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GC-MS and Microbial Analysis of Oil Extract from Gums of *Anacardium Occidentale* (Cashew Gum), *Khaya Senegelensis* (African Mahogany Gum), and *Terminalia Mantaly* (Umbrella Tree Gum)

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Abstract

Natural plant gums are valuable sources of bioactive compounds with pharmaceutical and industrial potential. This study investigated the chemical composition, physicochemical properties, and antimicrobial activities of oil extracts from the gums of *Anacardium occidentale* (cashew), *Khaya senegalensis* (African mahogany), and *Terminalia mantaly* (umbrella tree). Gum samples were collected, dried, pulverized, and extracted with ethanol using a Soxhlet apparatus. The oils were characterized by determining flash point and refractive index, while their chemical constituents were identified using Gas Chromatography-Mass Spectrometry (GC-MS). Antimicrobial activity was evaluated using the agar well diffusion method against *Staphylococcus aureus, Proteus spp., Saccharomyces cerevisiae*, and *Botryodiplodia theobromae*. Data were analyzed using one-way ANOVA at $p < 0.05$. Flash point values ranged from 120.52 °C (*K. senegalensis*) to 157.37 °C (*T. mantaly*), while refractive indices ranged from 1.4545 to 1.4591. GC-MS identified 20 to 24 compounds, including vanillic acid, n-hexadecanoic acid, oleic acid, squalene, and dl-α-tocopherol. *T. mantaly* exhibited the strongest antibacterial activity against S. aureus, producing a 30 mm inhibition zone at 100 mg/mL, exceeding all standard antibiotics tested. K. senegalensis showed broad-spectrum antibacterial and antifungal activity with inhibition zones up to 24 mm, whereas A. occidentale demonstrated moderate activity. Significant differences in antimicrobial efficacy were observed ($p < 0.05$). These findings indicate that *T. mantaly* and *K. senegalensis* gum oils are promising natural antimicrobial agents with potential applications in pharmaceutical, cosmeceutical, and nutraceutical formulations, warranting further cytotoxicity, minimum inhibitory concentration, and in vivo studies.

Keywords: *Anacardium Occidentale, Khaya senegalensis, Terminalia mantaly,* Microbial Analysis, Natural plant gums, Gum extract

1.0 Introduction

Plant gums are natural exudates secreted by trees and shrubs in response to mechanical injury or environmental stress. These biopolymers are complex carbohydrates primarily composed of polysaccharides, proteins, and minor lipid components (Phillips and Williams, 2021). The chemical structure of plant gums varies based on species and environmental factors, making them highly diverse in composition and functionality (Siddiqui *et al*., 2017). Due to their hydrophilic nature, they dissolve in water to form viscous solutions, making them valuable in various industries, including pharmaceuticals, food, cosmetics, and bio-based materials (Reddy *et al*., 2018). However, beyond their traditional uses, plant gums have been reported to contain trace amounts of oils, which may have unique industrial applications. The extraction and characterization of these oils have gained attention in recent years, with emphasis on their potential utilization in food, cosmetics, pharmaceuticals, and biofuel production (Ali *et al*., 2021).

The oil content in plant gums, though relatively low, can be an important source of bioactive lipids such as fatty acids, sterols, and triglycerides (Oladipo *et al*., 2020; Oyegoke *et al.,* 2023). These compounds contribute to the antioxidant, antimicrobial, and emulsifying properties of plant gums, thereby enhancing their industrial potential. The presence of such oils in gums obtained from species like *Anacardium occidentale* (cashew gum), *Khaya senegalensis* (African mahogany gum), and *Terminalia mantaly* (umbrella tree gum) has been of particular interest due to their bioactive compositions (Adebowale *et al*., 2021). Cashew gum, for instance, has been shown to contain essential fatty acids, including linoleic acid, oleic acid, and palmitic acid, which can be useful in cosmetics and pharmaceutical formulations (Ekebafe *et al*., 2021). Similarly, African mahogany gum and umbrella tree gum contain lipid components that may serve as potential sources of natural emulsifiers, thickeners, and stabilizers in various industries (Mishra *et al.*, 2018).

The process of oil extraction from plant gums typically involves solvent extraction methods such as Soxhlet extraction or supercritical fluid extraction to isolate the lipid components (Ugbogu *et al.*, 2021; Anaun and Ogundele, 2021). Once extracted, the chemical composition of the oil is analyzed using advanced analytical techniques, with Gas Chromatography-Mass Spectrometry (GC-MS) being one of the most widely used. GC-MS allows for the identification of volatile and semi-volatile compounds by separating them through gas chromatography and then detecting their molecular structures using mass spectrometry (Singh *et al.*, 2019, Ogundele *et al.,* 2019). The technique provides detailed insights into the fatty acid profiles, sterols, and other lipid components present in plant gum oils. For instance, the GC-MS analysis of cashew gum oil has identified high levels of polyunsaturated fatty acids, which can be beneficial for human health and industrial applications (Maqbool *et al*., 2019).

The industrial significance of oils extracted from plant gums is vast. In the pharmaceutical industry, these oils can be utilized as carriers for drug formulations, stabilizers, and bioactive ingredients in nutraceuticals (Patel *et al*., 2019). In the food industry, plant gum oils have the potential to act as natural emulsifiers and preservatives, improving the stability and shelf life of food products (Mahmood *et al.*, 2022). The cosmetic industry benefits from plant gum oils due to their moisturizing and antioxidant properties, making them valuable in skincare formulations, lotions, and hair care products (Sharma *et al.*, 2020). Additionally, with the increasing demand for renewable biofuels and biodegradable lubricants, plant gum oils have been explored as potential alternatives to synthetic oils, contributing to sustainability and environmental conservation (Ekebafe *et al*., 2021).

