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ORIGINAL ARTICLE

Analysis of chemical composition and biological efficiency of leaf essential oils isolated from seven species of *Cinnamomum* of the Western Ghats, India

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Abstract
The chemical profiles of essential oils (EOs) of 7 *Cinnamomum* species (*C. sulphuratum*, *C. perotettii*, *C. verum*, *C. wightii*, *C. camphora*, *C. glanduliferum*, and *C. malabatrum*) composed from assorted parts of Western Ghats were analyzed. The antioxidant, antibacterial, and larvicidal potentialities of 7 essential oils were also investigated. *Cinnamomum verum* was observed to have the maximum yield (1.96%) of EO followed by *C. malabatrum* (1.71%). GC-MS analysis of EOs reported the highest amount of eugenol (38.04%), shyobunol (13.31%) in *C. verum*, whereas *C. perrottetii* identified 14–42 phytocompounds including terpenes, sesquiterpenes, monoterpenes, diterpenes, and phenylpropanoids, esters, aldehydes, and fatty acids. In the antibacterial activity, the higher zone of inhibition of *C. malabatrum* showed against *S. pyogenes* (28 ± 2.5 mm) and *E. faecalis* (27 ± 1.24 mm) followed by *C. verum* has shown notable inhibition zone against *K. pneumoniae* (26 ± 1.15 mm) and *S. flexneri* (24.33 ± 0.88 mm), with MIC value against *C. verum*, showed MIC value (1.17 ± 0.34 µg/mL) against *S. flexneri, S. pyogenes,* and *K. pneumonia*. All the *Cinnamomum* oil samples exhibited appreciable antioxidant radical scavenging activities with IC_{50} values ranged 10.83–15.06 µg/mL. All the oil samples showed promising larvicidal activity against the late-third instar larvae of *Aedes aegypti*. Extending the present study with in vivo and in vitro animal studies will confirm the potentiality of *Cinnamomum* oils in the field of biomedicine to detect novel medicines.

Keywords *Aedes aegypti* · DPPH · GC-MS · *Cinnamomum verum* · IC_{50}

1 Introduction

Cinnamomum is reported to have higher medicinal value and larvicidal properties which can be considered a commercial cultivable species for the rural economy [1]. EOs are volatile lipophilic compounds secreted through aromatic plant cells with diverse morphology, synthesized through numerous plant organs such as buds, branches, stems, roots, berries, seeds, flowers, bark, and wood, and stored in secretory cells, channels, cavities, trichomes, or epidermal cells [2]. The secretory cells range from highly specialized to non-specialized cells, osmophores, ducts, and cavities reported in dissimilar plant parts are the repositories of Eos [3]. EOs is short-molecular-weight compound mixtures with a strong odor obtained from aromatic plants [4, 5]. They are the mixtures of a large number of compounds having differentiated structures mainly of terpenes or phenylpropanoid derivatives. EOs is significant derived metabolites reported in aromatic and medicinal plants used in fields and industries such as pharmaceuticals, cosmetics, food, and aromatherapy [6]. EOs obtained from plant sources are the best alternative to synthetic oils used in human health care. They are also reported as the best food additives to retain the quality and value of nutrition [7]. Moreover, spice and aromatic plants are reported to be prosperous sources of

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volatile compounds. Many of these secondary metabolites are approved for human consumption through the United States Food and Drug Administration, widely used in flavors and fragrances, food preservation, and pharmaceutical industries [8].

The different compounds in EO samples are recognized to have acquired an assortment of biological significance, which capacity has been accountable for therapeutic consequences such as insecticidal, anti-allergic, anti-inflammatory, antioxidant, and anticancer properties, along with their antimicrobial characteristics [9–11]. Multidrug resistance and side effects of antibiotics increased the interest and need for developing new antimicrobial agents from plants [12].

The antimicrobial activity of EO is also proposed to the morphological changes produced in bacteria by the volatile compounds, which modulate bacterial growth and are capable of trapping the free radicals and preventing oxidation [8]. The essential oils from cinnamon improve the activity of antioxidant enzymes by reducing the reactive oxygen-free radical [4]. Many studies on EO reported that the antioxidant activity is unpaid to the existence of several monoterpenes and sesquiterpene compounds [13, 14]. By the action of an antibacterial representative on bacteria, their ROS content increases. When it exceeds the antioxidant defense ability, the protein and further intracellular substances of the bacteria get injured and hence affect the metabolic process of the bacteria [15]. Also, the EOs of several plant species have been documented to exhibit toxicity effects against a few species of mosquitoes. Only substantial numbers of studies have been attempted on the usage of EOs of cinnamon as food additives and medicines. The comparative analysis of volatile composition among the species helps to understand the medicinal values of the EO-yielding species of a group [16].

The genus *Cinnamomum* contains shrubs and evergreen trees belonging to the Lauraceae family consisting of about species are 250 in subtropical and tropical regions, mainly in Asia, and a few in central and South America and Australia [17]. The bark of *Cinnamomum* is widely employed as a spice and denoted as a highly valued oldest spice which ranks second next to pepper and is commonly used for flavoring, preservation, cosmetics, and perfumery industries and in preparation of various ayurvedic and allopathic medicines [18]. The EOs from the species of *Cinnamomum* are reported as an important source of chemicals with pharmaceutical uses and hence, current investigate works were alerted on *Cinnamomum* ([19, 20]. The plants of this genus are reported with numerous pharmacological properties; antioxidant, antibacterial, and anti-inflammatory properties [21, 22]. The occurrence of bulky amounts of phytoconstituents such as camphor, α-terpineol, linalool, eucalyptol, and borneol in the EO is accountable for their biological activities [20]. The EOs of *Cinnamomum* are reported to be carminative and expectorant; the bark oil is an anthelmintic, aphrodisiac, and tonic and also useful in treating bronchitis, parched mouth, itching, diarrhoea, piles, flatulence, headache, heart, urinary diseases, etc. [23].

The current study of seven *Cinnamomum* species *C. verum*, *C. sulphuratum*, *C. perrottetii*, *C. camphora*, *C. wightii*, *C. glanduliferum*, and *C. malabatrum* selected and focused for the identification of secondary metabolites in essential oils by gas chromatography mass spectroscopy (GC-MS), extracted essential oils targeted pharmacological applications like free radical scavenging assay by DPPH method, antibacterial activity against two gram-positive bacteria, such as *Streptococcus pyogenes* and *Enterococcus faecalis* and four gram-negative bacteria, *Pseudomonas aeruginosa*, *Enterobacter aerogenes*, *Klebsiella pneumoniae*, and *Shigella flexneri* human pathogenic bacteria and larvicidal activity against *Aedes aegypti*.

2 Materials and methods

2.1 Plant materials

Fresh leaves of seven species of *Cinnamomum*, *Cinnamomum verum* J. Presl., *Cinnamomum sulphuratum* Nees., *Cinnamomum perrottetii* Meisn., *Cinnamomum camphora* (L.) J. Presl., *Cinnamomum wightii* Meisn., *Cinnamomum glanduliferum* (Wall.) Meisn., and *Cinnamomum malabatrum* (Burm.f.) Presl. were composed of dissimilar forest areas of the Western Ghats. The collected leaf specimens were properly identified with standard journalism and authenticated with official voucher specimens (Fig. 1). The receipt specimens of all the studied species (*Cinnamomum verum* J. Presl.: GUD 942, *C. sulphuratum* Nees.: GUD 943, *C. perrottetii*, Meisn.: GUD 944, *C. camphora* (L.) J. Presl.: GUD 945, *C.wightii* Meisn.: GUD 946, *C. glanduliferum* (Wall.) Meisn.: GUD 947, and *C. malabatrum* (Burm.f.) Presl.): GUD 948) were placed in the herbarium (GUD) of the Gandhigram Rural Institute (Deemed to be University), Department of Biology, Gandhigram, Dindigul (DT), Tamil Nadu.

