



**INTERNATIONAL JOURNAL OF
PHARMACEUTICAL SCIENCES**
[ISSN: 0975-4725; CODEN(USA): IJPS00]
Journal Homepage: <https://www.ijpsjournal.com>



Research Article

Comparative Evaluation of Antibacterial and Antioxidant Efficacy of Essential Oils of *Cymbopogon citratus* and *Eucalyptus globulus*

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ARTICLE INFO

Published: 8 Jan 2026

Keywords:

Hydro-distillation,
Clevenger Apparatus,
Essential Oils, Antibacterial
Activity, Antioxidant
Activity.

DOI:

10.5281/zenodo.18186237

ABSTRACT

Humans are constantly exposed to pathogenic vectors such as bacteria, fungi, viruses, mosquitoes, flies, and insects, which are responsible for the transmission of various diseases. While modern healthcare predominantly relies on allopathic medicines, their prolonged use may lead to adverse side effects. In search of a safer alternative, herein, Essential Oils (EOs) were extracted from the dried leaves of Lemongrass (*Cymbopogon citratus*) and Eucalyptus (*Eucalyptus globulus*) via hydrodistillation using a Clevenger apparatus. The EOs were characterized using GC-MS (Gas Chromatography Mass Spectroscopy). Further, the antibacterial efficacy was evaluated by the agar well diffusion method against *Staphylococcus aureus* (Gram-positive) and *Escherichia coli* (Gram-negative) bacteria. The results of the study have revealed that EO of Eucalyptus (*E. globulus*) exhibited higher efficacy against *E. coli*, while Lemongrass (*C. citratus*) EO was more effective against *S. aureus*. Furthermore, the antioxidant activity was assessed by 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay, with ascorbic acid as a standard, indicating that Lemongrass (*C. citratus*) EO possessed higher antioxidant activity than Eucalyptus (*E. globulus*) EO. GC-MS analysis identified the major volatile compounds in Lemongrass (*C. citratus*) EO as E-citral (26.32%), Z-citral (16.35%), Nerol (14.46%), and Geraniol (11.38%). The predominant compounds in Eucalyptus (*E. globulus*) EO were γ -terpinene (45.81%), β -pinene (16.21%), α -pinene (5.60%), Eucalyptol 1.94%, Camphene 1.91% and D-limonene 1.94%. The EOs of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*) exhibit promising antibacterial and antioxidant properties, supporting their potential applications in food preservation, cosmetics, mosquito repellents, and pharmaceutical formulations.

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Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.



INTRODUCTION

The growing global concern regarding the adverse health and environmental effects of synthetic additives has spurred a paradigm shift towards natural, sustainable, and bioactive alternatives. Plant-derived essential oils (EOs) have emerged as a major focus of scientific research due to their complex chemical composition and broad spectrum of biological activities. These volatile secondary metabolites, with a history of use in traditional medicine, are now recognized for their significant potential in modern industries such as food preservation, pharmaceuticals, cosmetics, and agrochemicals^[1].

The rise of antimicrobial resistance (AMR) necessitates an urgent quest for novel therapeutic agents to combat Drug-resistant pathogenic microorganisms. Simultaneously, diseases like cancer, neurodegenerative disorders, and cardiovascular conditions have increased the focus on identifying potent and safer antioxidants^[2]. To this end, medicinal plants have re-emerged as an invaluable reservoir of bioactive compounds, offering a promising complement or alternative to synthetic pharmaceuticals, which are often associated with adverse side effects and microbial resistance (World Health Organization, 2019).

The Indian traditional medicine system relies directly on herbs, which are preserved and cultivated by the tribal native communities. These herbs possess medicinal value for treating various diseases, as listed in Ayurveda texts such as Charak Samhita and Sushruta Samhita^[3]. Focusing on the needs of industries like food preservation, cosmetics, agrochemicals, repellents, and pharmaceuticals, two ethnomedicinal plant species were selected: Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*), to analyse their antibacterial and

antioxidant efficacy along with phytochemical analysis.

2. BACKGROUND

Cymbopogon citratus (DC.) Stapf, is a plant of the poaceae family generally named Lemongrass as shown in Fig.1. This perennial aromatic plant is cultivated around the globe, mostly in tropical and subtropical areas, particularly throughout Asia, Africa, and South America. Lemongrass (*C. citratus*) is commonly used in traditional medicine because of its variety of pharmacological effects as an antipyretic, analgesic, anti-inflammatory, and anti-diabetic^[4]. Historically, Lemongrass (*C. citratus*) has gained its therapeutic value due to essential oils' focus in abundance phytochemicals, with the being the most important. Indeed, volatile compounds of Lemongrass (*C. citratus*) essential oil consists primarily of higher levels of oxygenated monoterpenes. Citral, an isomeric mixture of geranial (trans-citral) and neral (cis-citral), is the most abundant component, constituting approximately 50-80% of the oil. Citral is important because it is identifying characteristic scent of Lemongrass and its documented bioactivity effects^[5]. In addition, the phenolic and flavonoid compounds contained within Lemongrass (*C. citratus*) are also credited with potent free radical scavenging anti-oxidant activity by stabilizing ROS and preventing their oxidative damage^[6].



