

Antioxidant Activity and Chemical Composition of *Origanum Majorana*, *Cedrus Atlantica*, and *Commiphora Myrrha* Essential Oils: Implications for Antifungal Applications

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Abstract

BACKGROUND/AIMS: Rising antifungal resistance has encouraged the investigation of plant-derived compounds as alternative or adjunctive agents in fungal infections. Essential oils possess bioactive constituents with antioxidant and antifungal properties. This study aimed to evaluate the antioxidant activity of essential oils obtained from *Origanum majorana*, *Cedrus atlantica*, and *Commiphora myrrha* and to characterize their chemical composition using gas chromatography-mass spectrometry (GC-MS), focusing on their potential relevance to antifungal applications.

MATERIALS AND METHODS: Antioxidant activity was assessed using the 2,2-diphenyl-1-picrylhydrazine (DPPH) radical scavenging assay, and half-maximal inhibitory concentration (IC_{50}) values were calculated with ascorbic acid (AA) as the reference standard. Chemical profiles of the essential oils were determined by GC-MS analysis. Statistical comparisons were performed using one-way ANOVA, with $p < 0.05$ considered statistically significant.

RESULTS: Among the tested essential oils, *C. myrrha* exhibited the strongest antioxidant activity (IC_{50} : 18.167 ± 0.972 mg/mL), followed by *C. atlantica* (IC_{50} : 213.037 ± 74.219 mg/mL) and *O. majorana* (IC_{50} : 333.207 ± 31.954 mg/mL). AA showed substantially higher antioxidant activity (IC_{50} : 0.021 ± 0.0003 mg/mL). GC-MS analysis revealed that *C. myrrha* oil was rich in oxygenated sesquiterpenes, particularly curzerene and furanoeudesma-1,3-diene; *O. majorana* was characterized by terpinen-4-ol and γ -terpinene; and *C. atlantica* was dominated by himachalene derivatives.

CONCLUSION: The findings demonstrate that *C. myrrha* essential oil possesses superior antioxidant capacity among the oils tested, likely attributable to its high content of oxygenated sesquiterpenes. While direct antifungal activity was not assessed, the observed antioxidant properties may support the use of myrrh essential oil as a natural adjuvant in antifungal formulations targeting fungal infections associated with oxidative stress.

Keywords: *Origanum majorana*, *Cedrus atlantica*, *Commiphora myrrha*, essential oil, antioxidant, DPPH

INTRODUCTION

Alternative approaches are of great importance in the treatment of fungal infections, especially due to increasing resistance problems.¹ In this context, the antimicrobial and especially antifungal properties of essential oils obtained from plants offer the potential for the development of new therapeutic strategies.² Essential oils have attracted attention with their antibacterial, antifungal, anticancer, and antioxidant effects

in various sectors such as food, cosmetics, pharmaceuticals, textiles, and agriculture.³ These natural compounds, secondary metabolites of plants, function as defense mechanisms against pathogens and exhibit broad-spectrum antifungal activities.⁴ With these properties, they constitute a promising therapeutic alternative, especially against pathogenic fungi such as *Candida* species in humans.⁵ In addition, the antioxidant capacity of essential oils may contribute to the treatment process by reducing oxidative stress caused by fungal infections.⁶

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Compared to other species in the *Origanum* genus, *Origanum majorana* (Sweet Marjoram) essential oil contains higher concentrations of monoterpene alcohols and hydrocarbons and lower concentrations of phenolic compounds such as carvacrol and thymol. Recent chromatographic studies indicate that the principal component of this oil is frequently terpinen-4-ol. Gamma-terpinene, sabinene, alpha-terpinene, cis-sabinene hydrate, and linalyl acetate are other important phytochemicals. Some chemotypes contain thymol and carvacrol, whereas *Origanum majorana* usually contains terpinen-4-ol and sabinene as its main constituents. These components give the oil its unique smell and make it function better within the body. The antioxidant properties of this essential oil are based on the scavenging ability of its monoterpenes. Terpinen-4-ol and gamma-terpinene specifically reduce reactive oxygen species, thereby preventing lipid peroxidation. The antifungal mechanism of action is highly effective and primarily targets the fungal cell membrane. The lipophilic structure of *Origanum majorana* essential oil penetrates the chitin and glucan layers of the fungal cell wall and integrates into the cell membrane. This makes the membrane less flexible and less permeable. It also stops the production of ergosterol, causing intracellular ions (such as potassium) and essential macromolecules to leak out. This stops energy synthesis in the fungal cell (mitochondrial failure), thereby killing the cell. This method shows a lot of inhibition, especially against *Candida* species and dermatophytes.⁷