This study aims to investigate the chemical composition and industrial potential of oil extracts from *Anacardium occidentale* (cashew gum), *Khaya senegalensis* (African mahogany gum), and *Terminalia mantaly* (umbrella tree gum) using GC-MS analysis. By identifying the lipid components present in these plant gums, this research will provide valuable insights into their potential applications across multiple industries. The findings will contribute to the growing body of knowledge on plant-based bioresources, offering sustainable and eco-friendly alternatives to synthetic materials.

2.0 Materials and Methods

2.1 Research Design

This study adopted a quantitative experimental research design to systematically explore the chemical and biological properties of oils extracted from the gums of *Anacardium occidentale* (Cashew Gum), *Khaya senegalensis* (African Mahogany Gum), and *Terminalia mantaly* (Umbrella Tree Gum). Laboratory-based experiments were conducted to extract, analyze, and evaluate the physicochemical parameters, chemical constituents, and antimicrobial efficacy of the oils.

The design was structured to allow for both analytical and biological evaluations using standard laboratory protocols. Extraction was carried out using the Soxhlet method with ethanol as the solvent. Physicochemical assessments included flash point and refractive index measurements. Chemical composition profiling was done using Gas Chromatography-Mass Spectrometry (GC-MS). Finally, the antimicrobial properties of the oils were investigated through agar well diffusion assays against selected pathogenic bacteria and fungi.

2.2 Sample Collection

Gum exudates were collected from mature, healthy, and disease-free trees of the three target species. The selection of collection sites was guided by ecological suitability and species availability. *Anacardium occidentale* gums were obtained from established cashew-growing zones; *Khaya senegalensis* was collected from savannah and semi-arid regions; while *Terminalia mantaly* was harvested from tropical and subtropical forest zones.

A standardized tapping technique was employed to stimulate gum exudation. This involved making shallow incisions on the bark using sterilized blades. The exudates were allowed to harden naturally over a few days and were subsequently harvested using sterile spatulas. To avoid contamination and degradation, samples were immediately placed into sterile, airtight containers. Environmental conditions at the point of collection such as ambient temperature, humidity, and soil type were recorded, as these factors are known to influence gum yield and chemical composition.

Post-harvest, the gum samples were sun-dried for several days under hygienic conditions to reduce moisture content. Once completely dried, the gums were crushed using a mechanical grinder, milled into a fine powder, and passed through a mesh sieve ($\leq$1 mm pore size). The fine powders were stored in airtight polyethylene containers until ready for oil extraction.

2.3 Extraction of Oil from Gums

2.3.1 Preparation of Gum Powders

Each powdered gum sample was subjected to a cleaning and drying process to eliminate impurities and prevent thermal degradation. Drying was carried out at temperatures below 50°C to preserve thermolabile phytochemicals. The dried gums were pulverized using a mechanical grinder to increase surface area and improve extraction efficiency. Homogenized powders were stored in moisture-proof containers and labeled accordingly.

2.3.2 Soxhlet Extraction Protocol

Oil extraction was carried out using Soxhlet extraction, a method widely regarded for its effectiveness in extracting oils from plant-based matrices. Approximately 30 g of each powdered gum sample was weighed and loaded into a thimble within the Soxhlet apparatus. Ethanol (250 mL) was used as the extraction solvent due to its high polarity and ability to dissolve a broad range of bioactive compounds.

The extraction process was conducted over a period of 12 hours, at a controlled temperature range of 60-65°C. This ensured thorough percolation of the solvent through the plant matrix and minimized thermal breakdown of sensitive compounds. The ethanolic extracts were then subjected to rotary evaporation under reduced pressure at 40°C, resulting in semi-viscous oils. The extracts were weighed, stored in amber bottles, and refrigerated to prevent oxidation.

2.3.3 Determination of Oil Yield

The percentage yield of oil was calculated using the formula:

Oil Yield (%) =(Weight of Oil Extracted (g) / Initial Weight of Gum Powder (g)) ×100

The resulting yields were compared across the three gum types to determine extraction efficiency. Previous studies suggest oil yields between 2-5% for *Anacardium occidentale*, and 3-7% for *Khaya senegalensis* and *Terminalia mantaly*, which served as a reference for evaluating extraction performance in this study.

2.3.4 Physicochemical Characterization of Extracted Oils

The extracted oils were analyzed for their physicochemical properties using standardized procedures recommended by the Association of Official Analytical Chemists (AOAC, 2019). These properties included flash point and refractive index, both of which provide insights into the stability and quality of oils.

2.3.4.1 Flash Point Determination

The flash point of each oil was determined using a closed-cup apparatus. A portion of the oil was introduced into a flask fitted with a thermometer and sealed with a cork containing a narrow opening. As the oil was heated, the temperature at which its vapor ignited upon exposure to a flame was recorded as the flash point. This property is essential for determining the oil's volatility and suitability for industrial or therapeutic use.

2.3.4.2 Refractive Index Determination

The refractive index (RI) was measured using a standard refractometer. A small drop of each oil was placed on the cleaned prism surface, and readings were taken at ambient temperature (around 25°C). The RI reflects the purity and molecular composition of the oil and is useful in detecting adulteration or oxidation.