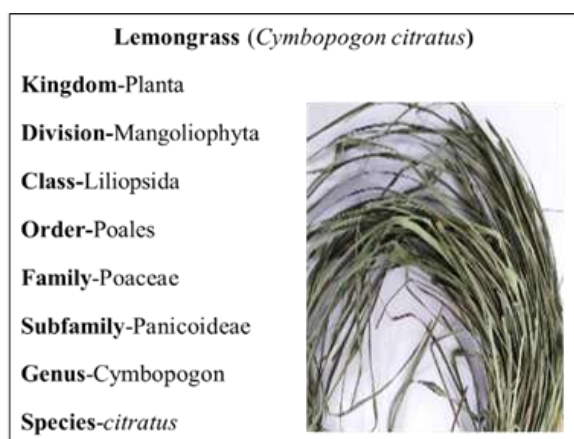


Fig.1 Taxonomic position of *Cymbopogon citratus*

The potent antibacterial effects of Lemongrass EO have been shown to be effective against bacteria, including multidrug-resistant bacteria like Methicillin-Resistant *Staphylococcus aureus* (MRSA) [7]. Potential mechanisms of action include cell membrane disruption, leakage of cellular contents, and biofilm inhibition [8]. The bioactivity of Lemongrass is directly related to the chemical profile, which can differ considerably depending on factors like geographical distribution, growing conditions, harvest time, and methods of extraction [9]. Therefore, accurate chemical characterization is important for the standardization of its therapeutic use.

Eucalyptus globulus (Labill.) is one of the most widely cultivated aromatic trees of the Myrtaceae family commonly named as Tasmanian blue gum globally, prized for its rapid growth and medicinal properties as shown in Fig.2. The essential oil from its leaves, known for its characteristic pungent, camphoraceous aroma, is traditionally used for its decongestant and antiseptic qualities [10]. The primary biological activities of Eucalyptus (*E. globulus*) EO are predominantly attributed to its major volatile constituents. 1,8-Cineole (eucalyptol) is consistently reported as the dominant compound (70-85%) and is chiefly responsible for its renowned antibacterial and anti-inflammatory effects [11]. It is frequently

accompanied by other bioactive monoterpenes like α -pinene, limonene, and p-cymene, which may act synergistically to enhance its overall efficacy [12]

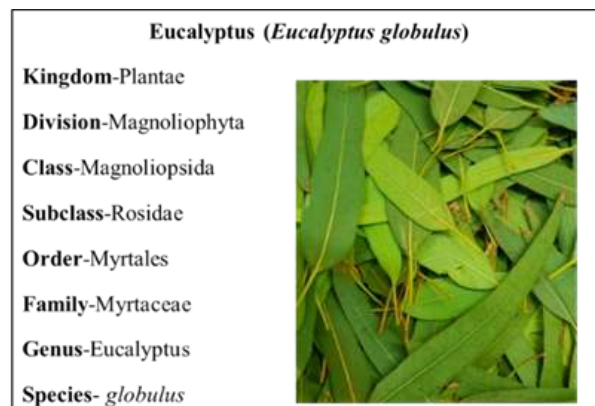


Fig.2 Taxonomic position of *Eucalyptus globulus*

E. globulus EO, with 1,8-cineole as the primary agent for its antibacterial ability, has been shown to act against a wide variety of foodborne pathogens (e.g., *Listeria monocytogenes*, *Escherichia coli*) and food spoilage microorganisms (e.g., *Aspergillus niger*) [13]. The mechanism of antibacterial action involves the disruption of microbial cell membranes, causing cell death. Additionally, the oil has a high antioxidant capacity, which allows it to scavenge free radicals and modulate lipid peroxidation, a primary reason for food rancidity and cosmetic spoilage [14]. In this sense, it is a powerful natural agent to extend the shelf-life of perishable products.

The compelling biological properties of *E. globulus* underpin the significant application potential of its EOs. In food preservation, they can be incorporated into active packaging films or emulsions to create a protective atmosphere against microbial spoilage and oxidation [15]. In cosmetics, their antibacterial and antioxidant qualities are valuable for developing natural preservatives and skincare formulations [16]. Their volatile nature also confers effective repellent activity against insects, offering a natural

alternative to synthetic repellents^[17]. In pharmaceutical formulations, their well-documented anti-inflammatory and antibacterial actions support use in topical antiseptics, inhalants, and wound healing preparations^[18]. However, the chemical profile and bioactivity of Eucalyptus (*E. globulus*) EO are influenced by factors such as geographical origin, season, extraction method, and plant age^[19] making precise chemical characterization an indispensable prerequisite for standardization.