2.2 Isolation of essential oils

The fresh leaves (200–500 g) of all the species of *Cinnamomum* were cut into small pieces (2–3 cm) individually and subjected to hydro-distillation with the Clevenger-type equipment (Borosilicate Glass) for about 4–6 h at ambient temperature (60–80 °C), the yield of EOs was calculated. The EOs collected from each species were dried over anhydrous Na_2SO_4 and then stored at 4 °C in a dark place for further analysis and further work [12, 13].

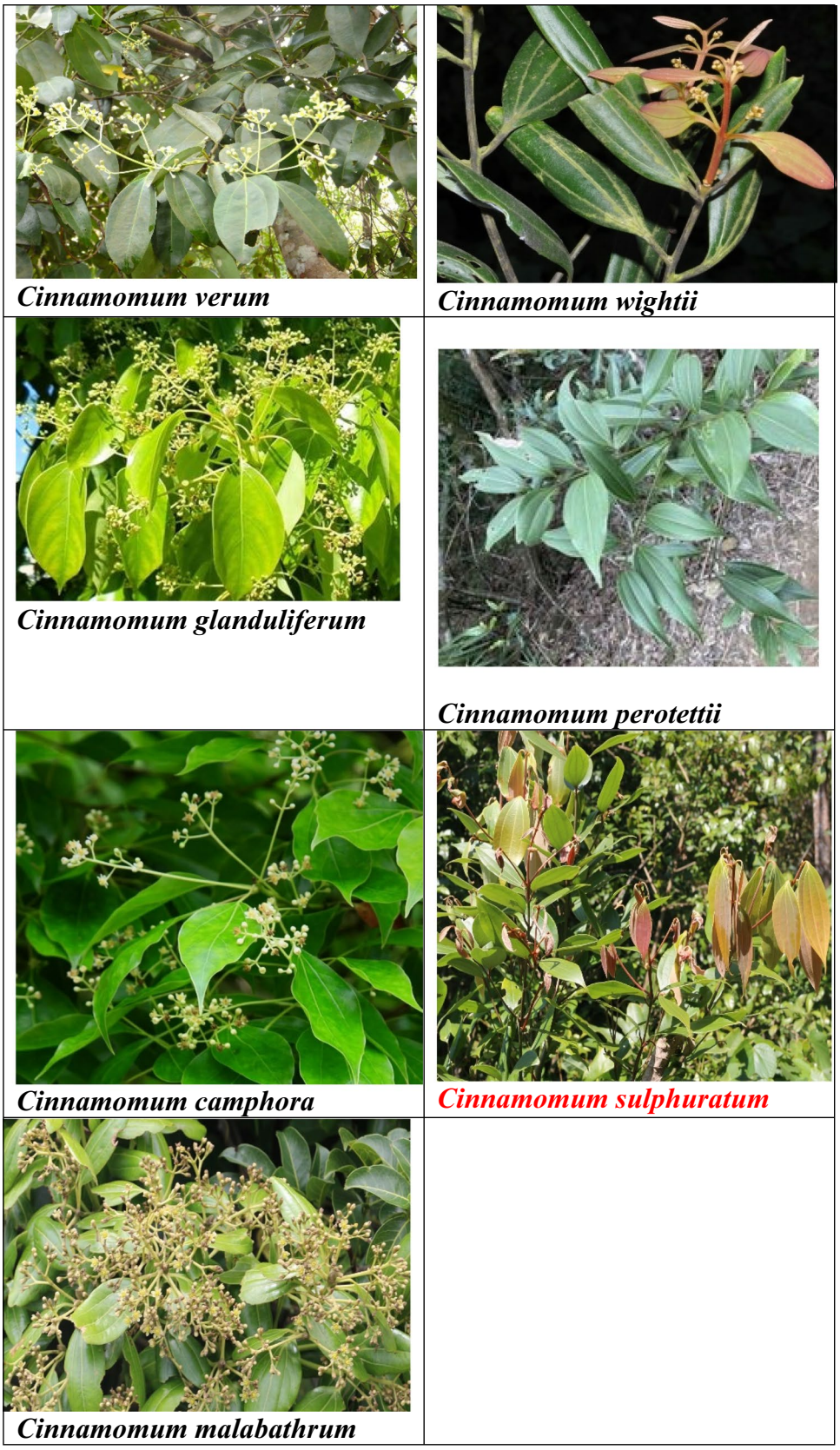

Fig. 1 List of *Cinnamomum* species selected for the investigation of essential oils

2.3 Analysis using gas chromatography and mass spectrometry (GC-MS)

The essential oil samples were diluted in n-hexane (10 µL/300 µL), and 10 µL of each solution was added to a split-splitless injector attached to an HP-5 column (30 m × 0.32 mm, film thickness 0.25 µm), which was used in a spit-mode gas chromatography experiment on an Agilent Technologies 7890 apparatus. As a carrier gas, helium (1 mL/min/210 °C) was employed. The column temperature was linearly programmed from 40 to 260 °C at 4 °C/min, while the injector and detector temperatures were set at 250 and 280 °C, respectively. Without using any correction variables, the percentage composition was calculated from the peak areas. The HPG 1800 C Series II GCD analytical instrument with an HP-5MS column was used to perform the GC-MS. Re-recorded mass spectra in the m/z 40–450 range were done so in EI mode (70 eV). By calculating the oils' Kovats retention index (RI) and comparing their mass spectra to reference compounds from the Nirst and Willey libraries, the volatile components of the oils were located [12].

2.4 Antibacterial activities of essential oils

Two gram-positive bacteria, such as *Streptococcus pyogenes* (MTCC 442) and *Enterococcus faecalis* (MTCC 439), and four gram-negative bacteria, *Pseudomonas aeruginosa* (ATCC 27853), *Enterobacter aerogenes* (MTCC 111), *Klebsiella pneumoniae* (MTCC 432), and *Shigella flexneri* (MTCC 1457) were used. The microbial cultures were obtained from the Laboratory of the Department of Biology, The Gandhigram Rural Institute, Dindigul, India. The diameter of the zone of inhibition of EOs was evaluated based on the qualitative determination of the antibacterial activity of essential oils [14]. Different concentrations (50–150 µg/mL) of sample solutions were added (50–90 µL) to each well, and the plates were incubated at respective temperatures of 37 °C for 24 h. Gentamicin (Laborate Pharmaceuticals India Ltd.) (10 µg/mL) and 1% DMSO (SRL chemical) were used as the experimental +ve and −ve controls. The triplicates were maintained, and the inhibition zone diameter (IZD) was observed and calculated for every organism and expressed the value as mean ± S.E. [14].

2.5 MIC (minimal inhibitory concentration) of EOs determination

The testing of MIC, seven essential oil samples selected, and the bacterial strains was determined through the method of microdilution as prearranged through the CLSI method by using 96-well microtiter plates (Tarsons trust delivered, Cat. No. 980040) with some modifications [24–26], minimal concentrations of essential oil samples that inhibit or kill the microorganisms were resoluted through the dilution of broth method. Each EO was subjected to dilution to 1 mg/mL for testing, followed by a series of two-fold dilutions from 150 to 0.29 µg/mL. Gentamicin (Laborate Pharmaceuticals India Ltd.) (1 mg/mL) and DMSO (SRL chemical) (100%) were taken as positive and negative controls respectively. The early bacterial inoculums were attuned to 5 × 106 CFU/mL and in a sterile 96-well microplate, nutrient broth (95 µL) as well as respective pathogenic culture (1×10^5 cells/ mL), were added into each well. The oil samples 100 µL were added to consecutive wells and incubated at 37 °C for 18 h. For MIC determination, resazurin (10 µL, 0.02% (w/v)) was transferred to each well followed by 2–4 h of further incubation. Incubated then wells were subjected to illustration evaluation of color changes that imply bacterial growth. The color changes indicated resazurin (HiMedia) reduction (blue/purple) to resorufin (pink). The 570 nm absorbance was read by Robonik Readwell Touch Automatic ELISA plate analyzer. The low concentration at which absolute inhibition of MIC values was recorded; all the experimental trials were performed in triplicates [16].