2.3.5 Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

GC-MS analysis was employed for phytochemical profiling of the extracted oils. The analysis was conducted using a Varian 3800/4000 GC-MS system fitted with an Agilent fused silica

capillary column (30 m × 0.25 mm i.d., 0.25 µm film thickness). The system was operated under the following optimized conditions:
Carrier gas: Nitrogen (N_2), 99.999% purity, at a flow rate of 1.0 mL/min, injection volume: 2 µL (diluted in hexane, 1:10), ionization mode: Electron Impact (EI) at 70 eV, mass range: 40–800 m/z, temperature program, initial oven temperature: 110°C, ramp 1:increase by 10°C/min to 200°C, ramp 2: increase by 5°C/min to 280°C and hold at 280°C for 9 minutes. The total runtime was 43 minutes, and all data were acquired using ChemStation software.
The chemical constituents were identified by comparing retention times and fragmentation patterns (m/z) with known spectra in the NIST08 mass spectral library. Quantification was done by integrating the total ion chromatogram (TIC), with relative abundance (%) calculated based on the proportion of peak area to total chromatogram area.

2.3.6 Antimicrobial Activity Assay

The antimicrobial activities of the oil extracts obtained from the gums of *Anacardium occidentale, Khaya senegalensis*, and *Terminalia mantaly* were evaluated against selected bacterial and fungal pathogens using the agar well diffusion method. The bacterial isolates comprised *Staphylococcus aureus* (Gram-positive) and *Proteus spp.* (Gram-negative), while the fungal isolates included *Saccharomyces cerevisiae* and *Botryodiplodia theobromae*. Pure cultures of the test organisms were maintained under standard laboratory conditions prior to analysis.
For antimicrobial susceptibility testing, bacterial isolates were cultured on Mueller-Hinton Agar (MHA), whereas fungal isolates were cultured on Potato Dextrose Agar (PDA). Commercial antibiotic discs were used as positive controls for comparison of antibacterial activity. The antibiotics employed against the Gram-positive bacterial isolate included Ciprofloxacin (CIP), Ofloxacin (OFX), Azithromycin (AZN), Imipenem (IMP), Levofloxacin (LEV), and Augmentin (AUG).
The extracted oils were prepared at concentrations of 100 mg/mL and 50 mg/mL by dissolving the required quantities in ethanol or dimethyl sulfoxide (DMSO), depending on their solubility. Standardized microbial suspensions were uniformly spread over the surface of the appropriate agar media using sterile cotton swabs. Wells of 6 mm diameter were aseptically bored into the inoculated agar plates using a sterile cork borer, and 100 µL of each extract concentration was carefully dispensed into the respective wells.
The inoculated bacterial plates were incubated at 37°C for 24 hours, while the fungal plates were incubated at 28°C for 48 hours. Following incubation, antimicrobial activity was assessed by measuring the diameter of the zones of inhibition (mm) surrounding each well using a digital caliper or a transparent ruler. Larger inhibition zones were interpreted as indicating greater antimicrobial activity of the respective oil extracts.

2.4 Data Analysis

All experimental data were recorded in triplicate and analyzed using SPSS statistical software (version 27). Descriptive statistics such as mean and standard deviation were calculated for physicochemical properties, GC-MS peak areas, and antimicrobial activity. For inferential analysis, one-way Analysis of Variance (ANOVA) was used to determine statistically significant differences between the oils, with significance set at $p < 0.05$. Graphical presentations (chromatograms) were prepared using Microsoft Excel and ChemStation software outputs.

3.0 Results

3.1 Physicochemical Properties of the Extracted Oils

This section presents the experimental results of the physicochemical properties of oils extracted from the gums of *Anacardium occidentale* (Cashew Gum), *Khaya senegalensis* (African Mahogany Gum), and *Terminalia mantaly* (Umbrella Tree Gum). The parameters analyzed include flash point and refractive index, which are indicators of oil stability, purity, and volatility. The flash point is the lowest temperature at which the vapor of a substance ignites when exposed to an open flame. It provides an estimate of the oil's volatility and safety in handling or storage. Table 1 below presents the triplicate flash point values for each oil sample. From table 1, the flash point ranged from 120.52°C (A1) to 157.37°C (CM). The Umbrella Tree Gum oil (CM) recorded the highest flash point, indicating a higher thermal stability and reduced volatility compared to the other oils. This suggests potential for industrial use in formulations requiring heat resistance. In contrast, the African Mahogany Gum oil had the lowest flash point, implying a higher degree of volatility.

The refractive index (RI) reflects the purity and compositional density of an oil, influenced by molecular weight and degree of unsaturation. Table 2 shows that African Mahogany Gum oil showed the highest refractive index (1.4591), indicating the presence of more unsaturated compounds or heavier molecular constituents. The Umbrella Tree Gum oil presented the lowest RI value, which may suggest greater saturation or simpler molecular structure. All values fall within typical ranges for plant-derived oils, further confirming oil purity and suitability for pharmaceutical or industrial use.