The essential oil of *Cedrus atlantica* exhibits a complex chemical composition, with sesquiterpenes constituting a major component. Recent studies show that the major constituents of the oil are beta-himachalene (30-50%), alpha-himachalene, and gamma-himachalene. The "himachalene" group is responsible for the biological activity of cedar oil. The oil also contains significant amounts of alpha-atlantone (a ketone), delta-cadinene, and cedrol, a sesquiterpene alcohol. Cedrol, a component of the oil, can crystallize, giving it its distinct aroma. The antioxidant properties of cedar essential oil are due to its sesquiterpenes, especially himachalenes, which can donate electrons. However, this effect is usually weaker than that of phenolic oils, such as thyme. Its antifungal mechanism is more narrowly focused. The essential oil of *Cedrus atlantica* has a unique ability to alter the structure of fungal biofilms. Studies show that this oil alters protein organization, facilitating fungal cell adhesion to surfaces and weakening the cell membrane. Studies show that the vapor phase of oil prevents mold, such as *Penicillium*, from forming spores and developing mycelium by irreversibly damaging the cytoplasmic membrane. The hydrophobic properties of sesquiterpene hydrocarbons interact with fungal membrane lipids, reducing the membrane potential and impeding adenosine triphosphate synthesis.⁸

The essential oil from *Commiphora myrrha* resin differs from other essential oils because it is rich in sesquiterpenes, specifically furano-sesquiterpenes that contain furan rings. Furano-eudesma-1,3-diene, curzeren, and lindrestrene are the primary components. There are also other sesquiterpenes, such as beta-elemene and germacrene. The unique chemical composition of myrrh oil, which includes molecules with furan rings, gives it strong therapeutic and antibacterial properties. The antioxidant properties of myrrh essential oil are attributed to furan-ring-containing compounds that mitigate oxidative stress and bind metal ions. It has a complex mechanism for combating fungi. *Commiphora myrrha* essential oil can directly inhibit enzymatic activity in fungal cells. Studies, particularly those focused on *Candida albicans*

and other pathogenic yeasts, have shown that myrrh oil selectively disrupts cell membrane permeability and promotes the coagulation of intracellular components. Furthermore, recent studies indicate that the sesquiterpenes in myrrh oil undermine the defensive mechanisms of fungi and inhibit hyphal (thread-like) development, thus preventing the spread of infection and avoiding secondary fungal infections during the wound healing process.^{9,10}

This study evaluated the antioxidant activity of marjoram (*Origanum majorana*), cedarwood (*Cedrus atlantica*), and myrrh (*Commiphora myrrha*) essential oils and characterized their chemical compositions using gas chromatography-mass spectrometry (GC-MS). The results are discussed in the context of their potential relevance to future antifungal applications.

MATERIALS AND METHODS

The current study was performed only using commercial essential oils and *in vitro* experimental techniques. No human subjects or experimental animals were used. Consequently, the ethical committee approval and informed consent were not needed for the present study.

Samples of Chemicals and Essential Oils

2,2-diphenyl-1-picrylhydrazine (DPPH), [ascorbic acid (AA); positive control], and methanol were purchased from Sigma-Aldrich (USA) and were of analytical reagent grade. Commercial essential oils of *Origanum majorana*, *Cedrus atlantica*, and *Commiphora myrrha* were obtained from a pharmacy in Adana, Türkiye, under the brand name Art de Huile. The oils were used as received, according to the product labeling, without additional botanical authentication or chemotype verification.

Antioxidant Activity

Free radical scavenging is measured using 1,1-diphenyl-2-picrylhydrazyl. The decrease in absorbance of the stable free radical DPPH at 517 nm can be used to evaluate the scavenging capacity of natural substances. DPPH is colorless when mixed with a scavenger and purple in its free-radical form. A 0.1 mM DPPH stock solution in methanol was prepared. This foil-wrapped solution is refrigerated to avoid degradation. The essential oil samples were tested for DPPH.^{11,12} Twelve MeOH dilutions were performed on essential oil samples. A 96-well plate was filled with 120 µL of diluted essential oil samples and AA. To initiate the reaction, 40 µL of 0.1 mM DPPH in MeOH was added. The absorbance of the reaction mixture at 517 nm was measured with a UV spectrophotometer after 45 minutes at room temperature. The mean was computed from three observations. Measurements were performed in triplicate wells (technical replicates), and mean values were used for calculations. The DPPH radical scavenging activity of samples and standards was calculated from the decrease in absorbance. The DPPH removal % was $[(A_{\text{Control}} - A_{\text{Sample}}) / A_{\text{Control}}] \times 100$. The half-maximal inhibitory concentration (IC_{50}) is then calculated. The antioxidant activities of the tested compounds were evaluated by comparing their IC_{50} values, which denote the concentrations necessary to block 50% of free-radical activity. Lower IC_{50} values correspond to higher antioxidant potency. Effective concentration (EC_{50}), antiradical power (ARP), and ascorbic acid equivalent antioxidant capacity (AEAC) were measured.¹³⁻¹⁶