2.6 Antioxidant activities of Eos

The capability of the DPPH radical scavenging assay of EO was also analyzed spectrophotometrically. DPPH (0.1 mM; 1 mL) solution was added to different concentrations (10–50 µg/mL) of sample solution and then incubated for about 30 min. The reaction mixtures were read by a spectrophotometer (GENESYS 180 UV – Visible spectrophotometer) at 517 nm. The standard ascorbic acid (Sigma-Aldrich) in various concentrations (10–50 µg/mL), the DPPH radical scavenging activity was calculated against sample concentrations, and IC_{50} values were obtained from the graph [24, 27–29].

$$\text{Scavenging activity}(\%) = \left[\frac{\text{Control OD} - \text{sampleOD}}{\text{Control OD}}\right] x100$$

2.7 Larvicidal bioassay

The larvicidal assay was experimented on the 3rd instar larvae of *Aedes aegypti* as per the methodology of WHO with minor modifications [30]. The larval colonies of *A. aegypti* were collected from ICMR, Madurai, CRME (Centre for Research in Medical Entomology). Dissimilar test concentrations (50–250 µg/ml) from the stock essential oils were prepared with methanol (0.1 mM) (HiMedia). The appropriate volume of methanol dissolved in distilled water has served as a negative control. Ten healthy 3rd instar larvae were released into every glass beaker containing the respective examination solution. The controls tested were also done in a similar with each duplicate. The death rate of larvae was intended after one hour of the revelation period with

an interval of 1 h up to 10 h. The motionless and unreactive larvae were considered dead. The departed larvae were noted and counted, and the regular % of transience was considered using Abbott's procedure.

$$\% \text{ Mortality} = \frac{(X - Y)}{X} * 100$$

where X is the survival % in the larvae population control, and Y is the % of survival in the larvae population treated.

2.8 Statistical analysis

The mortality of the larvae at various concentrations was analyzed by the probit analysis method described by Raymond in 1985. The attentiveness that caused 90% and 50% death (LC_{50}) have resulted in assurance intervals for mathematically important comparisons. The significance of the differences between the treatments versus concentrations was established by Oneway ANOVA and p values < 0.05, were measured as noteworthy. The data were analyzed statistically and completed using IBM SPSS (Statistics version 21), and the graphs were plotted using ggplot 2 version 3.3.6. in R version 4.1.1 [24].

3 Results and discussion

3.1 Chemical composition of essential oils

The present study confirmed that species of *Cinnamomum* were rich repositories of terpene compounds and oxygenated derivatives. A comparison between the chemical compositions of oils with the previous reports has indicated the compositional differences of essential oils according to the circulation and components accumulation in the same plant parts. The EOs isolated from *Cinnamomum* leaves were characterized by a diverse group of sesquiterpenes with varying areas of distribution. Few previous studies attempted on essential oils have shown variation in the chemical constituents from the same plant part, this variation was due to the influence of ecological factors such as climate and soil [31–33] genetic and developmental factors [33, 34] and also depends on the geographical region where the plant grow [35–37].

The yield of EOs from *Cinnamomum* species mottled from 0.142 to 1.96% v/w. The greatest surrender was obtained from *C. verum* (1.96% v/w) followed by *C. malabatrum* (1.71%), *C. sulphuratum* (1.17% v/w), *C. wightii* (1.014% v/w), *C. camphora* (0.32% v/w), and *C. glanduliferum* (0.171% v/w) whereas it was minimum yield from *C. perrottetii* (0.142% v/w). The higher quantity was obtained from *C. verum* (1.96% v/w) and *C.sulphuratum* (1.18% v/w) making these species a new natural potential source of volatile aromatic compounds. The GC-MS results demonstrated that sesquiterpenes were the principal class of compound in *C. sulphuratum*, *C. perrottetii*, *C. wightii*, *C. camphora*, and *C. glanduliferum*. In contrast to that, monoterpenes were predominantly reported in *C. verum* and esters in *C. malabatrum*. The predominant volatile components detected from the species of *Cinnamomum* were eugenol (38.04%), shyobunol (13.31%), isocaryophyllene (10.36%) along with germacrene D (7.89%), β-caryophyllene (11.33%), α-cadinol (10.7%), and Isoeugenol acetate (27.95%) in *C. verum*, *C. perrottetii*, *C. camphora*, *C. sulphuratum*, *C. wightii*, *C. glanduliferum*, and *C. malabatrum*, respectively (Table 1).

A total of 42 phytoconstituents were detected from *C. sulphuratum* including 31 sesquiterpenes, 7 sesquiterpenoids, and 4 monoterpenes. Germacrene D was established as the chief compound eluted at a retention time of 17.95 with a 7.89 % distribution. Furthermore, β-selinene (5.66%) and α-selinene (5.58%) were detected at the elution time of 18.128 and 18.266, respectively. Among the identified phytocompounds, the minor bioactive compounds of the essential oil were allo-ocimene (0.34%) and α-cubebene (0.32%) which were detected at the elution times 12.33 and 23.12 (Table 2 and Fig. 2a). The *C. sulphuratum* EO chemical composition from Kerala were rich with benzyl benzoate and another accession from Kerala has reported with (E)-cinnamyl acetate, linalool and benzyl benzoate as main constituents. *C. sulphuratum* from Kodagu district and Gudalur, Nilgiri region of Tamil Nadu showed dissimilarities of chemical constituents, in which α-phellandrene, (E)-caryophyllene, linalool, and benzyl benzoate were the major compounds from

Table 1 Characteristics of isolated essential oil

Sl. No.	Species	Yield (v/w)	Appearance	Color	Odor
1.	***C. verum* J. Presl.**	1.95%	Thick consistency	Pale yellow	Spicy and warm aroma
2.	***C. perrottetii* Meisn.**	0.142%	Thick consistency	Golden yellow	Slightly pungent aroma
3.	***C. sulphuratum* Nees.**	1.18%	Thick consistency	Greenish yellow	Mild aromatic
4.	***C. camphora* (L.) J. Presl.**	0.32%	Thick consistency	Brownish-yellow	Pungent
5.	***C. wightii* Meisn.**	1.014%	Thick consistency	Dark yellow	Piercing
6.	***C. glanduliferum* (Wall.) Meisn.**	0.171%	Clear, loose	Light green	Pungent
7.	***C. malabatrum* (Burm.f.) Presl.**	1.71%	Thick consistency	Dark yellow	Spicy aromatic smell

Table 2 List of phytocompounds detected from the essential oil of seven *Cinnamomum* spp. by GC-MS