This study, therefore aims to chemically characterize the essential oil extracted from Lemongrass (*C. citratus*), and Eucalyptus (*E. globulus*) using Gas GC-MS and phytochemical screening techniques, and empirically evaluate their antibacterial activity against human pathogens and their antioxidant potential through established assays. By integrating chemical analysis with bioactivity assessment, this research

seeks to signify a concrete scientific basis for the traditional use of these plants and elucidate the relationship between their chemical constitution and biological potential. Ultimately, this work aims to provide a scientific foundation for their upgradation as multi-functional, natural ingredients for innovative applications across food, cosmetic, repellent, and pharmaceutical industries

3. MATERIALS AND METHODS

3.1 Collection of Materials

Plant materials, i.e., leaves of Eucalyptus (*E. globulus*), were collected from the garden of the School of Studies in Physics, Jiwaji University, Gwalior - 474011, Madhya Pradesh, India. Leaves of Lemongrass (*C. citratus*) were procured from the Sizz Pharmaceuticals Pvt. Ltd., Gwalior, Madhya Pradesh, India, as shown in Fig.3.



Fig.3 Dried leaves of (A) *Eucalyptus globulus* and (B) *Cymbopogon citratus*

3.2 Instruments and Chemicals

Hydro-Distillation Clevenger Apparatus (Borosilicate glass) and Heating Mantle (Ambassador), Refrigerator (LG), and GC-MS (Agilent Technologies, Inc.) were used in experiments. All Chemicals used were of analytical grade.

3.3 Extraction of Essential Oils

Conventional hydro-distillation method was used to extracted Essential oils (EOs) from plant materials^[20]. The collected plant materials were washed, dried, and ground into a powder. One hundred grams (100 g) of the dried sample was mixed with approximately 800 ml. of distilled water in a Clevenger apparatus. The distillation was performed for 4 hours under optimal operational conditions at a temperature of approximately 40°C as shown in Fig.4. The

extracted EOs was collected and dehydrated using anhydrous Na_2SO_4 . The percentage yield of EOs was calculated as follows

$$\text{Yield \%} = \frac{\text{weight of extracted essential oil}}{\text{weight of sample used}} \times 100$$



Fig.4 Hydrodistillation Extraction of Essential Oil

3.4 Chemical Characterization of Essential Oils

The chemical profiling of EOs, viz., saponification value, acid values, ester value, and free fatty acid, were analyzed using the standard methods American Oil Chemists' Society (AOCS)

Determination of Saponification Value:

To estimate the saponification value of the essential oil, 0.5 ml of the sample was taken in a 50 ml conical flask. to this, 10 ml of 0.5 M ethanolic KOH was added and the mixture was stirred for approximately 1hr. then excess alkali was titrated against 0.5 M HCl using phenolphthalein as an indicator, until the pink color disappeared. The saponification value of the EOs was calculated using the following formula [21].

$$\text{SV} = \frac{(S-B) \times M \times \text{Molecular Weight of KOH}}{\text{volume of sample (ml)}} \text{mgKOH/g}$$

Where, S = Sample Titration Value, B = Blank Titration Value, M = Molarity of HCL, Molecular Weight of KOH=56.1mg

Determination of Acid Value:

For the estimation of acid value, 0.5 ml of essential oil was taken in a conical flask containing 50 ml of ethanol. To this mixture, 2–3 drops of phenolphthalein indicator were added. The solution was then titrated against 0.1 N KOH until a persistent pink color appeared. The volume of KOH consumed during titration was recorded, and the acid value of the EOs was calculated using the following formula [22].

$$\text{AV} = \frac{\text{Molecular weight of KOH} \times \text{Normality of KOH}}{\text{volume of sample (ml)} \times \text{titre value}} \text{mgKOH/g}$$

Where the Molecular weight of KOH=56.1mg, the Normality of KOH=0.1N

Determination of Free fatty acid:

One gram of essential oil was taken in a beaker and gently warmed. To this, 25 ml of methanol was added and the mixture was stirred thoroughly. Subsequently, 2 drops of phenolphthalein indicator and 1 drop of 0.14 N NaOH were introduced. The sample was titrated against the

NaOH solution until a light pink color appeared and persisted for about one minute. The endpoint reading was recorded and calculate the free fatty acid (FFA) using the formula equation^[23].

$$\text{FFA \%} = \text{Acid value} \times \frac{1}{2}$$

Determination of Ester Value:

The ester value of the oil was calculated as the difference between its acid value and saponification value. It is expressed using the following formula:

$$\text{Ester Value (mg KOH/g)} = \text{Saponification Value} - \text{Acid Value}$$

The ester value provides an estimate of the amount of esterified fatty acids present in the oil. A higher ester value generally indicates a greater proportion of esters, which is important for assessing the quality, stability, and potential industrial application of the oil^[24].