Composition Analysis of the Essential Oil by Gas Chromatography-Mass Spectrometry

The content analysis was conducted utilizing GC-MS.¹⁷ The content analysis was conducted on a Shimadzu GC-MS/QP2010 Ultra equipped with a DB-5MS stationary-phase column (i.d. 0.25 mm; film thickness 0.25 μ m). Upon completion of the analysis, intricate chromatograms were deconvoluted using AMDIS. Moreover, retention time adjustment and data matrix creation were performed using SpectConnect software. The correlation analysis was conducted using Microsoft Excel.

Statistical Analysis

For the purpose of expressing all of the data, the mean and standard deviation of the IC_{50} measurements were utilized. Statistical comparisons were performed using one-way analysis of variance (ANOVA). When the overall ANOVA was significant, Dunnett's post-hoc test was applied to compare each essential oil with AA, the reference standard. A p-value <0.05 was considered statistically significant.

RESULTS

As the IC_{50} value decreased, the antioxidant ability of the essential oil samples correspondingly increased. A lower IC_{50} value indicates the presence of antioxidant action. An antioxidant is deemed more potent if it has a low IC_{50} - EC_{50} value and a high ARP-AEAC value. The essential oils were evaluated for their antioxidant efficacy according to the specified criteria. AA (IC_{50} : 0.021 ± 0.0003 mg/mL) is more effective than *Commiphora myrrha* (myrrh) essential oil (IC_{50} : 18.167 ± 0.972 mg/mL), *Cedrus atlantica* (cedarwood) essential oil (IC_{50} : 213.037 ± 74.219 mg/mL), and *Origanum majorana* (marjoram) essential oil (IC_{50} : 333.207 ± 31.954 mg/mL) (Figure 1 and 2). The other antioxidant parameters are displayed in Table 1.

The GC-MS analysis of the essential oils revealed distinctive chemical profiles closely associated with their antioxidant potentials. The essential oil of *Origanum majorana* (marjoram) was predominantly composed of terpinen-4-ol, γ -terpinene, and cis-thujanol-4, accompanied by smaller proportions of α -terpinene and sabinene. The GC-MS profile of *Commiphora myrrha* (myrrh) revealed a complex mixture rich in sesquiterpenes, with curzerene, furanoeudesma-1,3-diene, lindestrene, and β -elemene as major constituents. The essential oil of *Cedrus atlantica* consisted mainly of himachalenes.

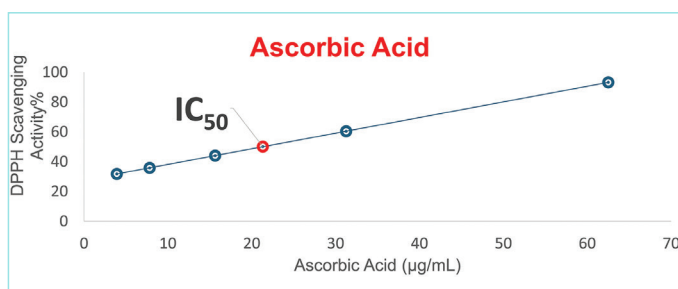


Figure 1. DPPH scavenging activity % of ascorbic acid as control group.

DPPH: 2,2-diphenyl-1-picrylhydrazine, IC_{50} : Half-maximal inhibitory concentration.

DISCUSSION

Essential oils contain bioactive compounds, such as terpenes and phenolics, that exhibit strong antifungal activity by disrupting fungal cell membranes, inhibiting cell wall synthesis, and interfering with mitochondrial function in fungi. They also inhibit fungal biofilm formation, which contributes to infection persistence and drug resistance. Marjoram (*Origanum majorana*) essential oil shows antifungal activity attributed to its content of oxygenated monoterpenes and terpenoids that disrupt fungal cell membranes and reduce ergosterol synthesis, essential for fungal cell viability. Cedarwood (*Cedrus atlantica*) essential oil exhibits antifungal effects mainly due to sesquiterpenes such as cedrol that impair fungal growth and mycelial integrity, with reported activity particularly against dermatophytes such as *Trichophyton* species, which are common agents of cutaneous fungal infections, including those affecting the hands. Myrrh (*Commiphora myrrha*) essential oil demonstrates antifungal properties through multiple mechanisms, including cell membrane disruption, mitochondrial dysfunction, and inhibition of fungal biofilm development, thereby enhancing its efficacy against persistent fungal infections. Studies report that essential oils have potential as natural antifungal agents for the treatment of hand fungal infections, with advantages including low toxicity, reduced development of resistance, and suitability for topical application. Their efficacy is linked to chemical composition variability dependent on plant source and extraction method.^{1,18,19}