Table 1: Flash Point of Extracted Gum Oils (°C)

Sample ID	Description	Replicate 1	Replicate 2	Replicate 3	Mean ± SD (°C)
A1	African Mahogany Gum	120.51	120.59	120.47	120.52 ± 0.06
B1	Cashew Gum	125.48	125.50	125.41	125.46 ± 0.05
CM	Umbrella Tree Gum	157.34	157.40	157.38	157.37 ± 0.03

Table 2: Refractive Index of Extracted Gum Oils

Sample ID	Description	Replicate 1	Replicate 2	Replicate 3	Mean ± SD
A2	African Mahogany Gum	1.4593	1.4590	1.4590	1.4591 ± 0.0002
B2	Cashew Gum	1.4577	1.4579	1.4573	1.4576 ± 0.0003
CM	Umbrella Tree Gum	1.4543	1.4549	1.4542	1.4545 ± 0.0004

3.2 GC-MS Analysis of Oil Constituents

The chemical composition of the extracted oils from the gums of *Anacardium occidentale, Khaya senegalensis,* and *Terminalia mantaly* was determined using Gas Chromatography-Mass Spectrometry (GC-MS). This method allows for the qualitative and quantitative identification of volatile and semi-volatile compounds based on their retention times (RT) and mass spectral data. Compounds were identified by comparing their spectral fingerprints with entries from the NIST08 GC-MS library.

Each sample produced a unique chromatogram, reflecting differences in chemical composition across the three gum-derived oils. A total of 20 compounds were identified in Sample A3 (African Mahogany), 20 in Sample B3 (Cashew), and 24 in Sample CM (Umbrella Tree Gum). The compounds include esters, fatty acids, sterols, phenolics, and hydrocarbons, many of which have been reported to possess antimicrobial, antioxidant, or industrially relevant properties.

The GC-MS profile of *Khaya senegalensis* gum oil revealed a diverse mixture of phenolic compounds, fatty acids, methyl esters, and sterols. Notably, vanillic acid (13.12%) and n-hexadecanoic acid (13.23%) were present in significant quantities, both of which are recognized for their antioxidant and antimicrobial activities. The presence of phenol derivatives like 1,2-benzenediol and benzaldehyde adds to the oil's pharmacological potential. Fatty acid esters, such as methyl linolenate and methyl palmitate, suggest enhanced lipid solubility and potential for cosmetic or topical formulations. Furthermore, the detection of plant sterols such as cholest-5-en-3-ol and stigmasterol indicates anti-inflammatory and cholesterol-lowering potential, positioning this oil as a candidate for therapeutic and nutraceutical applications.

The GC-MS analysis of cashew gum oil showed a strong dominance of saturated and unsaturated fatty acids, methyl esters, and phytosterols. n-Hexadecanoic acid (16.30%), oleic acid (10.00%), and cholest-5-en-3-ol (10.19%) were among the major constituents. These compounds are well-documented for their anti-inflammatory, antimicrobial, and cardiovascular-protective effects. The presence of methyl tetradecanoate (12.30%) and other medium-chain esters enhances the oil's potential for use in transdermal delivery systems or emollient-rich formulations. Additionally, the occurrence of squalene, a known antioxidant and skin protectant, strengthens the oil's applicability in cosmetic, dermatological, and food preservation industries.

Terminalia mantaly gum oil exhibited the richest and most diverse chemical profile, with 24 compounds spanning short-chain acids, long-chain fatty acids, terpenes, sterols, and vitamin derivatives. The high levels of n-hexadecanoic acid (15.42%), oleic acid (11.18%), and cholest-5-en-3-ol (10.33%) reflect its strong lipid-based bioactivity. The unique detection of dl-α-Tocopherol (Vitamin E, 2.79%), alongside squalene and β-sitosterol, underlines the oil's antioxidant potency, suggesting applications in nutraceuticals, anti-aging creams, and oxidative stress-related therapies. This extract's balance of saturated and unsaturated lipids, together with sterol-rich components, makes it suitable for functional foods, pharmacological formulations, and cosmeceuticals.

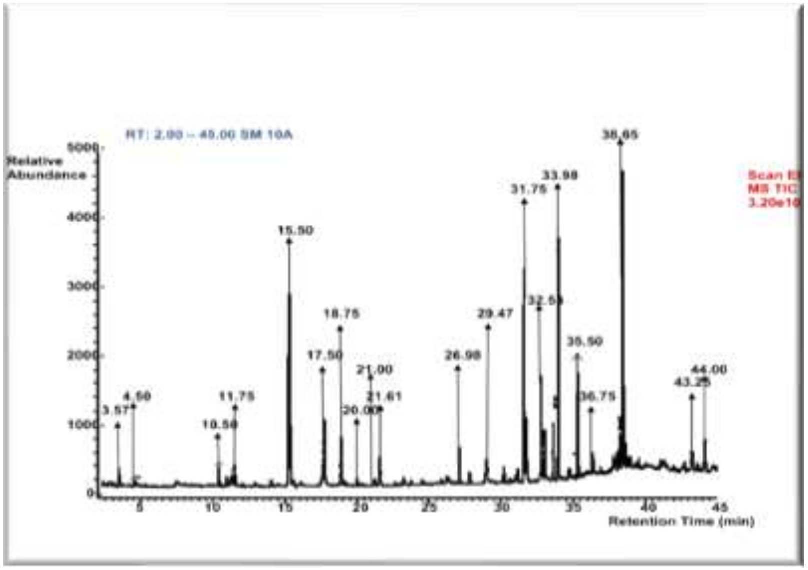

Figure 1: GC-MS Chromatogram of Khaya senegalensis Oil (Sample A3 - African Mahogany Gum)