3.5 Characterization of EOs through GC-MS

The chemical constituents of the extracted EOs were analyzed using Gas Chromatography–Mass Spectrometry (GC-MS) equipped with an HP-5ms capillary column (30 m × 250 µm × 0.25 µm)^[25,26]. The total run time was 36.7 minutes. Identification of compounds was carried out by comparing their retention times and mass spectra with data from the Wiley and NIST libraries, as well as with published literature and reference standards of known constituents.

3.6 Antibacterial activities of extracted Essential oils

The Antibacterial efficacy of the extracted EOs was evaluated using the agar well diffusion method on Mueller-Hinton Agar (MHA) plates. A comprehensive and reliable assessment of EOs'

antibacterial efficacy was ensured against both types of bacterial cell structures: Gram-positive and Gram-negative bacterial cell culture of *Staphylococcus aureus* (Gram-positive) and *Escherichia coli* (Gram-negative). Each strain was inoculated separately into Mueller-Hinton Broth (MHB) and incubated at 37 °C until the turbidity matched 0.5 McFarland standards, corresponding to approximately 1.5×10^8 CFU/ml. The prepared inoculum was then lawn-cultured onto MHA plates. For the assay, 6 mm size wells were created in the agar using a sterile cork borer. Different concentrations of the essential oils (25 µl, 50 µl, and 100 µl) were added into the wells, while Gentamycin (5 µl) was used as a standard control. The plates were kept at room temperature for 15 minutes to allow diffusion of the oils, followed by incubation at 37 °C for 18–24 hours. After incubation, Antibacterial activity was evaluated by measuring the diameter (in mm) of the clear inhibition zones around the wells^[27].

3.7 Antioxidant Activity of EOs through DPPH Assay

The antioxidant potential of EOs of Lemongrass (*C. citratus*), and Eucalyptus (*E. globulus*) was assessed by DPPH assay using ascorbic acid as the standard. 2,2-diphenyl-1-picrylhydrazyl (DPPH) is stable (in powder form) free radical with purple color which turns yellow when scavenged. The DPPH assay uses this character to show free radical scavenging activity. Antioxidants react with DPPH to reduce it to DPPH-H as the consequence absorbance decreases^[28].

Reagent preparation: A stock solution of 0.1 M DPPH was prepared by dissolving 4 mg of DPPH in a 100 ml volumetric flask and making up the volume with methanol. For the standard, 20 mg of ascorbic acid was dissolved in 20 ml of distilled water to obtain a concentration of 1 mg/ml. Similarly, 10 mg of essential oil was dissolved in



10 ml of methanol to prepare a solution with a concentration of 1 mg/ml. additionally, 80% methanol was prepared by mixing 80 ml of methanol with 20 ml of distilled water.

DPPH assay procedure: Serial dilutions of the ascorbic acid stock solution were prepared to obtain different concentrations: 250, 200, 150, 100, and 50 µl. Similarly, essential oil solutions were diluted to the same volumes (250, 200, 150, 100, and 50 µl). To each test tube, 80% methanol was added to bring the final volume to 1 ml. Subsequently, 1 ml of DPPH solution was added to all test tubes. The mixtures were incubated in 30 minutes in the dark, and the spectrophotometer was used to calculate the absorbance at 517MM.

Calculation of %inhibition activity or % Radical Scavenging Activity (RSA) calculated by using the formula:

$$\frac{c - A}{c} \times 100$$

Where, C is absorbance of control, A is absorbance of sample

3.8 Statistical Analysis

The results of antimicrobial and antioxidant Activity of Extracted Essential Oils were statistically analyzed by One-Way ANOVA followed by post hoc Dunnett's test with a $p < 0.05$ significance level. All of the statistical analyses were performed with GraphPad Prism Version 8.0.2 (263) and MS Excel 2019.

4. RESULTS

4.1 Yield of Extracted Essential Oils

The average yield of EOs of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*) was 0.6 % and 1 % (w/w), respectively. Eucalyptus (*E.*

globulus) EOs showed a higher percentage of yield in comparison to EOs of Lemongrass (*C. citratus*), as shown in Table 1.

Table 1 Yield % of EOs of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*)

Plants	Plants dried leaves powdered used (g)	Extracted Essential Oil (g)	% Yield
Lemongrass	100	0.6	0.6%
Eucalyptus	100	1	1%

4.2 Physicochemical Analysis of Essential Oils

The physicochemical Analysis of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*) EOs, as shown in Table 2 and Table 3 revealed that initially Eucalyptus EOs were colorless, but after about 2 weeks turned yellow in color. Both the EOs were soluble in acetone, hexane but insoluble in water.