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
1.	*C. sulphuratum* Nees.	1	1483	1490	[38]	7.74	0.71	β -Guaiene	Sesquiterpene	Sesquiterpenes : 31 Sesquiterpenoid : 7 Monoterpene : 4 Total : 42
		2	931	932	[39]	7.79	2.32	α -Pinene	Monoterpene	
		3	1118	1118	[40]	9.943	0.34	Allo-Ocimene	Monoterpene	
		4	1082	1082	[41]	10.144	0.76	Linalool	Monoterpene	
		5	1458	1458	[42]	11.305	0.73	Alloaromadendrene	Sesquiterpene	
		10	1398	1398	[42]	14.746	1.08	δ - Elemene	Sesquiterpene	
		11	1344	1345	[43]	15.006	0.32	α -Cubebene	Sesquiterpenoid	
		12	1353	1353	[44]	15.657	0.8	α -Copaene	Sesquiterpene	
		15	1398	1389	[42]	15.963	5.44	(-)-β -Elemene	Sesquiterpene	
		16	1413	1413	[45]	16.654	5.51	(-)- Isocaryophyllene	Sesquiterpene	
		17	1431	1430	[42]	16.798	2.25	G-Elemene	Sesquiterpene	
		18	1490	1471	[39]	16.931	0.52	α -Guaiene	Sesquiterpene	
		20	1440	1460	[13]	17.235	1.78	(*E*)- β -Farnesene.	Sesquiterpene	
		21	1452	1452	[42]	17.393	1.86	α -humulene	Sesquiterpene	
		22	1488	1488	[46]	17.789	2.49	γ.-Muurolene	Sesquiterpene	
		23	1479	1484	[42]	17.957	7.89	Germacrene D	Sesquiterpene	
		24	1729	1723	[38]	18.04	1.1	Isoledene	Sesquiterpene	
		25	1488	1489	[42]	18.128	5.66	β - Selinene	Sesquiterpene	
		26	1532	1499	[47]	18.17	1.48	Curzerene	Sesquiterpene	
		27	1474	1498	[42]	18.266	5.58	α -Selinene	Sesquiterpene	
		28	1409	1510	[13]	18.319	3.01	α-Farnesene	Sesquiterpene	
		29	1500	1506	[41]	18.426	1.03	β -Bisabolene	Sesquiterpene	
		30	1435	1513	[42]	18.57	0.94	γ - Cadinene	Sesquiterpene	
		31	1521	1522	[42]	18.664	2.47	D-Cadinene	Sesquiterpene	
		32	1527	1528	[48]	18.727	0.36	Cis-Calamenene	Sesquiterpene	
		33	1430	1433	[49]	18.904	0.4	(+)-β -Guajen	Sesquiterpene	
		34	1425	1401	[39]	19.036	0.67	Cyclosativene	Sesquiterpene	
		37	1465	1470	[50]	19.425	0.68	(-)-Selina-4,11-Diene	Sesquiterpene	

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		38	1540	1559	[42]	19.529	4.92	Germacrene B	Sesquiterpene	
		39	1575	1585	[41]	19.601	2.75	Spathulenol.	Sesquiterpenoid	
		40	1536	1583	[41]	19.91	2.28	Caryophyllene Oxide	Sesquiterpene	
		41	1650	1652	[51]	20.017	1.25	Cadinol	Sesquiterpenoid	
		43	1502	1502	[52]	20.205	0.92	Eremophilene	Sesquiterpene	
		45	1600	1608	[42]	20.460	0.4	(-)-Humulene Epoxide	Sesquiterpene	
		47	1561	1567	[53]	20.617	0.64	Germacrene D-4-Ol	Sesquiterpene	
		49	1632	1635	[54]	20.79	2.97	Isospathulenol	Sesquiterpenoid	
		50	1610	1602	[55]	20.879	0.7	Mintketone	Monoterpene	
		51	1628	1633	[50]	20.974	1.01	Tau-Muurolol	Sesquiterpenoid	
		52	1458	1458	[42]	21.153	0.66	Alloaromadendrene Oxide-(1)	Sesquiterpene	
		53	1500	1501	[39]	21.208	0.53	γ -Himachalene	Sesquiterpene	
		54	1652	1652	[51]	21.274	3.11	α-Cadinol	Sesquiterpenoid	
		56	1691	1693	[48]	21.476	0.35	Germacrone	Sesquiterpenoid	
2	*C. perrottetii* Meisn.	1	948	935	[41]	5.487	0.27	(+)- α -Pinene	Monoterpene	Sesquiterpene :17 Sesquiterpenoid :8 Monoterpene : 7 Monoterpenoid :2 Diterpene :1 Total : 35
		2	980	985	[56]	6.336	0.16	2-Menthene	Monoterpenoid	
		4	1018	1029	[57]	7.604	0.93	D-Limonene	Monoterpene	
		5	1650	1650	[57]	7.667	1.59	α-Elemene	Sesquiterpenoid	
		7	1054	1054	[42]	8.261	0.15	γ-Terpinene	Monoterpene	
		9	1082	1082	[57]	10.256	1.57	Linalool	Monoterpene	
		10	1169	1174	[51]	11.807	0.19	Terpinen-4-Ol	Monoterpene	
		11	1488	1488	[46]	15.647	0.36	γ -Muurolene	Sesquiterpene	
		12	1386	1389	[42]	15.947	1.45	(-)-β-Elemene	Sesquiterpenoid	
		13	1452	1452	[42]	16.7	10.91	α - humulene	Sesquiterpene	
		16	1118	1118	[58]	17.505	2.21	alloocimene	Monoterpene	
		18	1484	1484	[42]	17.983	8.9	Germacrene D	Sesquiterpene	
		20	1727	1727	[59]	18256	2.52	α - Muurolene	Sesquiterpene	
		21	1488	1488	[46]	18.568	0.97	γ.-Muurolene.	Sesquiterpene	
		22	1437	1537	[54]	18.684	4.4	Cadina-1,4-Diene	Sesquiterpene	
		23	1685	1688	[42]	18.752	2.68	Shyobunol	Sesquiterpenoid	
		25	1584	1588	[60]	18.946	0.36	Cubebanol	Sesquiterpene	

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		26	1476	1479	[61]	19.035	0.27	Zizanene	Monoterpene	
		27	1522	1545	[62]	19.28	018	Elemol	Sesquiterpenoid	
		29	1413	1413	[45]	19.496	0.34	Isocaryophyllene	Sesquiterpenoid	
		30	1567	1567	[63]	19.912	4.43	Germacrene D-4-Ol	Sesquiterpene	
		31	1583	1583	[57]	20.03	1.64	Caryophyllene Oxide	Sesquiterpene	
		32	1570	1570	[64]	20.085	0.34	(-)-Globulol	Sesquiterpene	
		33	1590	1590	[65]	20.308	5.96	Viridiflorol	Sesquiterpenoid	
		34	1530	1565	[41]	20.459	0.54	Ledol	Sesquiterpene	
		35	1608	1608	[42]	20.548	0.19	Humulene Epoxide	Sesquiterpene	
		37	1610	1613	[66]	20.861	1.4	Di-Epi-1,10, Cubenol	Sesquiterpene	
		38	1642	1642	[54]	20.984	0.23	Cubenol	Sesquiterpene	
		40	1652	1652	[51]	21.49	10.86	α-Cadinol	Sesquiterpene	
		41	1892	1895	[67]	21.585	0.21	1-Naphthalenol	Sesquiterpenoid	
		42	1688	1688	[42]	22.279	13.31	Shyobunol	Sesquiterpene	
		43	1739	1740	[68]	22.969	0.17	Mintsulfide	Monoterpenoid	
		48	1813	-	-	25.662	0.3	Voleneol Diacetate	Sesquiterpene	
		49	3942	-	-	25.805	0.23	1heptatriacotanol	Sesquiterpenoid	
		50	2066	2066	[69]	28.963	0.45	Phytol	Diterpene	
3.	*C. verum* J. Presl	2	1124	1002	[42]	4.75	0.04	α -Phellandrene Epoxide	Monoterpene	Monoterpene : 9 Sesquiterpene : 7 Ester : 5 Phenylpropanoid: 4 Sesquiterpenoid : 3 Monoterpenoid : 1 Total = 29
		3	929	924	[42]	5.326	0.02	α-Thujene	Monoterpene	
		4	1078	1086	[42]	5.489	0.08	Terpinolene	Monoterpene	
		6	1011	1003	[57]	7.065	0.14	1-Phellandrene	Monoterpene	
		7	932	1654	[70]	7.350	0.87	Isoelemicin	Monoterpene	
		8	950	-	-	7.420	1.9	α -Chlorohydrin	Sesquiterpene	
		11	1059	1031	[56]	7.718	0.1	1,8-cineol	Monoterpenoid	
		13	1056	1050	[50]	7.99	0.05	Trans- β -Ocimene	Monoterpene	
		14	1053	1054	[42]	8.261	0.09	γ-Terpinene	Monoterpene	
		15	931	932	[42]	8.901	0.08	(+)-α-Pinene	Monoterpene	
		19	1082	1082	[71]	10.202	3.81	Linalool	Monoterpene	
		20	1037	1033	[71]	11.344	8.13	Benzyl alcohol	Ester	
		26	1189	1179	[72]	13.603	4.81	Cinnamaldehyde	Phenylpropanoid	
		27	1319	1312	[73]	14.441	0.44	Cinnamic Alcohol	Ester	
		28	1392	1356	[42]	15.593	38.04	Eugenol	Phenylpropanoid	
		30	1361	1460	[42]	16.211	0.28	Methyleugenol	Phenylpropenoid	