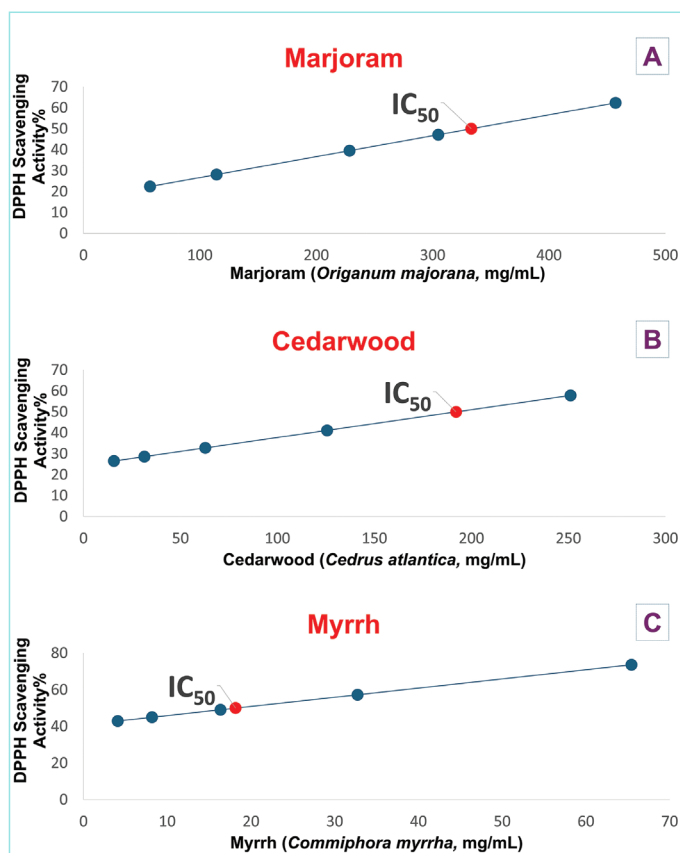


Figure 2. DPPH scavenging activity % of marjoram (A), cedarwood (B) and myrrh (C) essential oils.

DPPH: 2,2-diphenyl-1-picrylhydrazine, IC_{50} : Half-maximal inhibitory concentration.

Table 1. Antioxidant properties of the samples of essential oils

Compounds	IC ₅₀ (mg/mL)		EC ₅₀ (mg/mL)		ARP (mL/mg)		AEAC
	Mean ± SD	p-values	Mean ± SD	p-values	Mean ± SD	p-values	
Ascorbic acid	0.021±0.0003	-	0.0005±0.0000069	-	184937.934±2347.026	-	-
Marjoram (<i>Origanum majorana</i>) essential oil	333.207±31.954	0.000027	8.450±0.810	0.000027	11.834±1.087	0.0001	6.399
Cedarwood (<i>Cedrus atlantica</i>) essential oil	213.037±74.219	0.003	5.403±1.883	0.0038	18.509±8.976	0.0001	10.008
Myrrh (<i>Commiphora myrrha</i>) essential oil	18.167±0.972	0.0000027	0.461±0.024	0.000002	217.049±11.980	0.0001	117.363

IC₅₀ results are presented as means ± standard deviation from three measurements. Group differences were assessed using one-way ANOVA followed by Dunnett's post-hoc test (ascorbic acid as reference). The values of the essential oil samples with superscripts varied significantly ($p < 0.005$).

EC₅₀: Effective concentration, IC₅₀: Half-maximal inhibitory concentration, ARP: Antiradical power, AEAC: Ascorbic acid equivalent antioxidant capacity.

Similar studies have reported potential antioxidant effects of the same essential oils. The DPPH antioxidant activity of *Commiphora myrrha* resin was found to be IC₅₀: 26.86 mg/L, indicating moderate antioxidant activity.²⁰ For *Cedrus atlantica* essential oil, an IC₅₀ value of 0.126 mg/mL and similar values were reported using the DPPH method, indicating moderate-to-high antioxidant capacity.²¹ Antioxidant activity testing and health effects of *Origanum majorana* water and ethanol extracts; despite the high IC₅₀ value, potential antioxidant effects were reported.²²

In this study, the antioxidant capacity of AA is markedly superior to that of essential oils. These are natural results of potent antioxidants, such as AA. Among the essential oils tested, myrrh demonstrated the most potent antioxidant effect with IC₅₀ values of 18.167±0.972 mg/mL, showing a markedly higher radical scavenging activity compared to marjoram and cedarwood.