Table 2 Physical properties of EOs of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*)

Sr. No.	Physical properties	Lemongrass (<i>C. citratus</i>)	Eucalyptus (<i>E. globulus</i>)
1	Color	Light brown	Clear light yellow
2	Odour	Minty, woody, pungent	Much like lemon
3	pH	5.5	6
4	Solubility	Acetone, Hexane	Acetone, Hexane

Table 3 Chemical properties of EOs

Sr. No.	Chemical properties	Lemongrass (<i>C. citratus</i>) (Mean ± SEM)	Eucalyptus (<i>E. globulus</i>) (Mean ± SEM)
1	Acid Value (mg KOH/g)	4.5 ± 0.2	1.1±0.1
2	Saponification Value (mg KOH/g)	140.2 ± 4.6	258.0 ± 7.5
3	Ester value (mg KOH/g)	135.7 ± 2.8	256.9 ± 1.2
4	Free Fatty Acid (%)	2.2 ± 0.1	0.6 ± 0.0



4.3 Antibacterial Activity of Essential Oils

Essential Oils of Lemongrass (*C. citratus*) showed higher antibacterial activity against *Staphylococcus aureus* (with 30.0 ± 1.2 mm Zone

of Inhibition), as shown in Table 4 and Fig.5, and Eucalyptus (*E. globulus*) EOs showed higher antibacterial activity against *Escherichia coli* (with 30.0 ± 1.2 mm Zone of Inhibition), as shown in Table 5 and Fig.6.

Table 4 Antibacterial activities of EO of Lemongrass

Antibacterial activities				
Zones of Inhibition (Mean $\pm$ SEM in mm) against selected bacteria				
Microbial Species	Gentamicin Conc.	Lemongrass (<i>C. citratus</i>) EOs Conc.		
	5 μ l	25 μ l	50 μ l	100 μ l
<i>Staphylococcus aureus</i>	34.0 ± 0.6	16.0 ± 0.6	18.0 ± 0.6	30.0 ± 1.2
<i>Escherichia coli</i>	36.0 ± 1.2	22.0 ± 1.2	22.0 ± 0.6	24.0 ± 0.6

Table 5 Antibacterial activities of EO of Eucalyptus

Antibacterial activities				
Zones of Inhibition (Mean $\pm$ SEM in mm) against selected bacteria				
Microbial Species	Gentamicin Conc.	Eucalyptus (<i>E. globulus</i>) EOs Conc.		
	5 μ l	25 μ l	50	100 μ l
<i>Staphylococcus aureus</i>	32.0 ± 0.6	14.0 ± 1.2	16.0 ± 1.2	22.0 ± 1.2
<i>Escherichia coli</i>	36.0 ± 1.2	16.0 ± 0.6	20.0 ± 0.6	30.0 ± 1.2



(A)

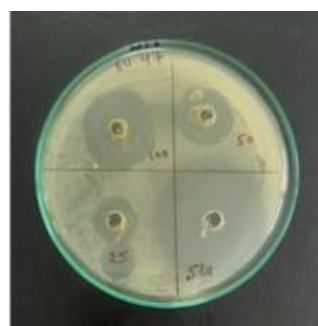


(B)

Fig.5 Antimicrobial activities of extracted EOs of Lemongrass (*Cymbopogon citratus*)
(A) *Staphylococcus aureus* (gram positive), (B) *Escherichia coli* (gram negative)



(C)



(D)

Fig.6 Antimicrobial activities of extracted EOs of Eucalyptus (*Eucalyptus globulus*)
(C) *Staphylococcus aureus* (gram positive), (D) *Escherichia coli* (gram negative)

4.4 Antioxidant Activity of Essential Oils

The antioxidant activity of Essential oils Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*) was assessed by DPPH assay using

ascorbic acid as a standard. % Inhibition of Lemongrass (*C. citratus*) was found to be higher than that of Eucalyptus (*E. globulus*) and lower than the standard, as shown in Table 6 and Fig.7.

Table 6 Radical Scavenging Activity (RSA) of EOs of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*)

Sr. No.	Concentration (μ l)	% RSA of Ascorbic Acid	% RSA of Lemongrass	% RSA of Eucalyptus
1	50	95.2 $\pm$ 1.2	71.4 $\pm$ 0.5	12.8 $\pm$ 1.6
2	100	96.1 $\pm$ 0.4	71.6 $\pm$ 0.4	14.2 $\pm$ 0.4
3	150	95.5 $\pm$ 0.2	73.2 $\pm$ 0.5	20.3 $\pm$ 4.0
4	200	96.7 $\pm$ 0.1	74.6 $\pm$ 0.4	30.5 $\pm$ 0.3
5	250	97.4 $\pm$ 0.1	77.6 $\pm$ 0.6	47.5 $\pm$ 0.4