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		31	1413	1413	[38]	16.677	2.78	Isocaryophyllene	Sesquiterpenoid	
		32	1451	1443	[42]	17.42	22.36	(E)-Cinnamyl Acetate	Ester	
		33	1474	1496	[48]	17.78	0.15	Valencene	Sesquiterpene	
		34	1511	1511	[53]	17.955	0.13	Germacrene D	Sesquiterpene	
		35	1509	1500	[42]	18.254	1	Bicyclogermacrene	Sesquiterpene	
		36	1552	1485	[74]	18.571	0.95	Eugenyl Acetate	Ester	
		37	1550	1563	[57]	19.178	0.1	Nerolidol	Sesquiterpene	
		39	1573	1573	[73]	20.03	0.56	Caryophyllene]Oxide	Sesquiterpene	
		40	1570	1570	[64]	20.255	0.09	Globulol	Sesquiterpenoid	
		42	1580	1573	[73]	21.151	0.1	Tau.-Muurolol	Sesquiterpenoid	
		43	1652	1652	[51]	21.38	0.12	α-Cadinol	Sesquiterpene	
		44	1760	1762	[69]	23.432	4.64	Benzyl Benzoate	Ester	
		45	2142	2142	[75]	36.774	0.11	Dihydroeugenol	Phenylpropanoid	
4	*C. wightii* Meisn.	1	1419	1411	[54]	5.48	0.18	Dodecyl Aldehyde	Aldehyde	Sesquiterpenoid : 6 Aldehyde : 2 Monoterpene : 5 Monoterpenoid : 2 Sesquiterpene : 25 Ester : 2 Fatty acids : 2 Diterpenes :1 Total :45
		2	943	932	[42]	6.445	0.16	Pinene	Monoterpene	
		4	1018	1024	[51]	7.606	0.58	δ -Limonene	Monoterpene	
		6	1046	1050	[50]	7.991	1.88	Trans- β -Ocimene	Monoterpene	
		7	1082	1097	[57]	10.259	2.77	Linalool	Monoterpenoid	
		8	1138	1165	[51]	11.671	0.3	Borneol	Monoterpenoid	
		9	1180	1188	[62]	12.072	0.47	Terpineol	Monoterpene	
		10	1337	1338	[57]	14.718	0.88	δ -Elemene	Sesquiterpene	
		12	1353	1353	[44]	15.64	0.63	α -Copaene,	Sesquiterpene	
		13	1398	1389	[42]	15.955	1.72	(-)-β -Elemene	Sesquiterpene	
		14	1402	1411	[54]	16.356	0.2	Dodecanal	Aldehyde	
		15	1456	1456	[39]	16.739	11.33	β -Caryophyllene	Sesquiterpene	
		16	1575	1575	[57]	16.803	0.55	Espatulenol	Sesquiterpene	
		17	1582	1575	[76]	16.845	0.33	B-Copaen-4-α-ol	Sesquiterpene	
		18	1518	1514	[77]	17.04	0.55	Elixene	Sesquiterpene	
		19	1582	1578	[57]	17.241	0.27	Spatulenol	Sesquiterpene	
		21	1118	1118	[58]	17.537	4.11	Alloocimene	Monoterpene	
		23	1492	1484	[42]	17.954	3.64	Germacrene D	Sesquiterpene	
		24	1419	1497	[78]	18.14	1.95	Virdiflorene	Sesquiterpenoid	
		25	1543	1537	[42]	18.268	3.37	α -Cadinene	Sesquiterpene	

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		26	1435	1513	[42]	18.592	3.04	γ -Cadinene	Sesquiterpene	
		27	1808	1808	[42]	18.712	5.14	Delta-Cadinene	Sesquiterpene	
		28	1537	1528	[57]	18.762	2.17	Cis-Calamenene	Sesquiterpenoid	
		29	1440	1482	[57]	18.95	0.39	Naphthalene	Sesquiterpene	
		30	1440	1483	[42]	19.042	1.47	α -Amorphene	Sesquiterpene	
		34	1530	1568	[62]	19.759	0.38	Palustrol	Sesquiterpenoid	
		36	1583	1583	[73]	20.03	3.21	Caryophyllene Oxide	Sesquiterpene	
		38	1570	1570	[64]	20.303	4.46	Globulol	Sesquiterpenoid	
		39	1413	1413	[45]	20.399	0.36	Isocaryophyllene	Sesquiterpene	
		40	1618	1613	[38]	20.462	0.94	Epicubenol	Sesquiterpenoid	
		41	1452	1452	[42]	20.55	0.52	Humulene	Sesquiterpene	
		42	1393	1386	[79]	20.63	0.94	Aromadendrane	Sesquiterpene	
		43	1782	1780	[80]	20.799	1.16	Cryptomeridiol	Sesquiterpene	
		44	1642	1642	[54]	20.87	1.54	Cubenol	Sesquiterpene	
		46	1484	1478	[73]	21.185	4.97	γ-Muurolene	Sesquiterpene	
		47	1452	1452	[42]	21.237	3.63	α - humulene	Sesquiterpene	
		49	1652	1652	[51]	21.493	6.51	α-Cadinol	Sesquiterpenoid	
		52	1689	1688	[38]	21.969	0.42	Bisabolol	Sesquiterpene	
		53	1688	1688	[42]	22.192	2.49	Shyobunol	Sesquiterpene	
		54	1722	1722	[73]	22.432	1.64	(Z,E)-Farnesol	Sesquiterpene	
		55	1760	1762	[69]	23.394	0.76	Benzyl Benzoate	Ester	
		57	2066	2066	[69]	28.954	0.26	Phytol	Diterpene	
		58	2078	2078	[81]	29.348	0.29	Linoleic-Acid	Fatty acid	
		59	2141	2141	[82]	29.441	0.28	Elaidic Acid	Fatty acid	
		60	3792	3790	[83]	38.562	2.5	γ -Sitostenone	Ester	
5	*C. camphora* (L.) J. Presl.	2	1489	1483	[42]	7.591	2.51	α -Amorphene	Sesquiterpene	Sesquiterpene : 24 Sesquiterpenoid : 5 Monoterpene : 4 Monoterpenoid : 3 Aldehyde :2 Diterpene : 2 Fatty acids : 1 Total = 41
		3	964	-	-	7.663	2.72	β -Cymene	Monoterpene	
		4	1082	1082	[57]	9.934	0.78	Linalool	Monoterpenoid	
		5	1172	1180	[46]	11.293	0.32	Borneol	Monoterpenoid	
		6	1197	1197	[46]	11.427	1.38	Terpineol	Monoterpene	
		9	996	996	[84]	12.726	0.14	Benzaldehyde	Aldehyde	
		10	1251	1244	[62]	12.985	0.16	Piperitone	Monoterpene	
		11	1337	1337	[85]	14.702	0.25	δ -Elemene	Sesquiterpene	

