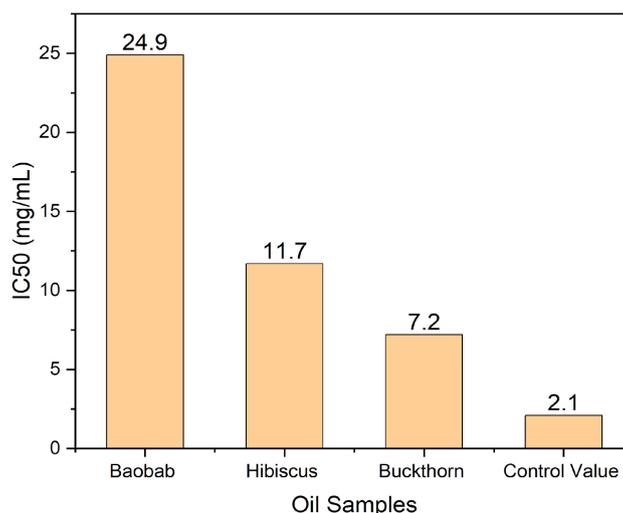


Figure 4. The IC₅₀ values for antioxidant activity in oil samples

3.3 Antimicrobial Activity

The antimicrobial effects of Baobab, Hibiscus, and Buckthorn seed oils were tested using the cup-plate agar diffusion method against three standard human pathogens: *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans* [36]. Baobab and Hibiscus oils showed only weak activity against *S. aureus*, with inhibition zones measuring 9.3 mm and 9.0 mm, respectively, which is below the commonly accepted range for moderate sensitivity (10–14 mm) [21]. Buckthorn oil showed no inhibition against *S. aureus*, indicating minimal antibacterial effect.

None of the oils were active against *E. coli* (gram-negative) or *C. albicans* (yeast), with inhibition zones of 0 mm, indicating a narrow antimicrobial spectrum. Overall, these results suggest that Baobab and Hibiscus oils exhibit only weak activity against Gram-positive bacteria, whereas Buckthorn oil is essentially inactive.

It should be emphasized that the antimicrobial effects observed are limited and unlikely to be clinically significant. The low activity may be due to poor diffusion of hydrophobic oil components in agar and to the polarity of bioactive compounds, which can limit their interaction with microbial cells. To better assess antimicrobial potential, future studies should determine minimum inhibitory concentrations (MICs) and explore formulation strategies such as combining oils or adjusting concentrations to enhance efficacy.

Statistical analysis using one-way ANOVA showed no significant differences in inhibition zones among Baobab, Hibiscus, and Buckthorn oils against *S. aureus* ($p = 0.204$), confirming that their antimicrobial activity is weak and not statistically meaningful under the tested conditions.

TABLE VI. Antimicrobial Activity of Oil Samples

Oil Samples	<i>S.aureus</i> Inhibition Zone (mm)	<i>E.coli</i> Inhibition Zone (mm)	<i>C. albicans</i> Inhibition Zone (mm)	Notes
Baobab	9.3 mm	0 mm	0 mm	Weak inhibition of <i>S. aureus</i>
Buckthorn	9.0 mm	0 mm	0 mm	Weak inhibition of <i>S. aureus</i>
Hibiscus	0 mm	0 mm	0 mm	No inhibition observed

4. CONCLUSIONS

This study provides a comprehensive evaluation of the chemical composition and bioactive properties of Baobab, Hibiscus, and Buckthorn seed oils, highlighting their distinct functional

potentials. GC/MS analysis revealed clear compositional differences among the oils. Hibiscus oil was characterized by a high content of unsaturated fatty acids, particularly 10(*E*), 12(*Z*)-conjugated linoleic acid and cis-13-octadecenoic acid, while Baobab oil was dominated by cis-vaccenic acid and n-hexadecanoic acid. In contrast, Buckthorn oil contained substantial levels of γ -sitosterol and γ -tocopherol, compounds known for their antioxidant and skin-protective properties.

The antioxidant activity, evaluated using the DPPH radical scavenging assay, demonstrated that Buckthorn oil exhibited the strongest activity ($IC_{50} = 7.2$ mg/mL), outperforming both Hibiscus and Baobab oils. Although its activity was lower than that of the reference standard (ascorbic acid, $IC_{50} = 2.1$ mg/mL), the results indicate a strong potential for Buckthorn oil as a natural antioxidant source, particularly in cosmetic and dermatological formulations.

In contrast, antimicrobial assays against *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans* revealed limited effectiveness. Baobab and Buckthorn oils exhibited weak inhibitory activity against *S. aureus*, with inhibition zones of 9.3 mm and 9.0 mm, respectively, while Hibiscus oil showed no detectable antibacterial effect. None of the tested oils demonstrated activity against *E. coli* or *C. albicans*. These findings underscore that, despite their strong antioxidant capacity, the oils possess limited antimicrobial potential in their crude form. The observed weak activity may be attributed to the poor diffusion of oil components in agar and the polarity of their bioactive compounds. It should not be interpreted as clinically significant. Future studies incorporating MIC testing or formulation optimization could help clarify and potentially enhance antimicrobial efficacy.

Overall, the results suggest that Baobab, Hibiscus, and Buckthorn seed oils are valuable sources of bioactive lipids with pronounced antioxidant properties but restricted antimicrobial activity. Future research should focus on optimizing extraction techniques, refining oil fractions, or developing synergistic formulations to enhance their biological efficacy and broaden their applicability in cosmetic, pharmaceutical, and functional product development.

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