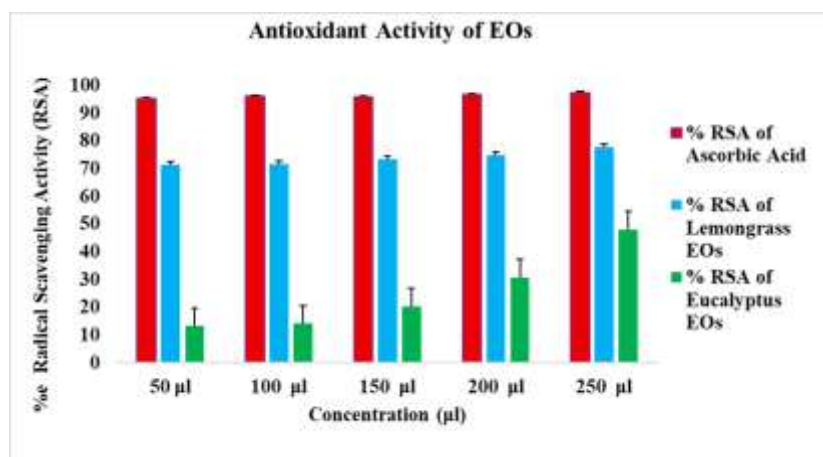


Fig.7 Graph Showing the comparative % Radical Scavenging Activity (RSA) of EOs

4.5 Phytochemical Analysis through GC-MS

GC-MS Chromatogram of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*) EOs contains various volatile compounds and were identified from Willey and NIST library as shown in Fig.8, Fig.9, Fig.10 and Fig.11, Table 7 and

Table 8. These chemical compounds are tabulated along with peak, retention time, and percent composition area for each EOs. A total of 24 chemical compounds were identified in Lemongrass (*C. citratus*) and 22 in Eucalyptus (*E. globulus*).

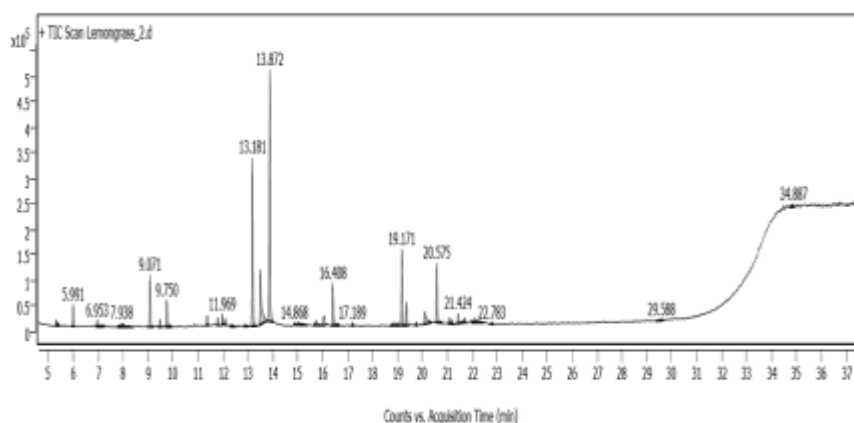
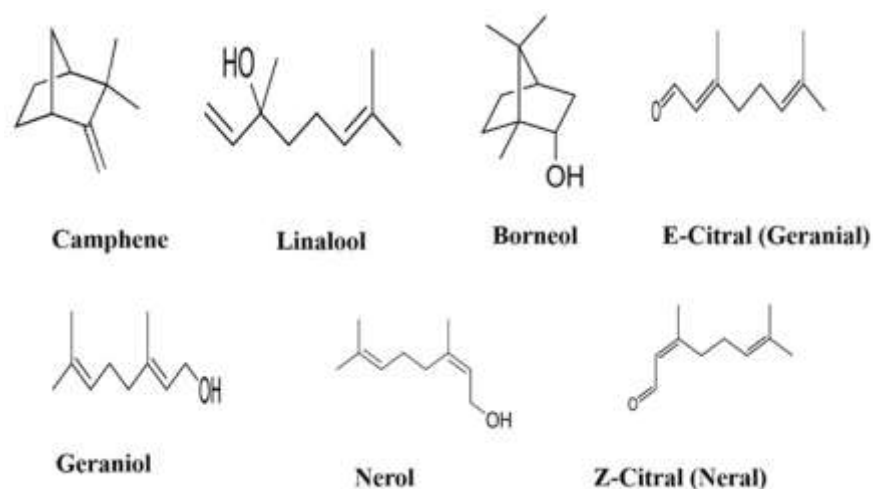


Fig.8 GC-MS Chromatogram of Lemongrass (*C. citratus*)



Fig.9 Chemical structure of volatile compounds of EO of Lemongrass (*C. citratus*)Table 7 Compounds Identified through GC-MS of Extracted EOs of Lemongrass (*C. citratus*)

Peak No.	Retention time (min)	Compound	Area %
1	5.311	Propanamide	0.62
2	5.991	Camphene	1.91
3	6.953	N-(3-Methylaminopropyl)-N-methylformamide	0.58
4	9.071	4-nonanone	4.81
5	9.467	1,2-Benzenediol, 4-[1-hydroxy-2-(methylamino)ethyl]-, (R)	0.83
6	9.75	L-linalool	2.55
7	11.358	1-borneol	1.59
8	11.788	Pyridinium	0.97
9	11.969	2-butanamine	1.28
10	12.083	4H-Pyran-4-one	0.95
11	13.181	Z-citral	16.35
12	13.498	Geraniol	11.38
13	13.872	E-citral	26.32
14	15.004	Methylpent-4-enylamine	0.61
15	15.717	2-hexanamine	1.04
16	16.068	2,4-dimethylamphetamine	1.55
17	16.408	Nerol (2,6-Octadien-1-ol, 3,7-dimethyl-, (Z))	4.46
18	19.171	Naphthalene	7.65
19	19.352	Delta-cadinene	2.41
20	20.088	Propanamide	3.73
21	20.575	Caryophyllene oxide	5.95
22	21.084	2-Pyridinepropanoic acid	0.68
23	21.424	Benzeneethanamine	1.20
24	21.696	2-hexanamine	0.56