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		13	1353	1353	[44]	15.634	1.41	Copaene	Sesquiterpene	
		14	1212	1213	[72]	15.918	2.97	Limonene	Monoterpenoid	
		15	1310	1310	[86]	16.152	0.55	Undecenal	Aldehyde	
		17	1413	1413	[45]	16.653	10.36	Isocaryophyllene	Sesquiterpene	
		18	1430	1436	[78]	16.834	1.19	α -Bergamotene	Sesquiterpene	
		19	1386	1585	[41]	17.01	0.53	Spatulenol	Sesquiterpene	
		20	1392	1386	[79]	17.212	0.54	Aromadendrene	Sesquiterpenoid	
		21	1569	1489	[42]	17.37	2.03	β -Selinene	Sesquiterpene	
		22	1590	1590	[65]	17.469	2.41	Viridiflorol	Sesquiterpenoid	
		23	1488	1488	[46]	17.741	1.81	γ -Muurolene	Sesquiterpene	
		24	1483	1483	[42]	17.831	1.31	α -Amorphene	Sesquiterpene	
		25	1512	1512	[87]	18.076	1.97	Γ-Cadinene	Sesquiterpene	
		26	1118	1118	[58]	18.222	2.43	Allo Ocimene	Monoterpene	
		28	1398	1389	[42]	18.637	3.25	β -Elemene	Sesquiterpenoid	
		30	1543	1537	[42]	19.009	0.78	α -Cadinene	Sesquiterpene	
		32	1461	1456	[39]	19.361	0.91	β -Caryophellene	Sesquiterpene	
		33	1562	1561	[51]	19.479	1.32	Nerolidol	Sesquiterpene	
		35	1541	1541	[40]	19.751	0.87	Maaliol	Sesquiterpene	
		36	1536	1522	[42]	19.915	3.68	δ -Cadinene	Sesquiterpene	
		37	1583	1583	[57]	20.036	8.74	β -Caryophylene Oxide	Sesquiterpene	
		38	1452	1452	[42]	20.248	2.39	Humulene	Sesquiterpene	
		39	1600	1600	[88]	20.456	1.74	Rosifoliol	Sesquiterpenoid	
		40	1452	1452	[42]	20.538	1.03	Humulene	Sesquiterpene	
		43	1642	1642	[54]	20.852	1.08	Cubenol	Sesquiterpene	
		44	1535	1635	[54]	20.969	0.99	Isospathulenol	Sesquiterpene	
		45	1580	1585	[41]	21.185	3.73	Spatulenol	Sesquiterpene	
		46	1652	1652	[42]	21.439	8.71	α -cadinol	Sesquiterpenoid	
		47	1873	1895	[89]	21.563	0.64	1-Naphthalenol	Sesquiterpene	
		49	1689	1683	[38]	21.94	0.72	Bisabolol	Sesquiterpene	
		51	1722	1722	[73]	22.406	0.67	Farnesol	Sesquiterpene	
		54	1754	1846	[90]	24.62	0.19	Hexahydrofarnesyl acetone	Diterpene	
		57	1968	1969	[62]	26.658	0.29	Palmitic Acid	fatty acid	
		58	2066	2066	[69]	28.962	0.54	Phytol	Diterpene	
6.	*C. glanduliferum* (Wall.) Meisn.	1	1004	1004	[42]	7.315	0.36	Carene	Monoterpene	Sesquiterpene : 17 Sesquiterpenoid :7 Monoterpene : 4 Monoterpenoid : 2 Diterpene : 1 Fatty acids : 1 Total : 32

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		3	1036	1036	[47]	7.992	0.49	β -Cis-Ocimene	Monoterpene	
		5	1014	1014	[42]	8.261	0.56	Terpinene	Monoterpene	
		8	1197	1197	[46]	11.903	0.98	Terpineol	Monoterpene	
		10	1175	1198	[91]	12.425	0.27	Piperitol	Monoterpenoid	
		14	1300	1300	[92]	14.681	1.46	Ascaridole	Monoterpenoid	
		15	1353	1353	[44]	15.951	3.51	Copaene	Sesquiterpene	
		16	1398	1389	[42]	15.951	6.31	β -Elemene	Sesquiterpenoid	
		17	1456	1456	[39]	16.652	9.35	β -Caryophyllene	Sesquiterpene	
		18	1430	1436	[78]	16.853	1.39	α -Bergamotene	Sesquiterpene	
		19	1452	1452	[42]	17.484	1.77	Humulene	Sesquiterpene	
		20	1386	1578	[57]	17.484	2.03	Espatulenol	Sesquiterpene	
		22	1524	1510	[93]	17.855	0.54	α -Curcumene	Sesquiterpene	
		23	1493	1484	[42]	17.923	1.5	Germacrene-D	Sesquiterpene	
		24	1469	1489	[42]	18.104	2.69	β -Selinene	Sesquiterpene	
		25	1449	1442	[94]	18.241	4.62	α - Guajen	Sesquiterpene	
		27	1531	1522	[42]	18.662	5.9	δ -Cadinene	Sesquiterpene	
		28	1537	1528	[48]	18.725	1.13	Cis-Calamenene	Sesquiterpenoid	
		29	1514	1514	[95]	19.02	0.58	Cadina-3-9-diene	Sesquiterpene	
		31	1564	1563	[57]	19.477	1.53	Nerolidol	Sesquiterpene	
		33	1586	1386	[79]	19.903	3.16	Aromadendrene	Sesquiterpenoid	
		35	1570	1570	[64]	20.084	2.61	Globulol	Sesquiterpenoid	
		36	1590	1590	[65]	20.252	2.29	Viridiflorol	Sesquiterpenoid	
		37	1652	1652	[42]	20.458	1.07	α -Eudesmol	Sesquiterpenoid	
		38	1452	1452	[42]	20.538	0.3	Humulene	Sesquiterpene	
		39	1642	1642	[54]	20.617	0.5	Cubenol	Sesquiterpene	
		43	1607	1607	[38]	20.963	0.85	Isocaryophellene	Sesquiterpene	
		44	1727	1727	[59]	21.143	5.11	α-Muurolene	Sesquiterpene	
		46	1652	1652	[51]	21.44	10.7	α -Cadinol	Sesquiterpenoid	
		48	1722	1722	[73]	22.405	0.47	Farnesol	Sesquiterpene	
		49	2066	2066	[69]	28.962	0.51	Phytol	Diterpene	
		50	2141	2141	[82]	29.471	0.31	Elaidic Acid	Fatty Acid	
7.	*C. malabatrum* (Burm.f.) Presl.	2	933	-	-	5.205	0.03	Methyl sulfonylmethane	Sulfone	Ester : 8 Sesquiterpene : 2 Phenyl propanoid: 2 Monoterpene: 1 Sulfone: 1 Total : 14
		3	1001	1002	[64]	5.519	0.03	Alpha Fellandrene	sesquiterpene	
		5	1430	-	-	7.05	0.05	Mesitylenic acid methyl ester	Ester	