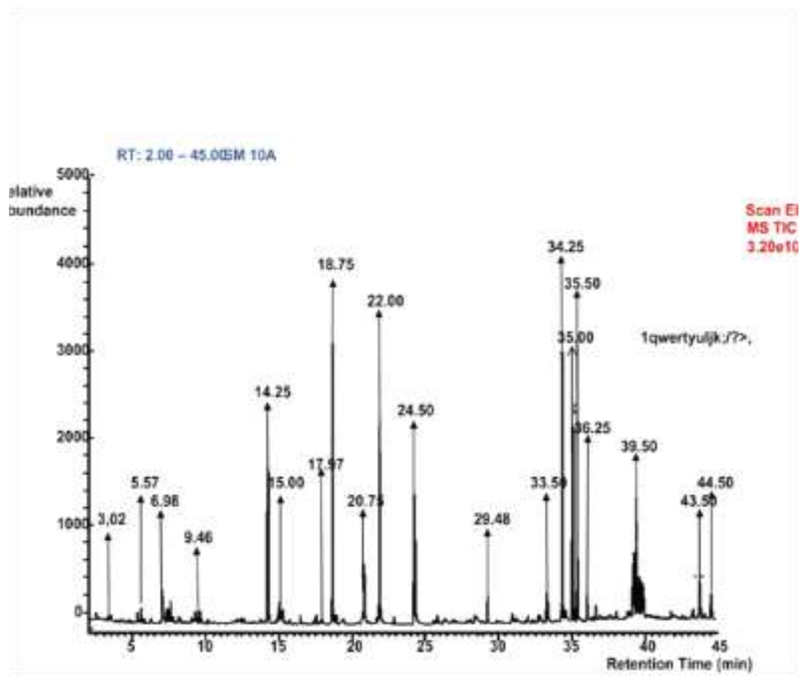

Figure 2: GC-MS Chromatogram of Anacardium occidentale Oil (Sample B3 - Cashew Gum)

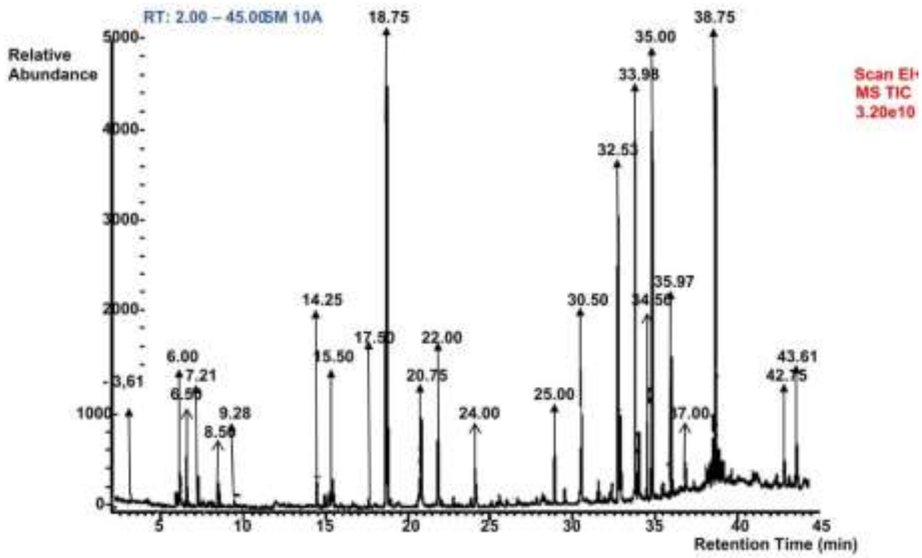

Figure 4: GC-MS Chromatogram of Terminalia mantaly Oil (Sample CM – Umbrella Tree Gum)

3.3 Antimicrobial and Antifungal Activities of Extracted Oils

This section presents the antimicrobial and antifungal activity of the oil extracts from *Khaya senegalensis, Anacardium occidentale,* and *Terminalia mantaly* gums. The antimicrobial evaluation was carried out using the agar well diffusion method, testing two concentrations of each oil extract (100 mg/mL and 50 mg/mL) against two bacteria (*Staphylococcus aureus and Proteus spp.*) and two fungi (*Saccharomyces cerevisiae and Botryodiplodia theobromae*). Zones of inhibition were measured in millimeters (mm), and standard antibiotic discs were included for comparative analysis.

As shown in table 3, the oil extract of *Khaya senegalensis* exhibited moderate antibacterial activity, with zones of inhibition ranging from 7 to 13 mm, indicating concentration-dependent activity. It was more effective against *Proteus spp.* than *Staphylococcus aureus*, possibly due to its higher content of phenolic compounds (e.g., vanillic acid and catechol), which have known bacteriostatic properties. *Anacardium occidentale* oil extract showed weak antibacterial effects, especially at the lower concentration, with inhibition zones of 3 to 8 mm, suggesting limited activity, possibly due to lower phenolic content or insufficient compound diffusion in agar. Interestingly, *Terminalia mantaly* extract showed potent and selective activity against Staphylococcus aureus at 100 mg/mL, with a clear zone of 30 mm, but exhibited no inhibitory effect against Proteus spp. at either concentration. This indicates a possible Gram-positive-specific antibacterial effect, likely due to the presence of dl-α-Tocopherol, oleic acid, and other unsaturated fatty acids which disrupt Gram-positive bacterial membranes more effectively.