The GC-MS analysis confirmed that each essential oil possesses a unique chemical composition, which influences its antioxidant potential. *Commiphora myrrha* oil contained high levels of curzerene and furanoeudesma-1,3-diene, that are strongly associated with antioxidant and cytoprotective effects. Consequently, myrrh demonstrated the most potent antioxidant activity among the tested oils. *Origanum majorana* oil, rich in terpinen-4-ol and γ -terpinene, exhibited moderate activity, reflecting the contribution of oxygenated monoterpenes. In contrast, *Cedrus atlantica* oil, dominated by hydrocarbon sesquiterpenes such as β -himachalene, showed weaker antioxidant activity. These findings highlight the close relationship between chemical composition and biological activity. The presence of oxygenated terpenes, particularly furanosesquiterpenes and alcohol monoterpenes, appears to enhance

antioxidant capacity, suggesting that *Commiphora myrrha* essential oil could serve as a promising natural antioxidant source for pharmaceutical or nutraceutical applications.

Study Limitations

This work assessed antioxidant activity using an in vitro DPPH assay and did not include direct antifungal testing (e.g., MIC/MFC, inhibition zones, or dermatophyte culture models). In addition, the essential oils were purchased as commercial products from a local market, and no independent botanical authentication, chemotype verification, or batch standardization was performed; therefore, compositional variability may have influenced the measured activity.

CONCLUSION

This study aimed to evaluate the antioxidant potential of three essential oils, *Origanum majorana* (marjoram), *Cedrus atlantica* (cedarwood), and *Commiphora myrrha* (myrrh), and to determine their chemical composition using GC-MS. The antioxidant activities were assessed using the DPPH radical-scavenging method, and the IC₅₀ values were compared with those of AA. The results showed that *Commiphora myrrha* exhibited the highest antioxidant activity with IC₅₀: 18.167±0.972 mg/mL among other essential oils, which may be related to its high content of oxygenated sesquiterpenes. No direct antifungal assays were performed; the present findings should be interpreted as supportive evidence for potential formulation research rather than treatment efficacy. Further in vitro and in vivo studies are required to evaluate antifungal activity, safety, and batch-to-batch variability.

MAIN POINTS

- The study evaluates the antioxidant activities of *Origanum majorana*, *Cedrus atlantica*, and *Commiphora myrrha* essential oils using the 2,2-diphenyl-1-picrylhydrazine radical-scavenging assay.
- *Commiphora myrrha* essential oil exhibited the highest antioxidant capacity (IC_{50} : 18.167 ± 0.972 mg/mL) among the tested oils.
- Gas chromatography-mass spectrometry analysis revealed that the high antioxidant potential of *Commiphora myrrha* is associated with its rich content of oxygenated sesquiterpenes, such as curzerene and furanoeudesma-1,3-diene.
- The findings suggest that *Commiphora myrrha* essential oil could serve as a natural adjuvant in antifungal formulations targeting oxidative stress-related fungal infections.
- This study highlights the potential of essential oils as alternatives for antifungal formulations, particularly as antioxidant adjuncts, and emphasizes the need for direct antifungal efficacy testing.

ETHICS

Ethics Committee Approval: The author declare that the materials and methods used in the current study do not need approval from an ethics committee or special legal permission. The study was not conducted on humans or experimental animals.

Informed Consent: The study did not include human participants or samples. Informed consent was not applicable.

Acknowledgement

During the preparation of this work, the author(s) used Perplexity to generate summaries of research articles relevant to the topic. The summaries were assessed by juxtaposing them with expert-authored summaries in the field. After the precision and pertinence of the generated summaries were verified, they were incorporated into the literature review section of the article. QuillBot was used to edit the language of the article. After carefully reviewing and editing the content as necessary, the author(s) take full responsibility for the publication's content. The integration of AI tools substantially improved the efficiency of the literature review process and the thoroughness of the collected research insights.

DISCLOSURES

Financial Disclosure: The author declared that this study received no financial support.

Declaration of Generative AI and AI-assisted Technologies in the Writing Process: During the preparation of this work, the author(s) used ChatGPT to improve “readability” and “language”. After using this tool/service, the author(s) reviewed and edited the content as needed and took full responsibility for the content of the publication.

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