Table 2 (continued)

Sl. no.	Name of the species	Peak	Retention Index (calculated)	Retention Index (References)		Retention time	Area %	Compound	Compound nature	Total no. of compounds and types
		6	1500	1530	-	7.46	0.03	Geranyl crotonoate	Ester	
		7	1082	1082	[96]	9.221	0.56	Linalool	Monoterpene	
		8	1216	1214	[97]	10.68	1.02	Benzyl acetate	Ester	
		10	1200	1179	[98]	13.304	4.28	Cinnamaldehyde	Phenyl propanoid	
		11	1624	1623	[99]	15.14	27.95	Isoeugenol acetate	Phenyl propanoid	
		12	1620	1620	[99]	15.42	0.26	Hydrocinnamyl acetate	Ester	
		13	1509	1510	[13]	16.63	0.34	Alpha Farnesene	Sesquiterpene	
		14	1400	1430	[42]	17.075	4.5	(E)-Cinnamyl acetate	Ester	
		15	1760	1762	[69]	23.38	0.57	Benzyl Benzoate	Ester	
		16	1918	1917	[100]	34.85	0.91	Phthalic acid, di(2-propylpentyl)ester	Ester	
		17	2519	2519	[101]	37.47	0.93	Diisooctyl phthalate	Ester	

Kodaku [102] and (E)-cinnamyl acetate, linalool, α-cadinol, benzyl benzoate, α-cadinene, and α-muurolol from Gudulur. The Northeast India leaf has also shown disparity with a significant presence of geranial, neral, and geraniol [103].

Out of 35 compounds reported from the E of *C. perrottetii*, 17 sesquiterpenes contributed to the main division of the EOs, along with 8 sesquiterpenoids, 7 monoterpenes, 2 monoterpenoids, and 1 diterpene. Shyobunol (13.31%) was detected as the dominant compound recorded at the elution time of 22.27 followed by caryophyllene (10.91%), α-Cadinol (10.86%), germacrene-D (8.9%), viridiflorol (5.96%), and germacrene D-4-Ol (4.43%) at the retention time of 16.7, 21.49, 17.98, 20.30, and 19.91, respectively (Table 2 and Fig. 2b). Schmidt et al. [104] and Boniface et al. [105] reported eugenol (74.9%) as the basic constituent of the *C. perrottetii* oil from Srilanka. Although, (E)-cinnamaldehyde (37.6%), cinnamyl benzoate (16.4%), and cinnamyl acetate (23.7%), as major constituents of the fresh leaves of *C. perrottetii*. The studies carried out by Unlu et al. [106] found that the major compounds *C. perrottetii* oil were (E) (E)-cinnamyl acetate (7.44%), cinnamaldehyde (68.95%), and benzaldehyde (9.94%). When compared to data observed from the current study with prior reports, the major components of *C. perrottetii*, have indicated that there was no similarity recorded in the principle component, shyobunol (13.31%) from the EOs.

Volatile phytocompounds 29 were identified from the EO of *C. verum* including 9 monoterpenes, 7 sesquiterpenes, 5 esters, 4 phenylpropanoids, 3 sesquiterpenoids, and 1 monoterpenoid. Eugenol (38.04%), one of the important phenylpropanoids was prominently detected at the elution time of 15.593. Cinnamyl acetate (22.36%), benzyl alcohol (8.13%), and cis-cinnamaldehyde (4.81%) were the other compounds subsequently eluted at 17.42, 11.344, and 13.603 retention times (Table 2 and Fig. 2c).

The EO chemical combination of *C. verum* collected from various geographical regions confirmed the dissimilarities of phytocompounds, in which (E)-linalool (2.30%), eugenol (86.02%), and caryophyllene (5.70%) recorded from the leaf samples of Fiji islands [107]. By the present study, *C.verum* essential oil from Palni hills, Tamil Nadu has also detected eugenol as a major compound (3.9%) [108]. *C. verum* essential oil from Brazil recorded linalool (5.4%), E-cinnamaldehyde (4.0%), β-phellandrene (3.4%), eugenol (3.4%), α-pinene (3.9%), and benzaldehyde (2.7%) as major constituents [109] and the Vietnam samples reported linalool (22%), caryophyllene oxide (5.6%), methyl eugenol (0.7%), β-bisabolene (7.7%), eugenol (0.1%), bicyclogermacrene (11.2%), (E)-nerolidol (1.3%), and γ-cadinene (4.0%) as significant compounds [110]. The present study has reported eugenol and benzyl benzoate compared to the previous studies [23, 109, 111–113].

GC-MS chromatogram of essential oil of *C. wightii* has detected 45 compounds, of which, 25 were sesquiterpenes and the rest of them were categorized into 6 sesquiterpenoids, 5 monoterpenes, 2 monoterpenoids, together with 2