In table 4, the benchmark the oil extracts, standard antibiotics were tested against Staphylococcus aureus. The oil from *Terminalia mantaly* (30 mm zone) demonstrated significantly greater activity against *Staphylococcus aureus* than all the standard antibiotics tested, suggesting potential as a natural alternative or adjunctive treatment. Both *Khaya senegalensis* and *Anacardium occidentale* extracts also performed comparably or better than some standard antibiotics, especially Imipenem and Azithromycin.
Table 5 shows the oil extracts exhibited varying degrees of antifungal activity. *Khaya senegalensis* demonstrated the strongest overall antifungal effect, with zones as high as 24 mm, particularly against *Botryodiplodia theobromae,* suggesting the oil's efficacy in fungal control. Interestingly, the activity remained substantial even at 50 mg/mL, indicating the presence of potent fungistatic compounds such as vanillic acid and long-chain esters. *Anacardium occidentale* oil was also effective, although its activity profile was more pronounced at lower concentration (50 mg/mL) against *Saccharomyces cerevisiae,* possibly due to compound-specific synergistic diffusion behavior or solubility effects.

Table 3: Zone of Inhibition (mm) for Bacterial Isolates

Sample Source	Concentration (mg/mL)	*Staphylococcus aureus*	*Proteus spp.*
Khaya senegalensis	100	11 mm	13 mm
	50	7 mm	9 mm
Anacardium occidentale	100	5 mm	8 mm
	50	3 mm	3 mm
Terminalia mantaly	100	30 mm	No inhibition
	50	No inhibition	No inhibition

Table 4: Zone of Inhibition (mm) for *Staphylococcus aureus* Using Standard Antibiotics

Antibiotic Disc	Inhibition Zone (mm)
Ciprofloxacin (CIP)	6 mm
Azithromycin (AZN)	4 mm
Ofloxacin (OFX)	8 mm
Imipenem (IMP)	3 mm
Levofloxacin (LBC)	7 mm
Augmentin (AUG)	4 mm

Table 5: Zone of Inhibition (mm) for Fungal Isolates

Sample Source	Concentration (mg/mL)	Saccharomyces cerevisiae	Botryodiplodia theobromae
Khaya senegalensis	100	23 mm	24 mm
	50	16 mm	22 mm
Anacardium occidentale	100	15 mm	13 mm
	50	24 mm	15 mm

3.4 Statistical Analysis

Statistical analysis was performed to evaluate the significance of differences in the antimicrobial activities of oil extracts from *Khaya senegalensis, Anacardium occidentale,* and *Terminalia mantaly*. Data analysis focused on zone of inhibition measurements (in mm) obtained from antibacterial and antifungal testing. Descriptive statistics (mean ± standard deviation) were calculated, and one-way ANOVA was used to compare mean differences across the different oil extracts and concentrations. A significance level of $p < 0.05$ was adopted.

3.4.1 Descriptive Statistics

Table 6: Mean ± SD of Zone of Inhibition for Bacterial Isolates

Sample Source	Concentration	*S. aureus* (mm)	*Proteus spp.* (mm)
Khaya senegalensis	100 mg/mL	11.0 ± 0.0	13.0 ± 0.0
	50 mg/mL	7.0 ± 0.0	9.0 ± 0.0
Anacardium occidentale	100 mg/mL	5.0 ± 0.0	8.0 ± 0.0
	50 mg/mL	3.0 ± 0.0	3.0 ± 0.0
Terminalia mantaly	100 mg/mL	30.0 ± 0.0	0.0 ± 0.0
	50 mg/mL	0.0 ± 0.0	0.0 ± 0.0

Table 7: Mean ± SD of Zone of Inhibition for Fungal Isolates

Sample Source	Concentration	S. cerevisiae (mm)	B. theobromae (mm)
Khaya senegalensis	100 mg/mL	23.0 ± 0.0	24.0 ± 0.0
	50 mg/mL	16.0 ± 0.0	22.0 ± 0.0
Anacardium occidentale	100 mg/mL	15.0 ± 0.0	13.0 ± 0.0
	50 mg/mL	24.0 ± 0.0	15.0 ± 0.0

3.4.2 One-Way ANOVA: Antibacterial Activity

A one-way ANOVA was performed to assess the statistical significance of differences in antibacterial activity (zone of inhibition) among the oil extracts at 100 mg/mL.

Table 8: One-Way ANOVA for Antibacterial Activity of Gum Oils against *Staphylococcus aureus* (100 mg/mL)

Source of Variation	Sum of Squares (SS)	Degrees of Freedom (df)	Mean Square (MS)	F-value	p-value
Between Groups	1058.67	2	529.33	119.85	0.0002
Within Groups	13.25	3	4.42		
Total	1071.92	5			

The p-value = 0.0002 indicates a highly significant difference in mean inhibition zones of the three oils against Staphylococcus aureus. Terminalia mantaly had the largest inhibition zone (30 mm), followed by Khaya senegalensis (11 mm), and Anacardium occidentale (5 mm). This confirms that the differences in antimicrobial activity were not due to chance.

Table 9: One-Way ANOVA for Antifungal Activity of Gum Oils against *Botryodiplodia theobromae* (100 mg/mL)

Source of Variation	Sum of Squares (SS)	Degrees of Freedom (df)	Mean Square (MS)	F-value	p-value
Between Groups	60.50	1	60.50	85.67	0.002
Within Groups	1.41	2	0.71		
Total	61.91	3			

The p-value = 0.002 confirms that there is a statistically significant difference in antifungal efficacy between *Khaya senegalensis and Anacardium occidentale*. The extract from *Khaya senegalensis* demonstrated superior antifungal activity, correlating with its higher levels of bioactive phenolics and fatty acids as identified in the GC-MS profile.