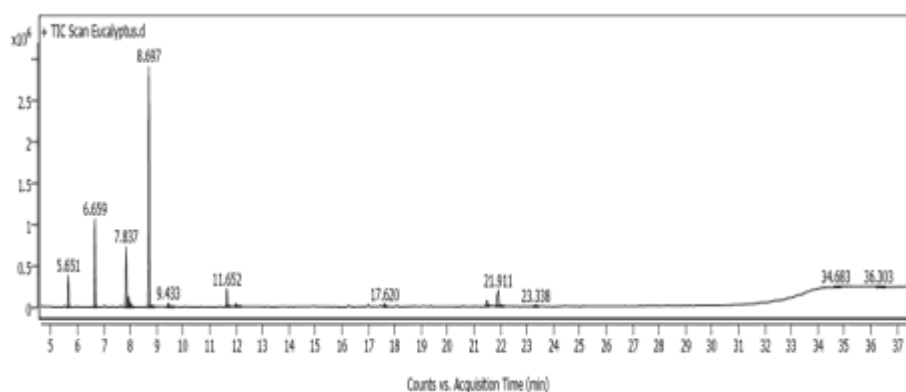


Fig.10 GC-MS Chromatogram of Eucalyptus (*E. globulus*)

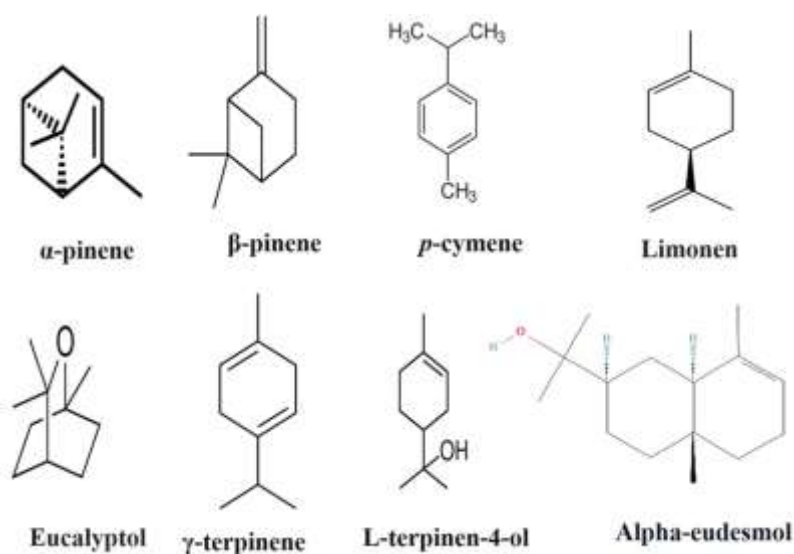


Fig.11 Chemical structure of volatile compounds of EO of Eucalyptus (*E. globulus*)

Table 8 Compounds Identified through GC-MS of Extracted EOs of Eucalyptus (*E. globulus*)

Peak No.	Retention time (min)	Compound	Area %
1	5.651	α -pinene	5.60
2	6.659	β -pinene	16.21
3	7.021	Beta-myrcene	0.37
4	7.633	α -terpinene	0.26
5	7.837	Benzene, 1-methyl-4-(1-methylethyl)-	11.07
6	7.938	D-limonene	1.94
7	8.018	Eucalyptol	0.65
8	8.697	γ -terpinene	45.81
9	9.433	Alpha-terpinolene	0.78
10	11.652	L-terpinen-4-ol	4.16
11	11.981	3-carene	0.72
12	16.261	3-ethoxyamphetamine	0.29
13	16.997	N-Acetyl p-ethoxyamphetamine	0.41
14	17.62	Tocainide	0.63
15	18.084	Benzenemethanol	0.40
16	20.586	Pterin-6-carboxylic acid	0.27
17	21.492	Delta-selinene	2.04
18	21.911	Alpha-eudesmol	6.28

19	23.338	2H-Azepin-2-one, hexahydro-1-methyl-	0.47
20	28.32	2-heptanol	0.26
21	31.887	Methylpent-4-enylamine	0.63
22	32.883	Cyclotrisiloxane	0.36

5. DISCUSSION

In this study, essential oils (EOs) were extracted from the dried leaves of Lemongrass (*C. citratus*) and Eucalyptus (*E. globulus*) through hydrodistillation. Their chemical compositions were analyzed using GC-MS, and their biological activities were tested. The yield of oil extraction after four hours of distillation was found to be higher in Eucalyptus (*E. globulus*) (1% w/w) compared to Lemongrass (*C. citratus*) (0.6% w/w). Lemongrass (*C. citratus*) EOs showed the highest Antibacterial activity against *Staphylococcus aureus* with 30.0 ± 1.2 mm Zone of Inhibition in (Fig.5). Eucalyptus (*E. globulus*) EOs showed the highest Antibacterial activity against *Escherichia coli* with 30.0 ± 1.2 mm Zone of Inhibition as shown in (Fig.6).