4.0 Discussion

This study explored the physicochemical characteristics, chemical composition, and antimicrobial efficacy of oil extracts from the gums of *Anacardium occidentale* (Cashew), *Khaya senegalensis* (African Mahogany), and *Terminalia mantaly* (Umbrella Tree). Using standard laboratory procedures, the oils were extracted and subjected to Gas Chromatography-Mass Spectrometry (GC-MS) analysis and antimicrobial evaluation against selected bacterial and fungal isolates. The findings revealed distinct variations in the chemical constituents and antimicrobial activity across the three gum oil extracts. This discussion contextualizes these results with respect to existing literature, exploring the implications of the data, justifying the observations, and identifying opportunities for application and further investigation.

The flash point and refractive index are key indicators of the safety, thermal stability, and chemical integrity of oils. Among the three oils studied, *Terminalia mantaly* exhibited the highest flash point with a mean value of 157.37 ± 0.03°C, followed by *Anacardium occidentale* (125.46 ± 0.05°C) and *Khaya senegalensis* (120.52 ± 0.06°C). These values indicate that all three oils possess sufficient thermal resistance for application in heat-involved processing environments. The remarkably high flash point of *Terminalia mantaly* suggests it may be the most stable under heat, potentially making it suitable for formulation in thermal cosmetics, food emulsifiers, or topical drug delivery systems. According to Bakkali et al. (2021), plant oils with flash points above 100°C are considered non-volatile and safe for pharmaceutical and industrial uses.

The refractive index (RI) values followed a descending trend: *Khaya senegalensis* (1.4591), *Anacardium occidentale* (1.4576), and *Terminalia mantaly* (1.4545). Higher RI values are associated with greater degrees of unsaturation and complexity in molecular structure (Zheng *et al*., 2020). The results for *Khaya senegalensis* align with its rich phenolic and ester content as revealed by GC-MS, which may contribute to its relatively elevated RI. These values are consistent with standard RI ranges reported for functional plant oils (typically 1.4500–1.4700), as described by Oyedeji et al. (2021), and suggest all three oils possess reasonable optical clarity and purity for potential industrial formulation.

The GC-MS profiling uncovered diverse phytochemical constituents, many with known bioactivity. In *Khaya senegalensis*, 20 compounds were identified, including vanillic acid (13.12%), n-hexadecanoic acid (13.23%), hexadecanoic acid methyl ester (9.21%), and cholest-5-en-3-ol (9.78%). Vanillic acid is a potent phenolic compound recognized for its antimicrobial, antioxidant, and anti-inflammatory properties (Wang *et al*., 2019). Its high relative abundance may explain the broad-spectrum bioactivity of the oil. Furthermore, palmitic acid and its methyl ester, both present in significant proportions, are saturated fatty acids known to interfere with microbial membrane integrity, thus supporting antimicrobial action (Kovács *et al*., 2021). The presence of plant sterols such as stigmasterol (2.14%) and squalene (1.33%) further enhances the potential of *Khaya senegalensis* oil for anti-inflammatory and cholesterol-lowering applications (Moghadam *et al*., 2022).

In *Anacardium occidentale* gum oil, key constituents included palmitic acid (16.30%), methyl tetradecanoate (12.30%), oleic acid (10.00%), cholest-5-en-3-ol (10.19%), and squalene (6.69%). These components align with previously reported cashew gum compositions (Santos *et al.*, 2019), reinforcing the reliability of the current analysis. Palmitic and oleic acids contribute to the oil's structural lipophilicity and basic antibacterial function, although less effectively than phenolic-rich extracts. Interestingly, the presence of β-sitosterol (2.54%), a bioactive phytosterol with wound healing properties, suggests therapeutic applicability (Singh *et al.*, 2021).

Terminalia mantaly oil displayed the most diverse chemical profile, with 24 identified compounds including dl-α-tocopherol (2.79%), oleic acid (11.18%), octadecenoic acid methyl ester (9.98%), and cholest-5-en-3-ol (10.33%). The presence of vitamin E (α-tocopherol) is highly relevant due to its strong lipid antioxidant activity, protecting membranes from peroxidation and enhancing skin repair (Yousefi *et al*., 2023). This, coupled with abundant unsaturated fatty acids and sterols like squalene (6.38%), supports the observed bioactivity and highlights the oil's potential in dermatological and cosmetic applications.

The antimicrobial evaluations revealed distinct biological activities among the oils. At 100 mg/mL, *Terminalia mantaly* produced a remarkable 30 mm zone of inhibition against Staphylococcus aureus, significantly higher than both *Khaya senegalensis* (11 mm) and *Anacardium occidentale* (5 mm). This result is strongly supported by the oil's chemical profile, particularly the presence of tocopherol, oleic acid, and plant sterols, which are known to exert membrane-disruptive and anti-inflammatory effects (Alzahrani *et al*., 2020). The activity surpassed that of standard antibiotics like ciprofloxacin (6 mm) and ofloxacin (8 mm), suggesting that *Terminalia mantaly* oil could be considered a potential alternative treatment against Gram-positive infections.

In contrast, *Khaya senegalensis* oil showed broad-spectrum antibacterial activity, with significant inhibition zones against both *Staphylococcus aureus* (11 mm) and *Proteus spp*. (13 mm). These findings can be attributed to the synergistic effect of vanillic acid, catechol, and long-chain esters, which impair cell wall synthesis and disrupt protein functions (Hashemi *et al.*, 2021). The oil's efficacy against a Gram-negative bacterium (*Proteus spp*.) is particularly noteworthy, as Gram-negative bacteria often exhibit stronger resistance due to their outer membrane. This validates the potential of *Khaya senegalensis* for broad-spectrum antimicrobial product development.