Radical scavenging activity (RSA) of ascorbic acid, Lemongrass (*C. citratus*), and Eucalyptus (*E. globulus*) extracts at different concentrations (50–250 μ l) was measured. Ascorbic acid, used as the standard, consistently exhibited the highest antioxidant activity (94–96%) across all concentrations, showing its strong and stable free radical scavenging potential even at lower doses. Among the extracts, Lemongrass (*C. citratus*) demonstrated moderate activity, increasing gradually from about 70% at 50 μ l to nearly 77% at 250 μ l, indicating a clear dose-dependent effect. In contrast, Eucalyptus (*E. globulus*) showed the lowest RSA, starting from 12–14% at lower concentrations and rising steadily to 47% at 250 μ l, suggesting a weaker antioxidant capacity that improves with increasing concentration. Overall, the findings highlight ascorbic acid as the most

potent antioxidant, followed by Lemongrass (*C. citratus*) as a promising natural alternative, while Eucalyptus (*E. globulus*) exhibited comparatively lower but concentration-dependent activity.

Based on GC-MS analyses, the chemical profiles of Lemongrass and Eucalyptus (*E. globulus*) essential oils revealed significant potential for multi-functional biological activity, including antibacterial effects against both Gram-positive *Staphylococcus aureus* and Gram-negative *Escherichia coli*, potent antioxidant properties, and effective mosquito repellency. The Lemongrass (*C. citratus*) oil is characterized by high concentrations of citral isomers (over 42% combined) and geraniol (11.38%), compounds widely documented for their antibacterial activity through membrane disruption and strong free-radical scavenging capacity^[29]. Similarly, Eucalyptus (*E. globulus*) oil demonstrates a composition rich in γ -terpinene (45.81%), β -pinene (16.21%), and p-cymene (11.07%), all recognized for their broad-spectrum antibacterial and antioxidant actions.^[30,31,32] Importantly, key components in both oils—particularly geraniol, citral, limonene, and pinene derivatives are well established in scientific literature as effective mosquito repellents, interfering with insect host-seeking behavior^[33]. This synergistic combination of bioactive compounds positions these essential oils as promising natural agents for pharmaceutical, preservative, and vector control applications

6. CONCLUSION

The present study shows that essential oils extracted from Lemongrass (*C. citratus*) and



Eucalyptus (*E. globulus*) are potent safer and biologically effective agents. Lemongrass oil, rich in citral and geraniol, exhibited stronger antibacterial and antioxidant properties, making it a promising natural alternative to synthetic drugs. Eucalyptus (*E. globulus*) oil, although yielding higher extraction efficiency and showing notable antibacterial activity against *E. coli*, displayed comparatively weaker antioxidant potential but still demonstrated valuable bioactivity. Both oils contained compounds (e.g., citral, geraniol, limonene, and pinene derivatives) recognized for their insect-repellent properties, further supporting their multifunctional role. Overall, the synergistic chemical composition of these essential oils highlights their potential as safer, plant-based alternatives for antibacterial, antioxidant, preservative, and mosquito control applications.

ACKNOWLEDGEMENT

The authors express their gratitude to the late Professor GBKS Prasad of Jiwaji University for his enduring support and motivation. They also extend thanks to the Test and Evaluation Division, Defence Research & Development Establishment (DRDE), Gwalior for granting access to their GC-MS equipment. Furthermore, acknowledgment is given to the Department of Botany at S. M. S. Government Model Science College, Gwalior and to Jiwaji University, Gwalior for supplying the necessary infrastructural resources

FUNDING

Ajay Kumar Ahirwar – (Author) awarded SC/ ST Ph. D. Research Fellowship of Department of Higher Education, Bhopal, Madhya Pradesh, India, for conducting this research work.

CONFLICT OF INTEREST

The authors have no conflict of interest to declare.

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HOW TO CITE: Ajay Kumar Ahirwar, Jyoti Sharma, Indu Gautam, Madhav Varshney, Garima Sharma, Suman Jain, Vinod Kumar Sewariya, Comparative Evaluation of Antibacterial and Antioxidant Efficacy of Essential Oils of *Cymbopogon citratus* and *Eucalyptus globulus*, *Int. J. of Pharm. Sci.*, 2026, Vol 4, Issue 1, 691-706. <https://doi.org/10.5281/zenodo.18186237>