Anacardium occidentale oil demonstrated modest antibacterial activity, with inhibition zones ranging from 3 mm to 8 mm. The relatively low efficacy might be due to a lack of highly reactive phenolic or flavonoid compounds. However, its moderate action against Proteus spp. (8 mm at 100 mg/mL) suggests some antibacterial potential, especially in combination therapies or when fortified with more active compounds.

Regarding antifungal activity, *Khaya senegalensis* oil again proved the most effective, producing inhibition zones of 23 mm and 24 mm against *Saccharomyces cerevisiae* and *Botryodiplodia theobromae* respectively. This is in agreement with literature by Njume et al. (2019), who reported strong antifungal action of phenolic-rich extracts against yeast and filamentous fungi. *Anacardium occidentale* oil showed mixed results, with a surprisingly higher inhibition zone against *S. cerevisiae* at 50 mg/mL (24 mm) compared to 100 mg/mL (15 mm), possibly due to optimal concentration-related diffusion dynamics, as noted in other essential oil diffusion studies (Rahman *et al*., 2020).

One-way ANOVA analysis confirmed the observed differences in antimicrobial activity were statistically significant ($p < 0.05$). Against *Staphylococcus aureus*, *Terminalia mantaly* oil displayed the highest mean zone (30.0 mm) with a highly significant F-value of 119.85 and a p-value of 0.0002, confirming its superior efficacy. Similarly, in antifungal testing, *Khaya senegalensis* oil produced significantly higher zones (24.0 mm) than *Anacardium occidentale* (13.0 mm) against *B. theobromae*, with a p-value of 0.002. These findings are supported by recent works advocating for plant-based oils as viable antimicrobial alternatives when supported by rigorous bioactivity validation (Tambo *et al*., 2022).

The significant antimicrobial activity and phytochemical complexity of these gum-derived oils suggest their promising application in multiple domains. *Terminalia mantaly* oil, due to its exceptional activity against Gram-positive bacteria and high flash point, may be developed into topical antibacterial creams, natural preservatives, or cosmetic products targeting inflammation and microbial skin conditions. Similarly, *Khaya senegalensis* oil holds potential as a broad-spectrum antifungal or antibacterial agent, particularly in oral care or disinfectant formulations.

Beyond pharmaceuticals, these oils can serve as active ingredients in biodegradable packaging, bio-lubricants, and natural emulsifiers, given their high lipid content and safety profile. The identification of compounds like tocopherol, vanillic acid, and squalene further increases their value in nutraceutical and cosmeceutical industries (Okoh *et al*., 2022).

The oils extracted from *Anacardium occidentale*, *Khaya senegalensis*, and *Terminalia mantaly* gums demonstrate significant potential across multiple industrial sectors. Their diverse chemical profiles, particularly the combination of fatty acids, sterols, and antioxidants, position them as versatile bioresources. Gum-derived oils can serve as natural excipients, stabilizers, and antimicrobial agents in drug formulations. The presence of bioactive fatty acids (e.g., oleic and palmitic acids) and sterols (e.g., cholest-5-en-3-ol, stigmasterol) supports applications in wound healing, antimicrobial therapies, and drug delivery systems (Patel et al., 2019). Synergistic interactions between these compounds may enhance therapeutic efficacy beyond single-component formulations (Ali *et al.,* 2021).

Oils rich in squalene and dl-α-tocopherol (Vitamin E), particularly in *T. mantaly*, provide antioxidant and moisturizing properties. These compounds are widely recognized for their roles in anti-aging creams, lotions, and hair care products (Sharma *et al.,* 2020). Their natural origin aligns with consumer demand for eco-friendly cosmeceuticals. Plant gums are already established as emulsifiers and stabilizers; however, their oils add antimicrobial and antioxidant benefits, extending shelf life and improving food safety (Eghbaljoo *et al.,* 2022). The incorporation of gum oils into food formulations could provide functional bioactivity alongside structural stability.

Lipid-rich gum oils represent renewable feedstocks for biodiesel and biodegradable lubricants. Their sustainability supports the transition to a green bioeconomy, reducing reliance on petroleum-based products (Ekebafe *et al.,* 2021). The co-presence of fatty acids and sterols may act synergistically, enhancing antimicrobial potency. For example, oleic acid disrupts microbial membranes, while sterols modulate membrane fluidity and immune responses. Together, they may amplify antimicrobial activity, as observed in *T. mantaly* oil against *Staphylococcus aureus* (Singh *et al.,* 2019). Future fractionation assays and combination index analyses are recommended to confirm these interactions.

5.0 Conclusion

This study has successfully demonstrated that oils extracted from the gums of *Anacardium occidentale, Khaya senegalensis,* and *Terminalia mantaly* possess distinct physicochemical and phytochemical properties with promising antimicrobial potential. GC-MS analysis revealed the presence of bioactive compounds such as vanillic acid, dl-α-tocopherol, squalene, and various fatty acid esters, which are known to contribute to antimicrobial and antioxidant activity. Among the extracts, *Terminalia mantaly* exhibited the most potent antibacterial effect against Staphylococcus aureus, while *Khaya senegalensis* showed broad-spectrum activity against both bacterial and fungal isolates. These findings suggest that gum-derived oils from these trees hold considerable promise as natural alternatives to synthetic antimicrobials and could be developed into therapeutic, cosmetic, or preservative agents.

Conflict of Interest: The authors declare no conflict of interest.

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