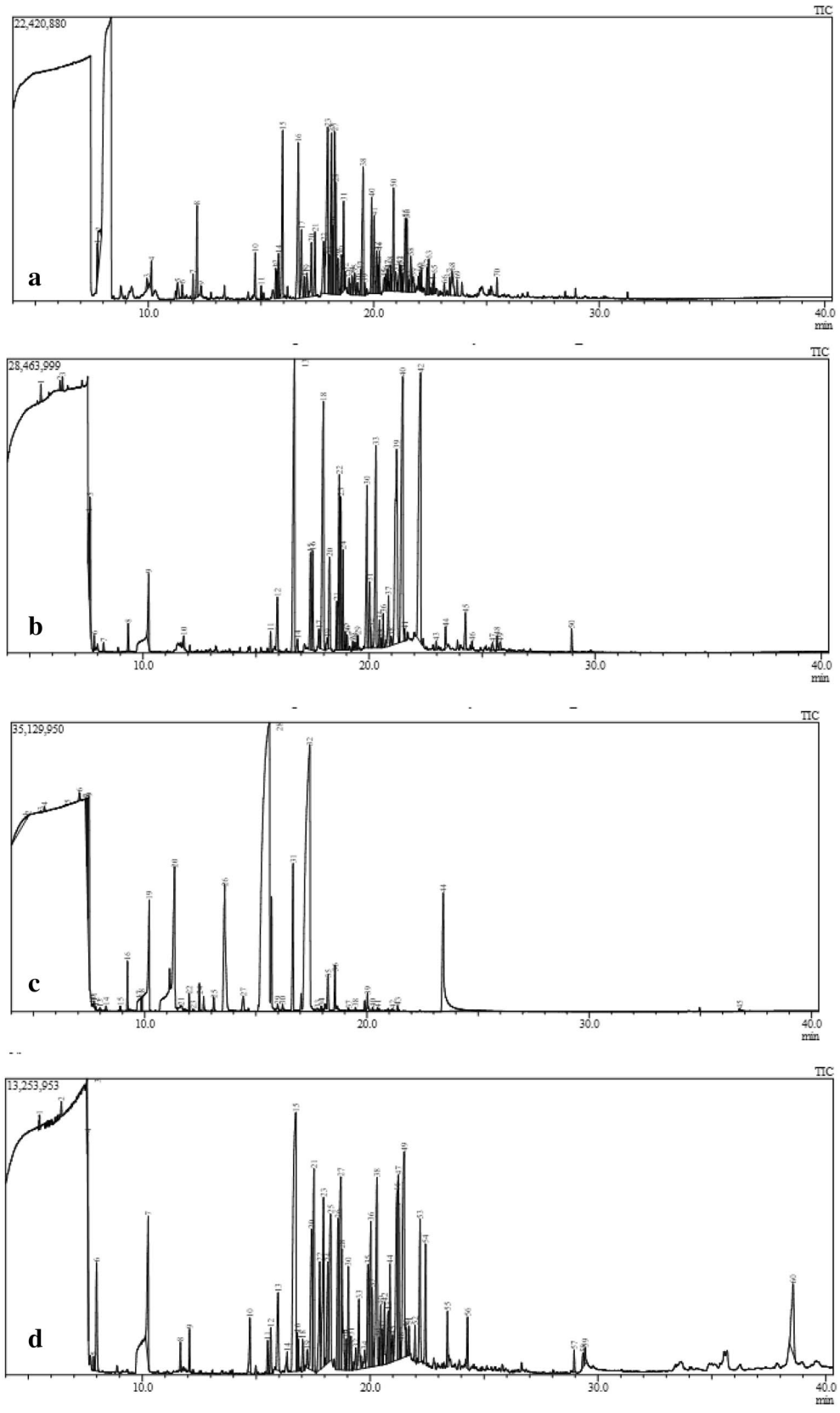

Fig. 2 **a** GC-MS Chromatogram of *C. sulphuratum* leaf essential oil. **b** GC-MS Chromatogram of *C. perrottetti* leaf essential oil. **c** GC-MS Chromatogram of *C. verum* leaf essential oil. **d** GC-MS Chromatogram of *C. wightii* leaf essential oil. **e** GC-MS Chromatogram of *C. camphora* leaf essential oil. **f** GC-MS Chromatogram of *C. glanduliferum* leaf essential oil. **g** GC-MS Chromatogram of *C. malabatrum* leaf essential oil

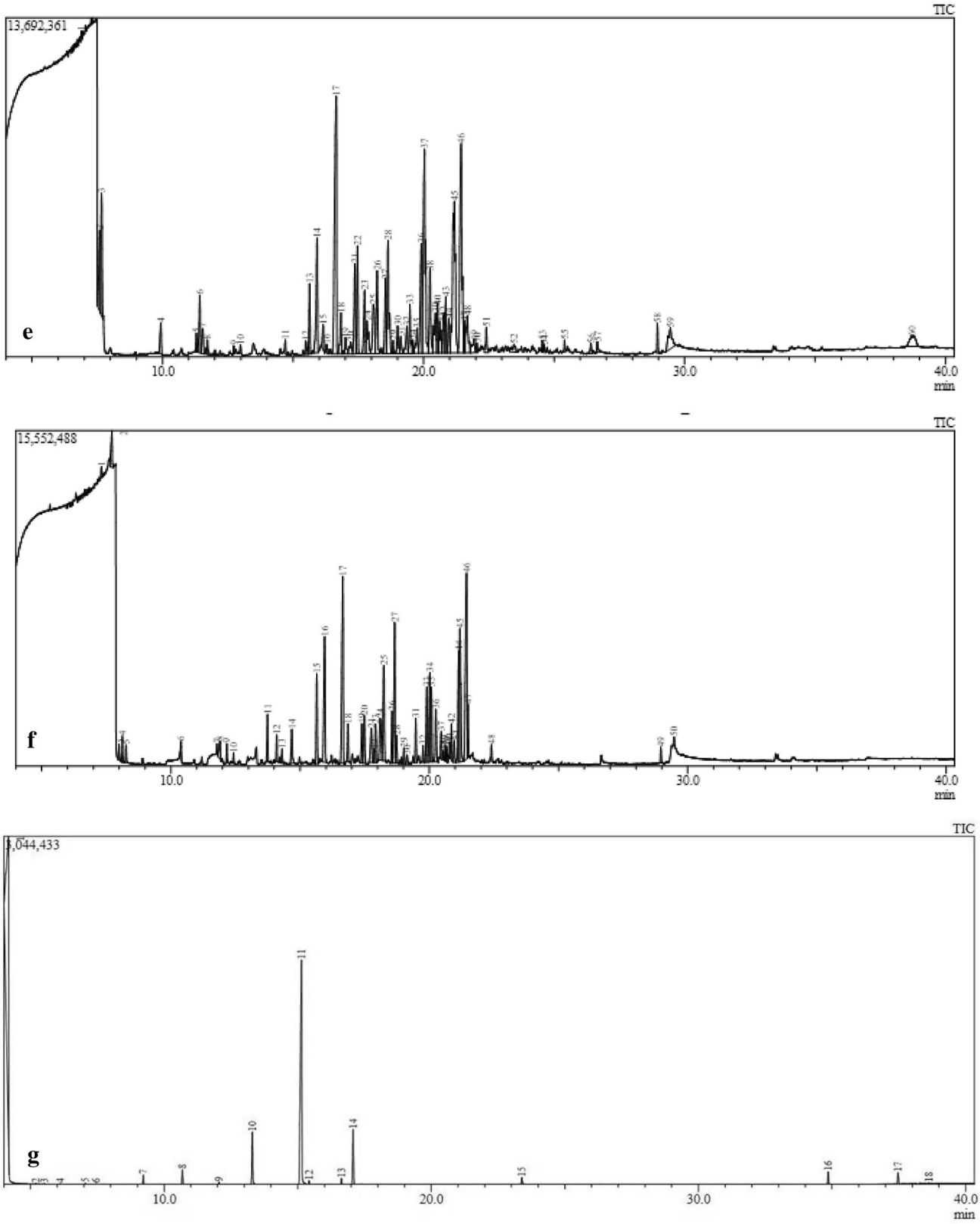

Fig. 2 (continued)

esters, 2 fatty acids, 2 aldehydes, and 1 diterpene(Table 2 and Fig. 2d). β-caryophyllene was reported with an area of 11.33% which has eluted at 16.73 along with α cadinol. *δ*-cadinene (5.14%), γ-muurolene (4.49%), globulol (4.46%), and allo-ocimene (4.11%) were further recognized based on the elution time. The oil leaf EO from seven different accessions of the Western Ghats showed a disparity in the chemical composition compared to the present study with the presence of monoterpenes such as β-phellandrene, α-pinene, linalool, limonene, p-cymene, methyl eugenol, (E)-caryophyllene, elemicin, α-phellandrene, and spathulenol [114].

Among 41 phytocompounds reported from the essential oil of *C. camphora*, 24 sesquiterpenes 5 sesquiterpenoids, 4 monoterpenes, 3 monoterpenoids, 2 diterpenes, 2 aldehydes, and 1 fatty acid were reported. Isocaryophyllene showed a major peak at a maintenance time of 16.65 with a relation profusion of 10.36%. The caryophyllene oxide (8.74%) and α-cardinal (8.71%) were reported as leading compounds eluted at 20.03 and 21.43 (Table 2 and Fig. 2e). By analyzing the result of GC-MS data of *C. camphora*, phytone (0.19%), piperitone (0.16%), and benzaldehyde (0.14%) were the meagre compounds eluted at 24.62, 12.98, and 12.72 retention times respectively. The *C. camphora* compound composition of oil has differed considerably from the previous reports by having isocaryophyllene (10.36%), β-caryophyllene oxide (8.74%), and α-cadinol (8.71) as the major constituents. However, all the phytocomponents reported by this study were similar to the previous study reports at different geographical regions [43, 115–119]. Comparatively, a different compound composition of *C. camphora* essential oil from China was reported with borneol as the major constituent, and another study reported a dissimilar composition with eucalyptol (16.8%), isoborneol (8.1%), camphor (5.0%), linalool (26.6%), and α-terpineol (8.7%), and β-phellandrene (5.1%) as major compounds [120].

Furthermore, 32 compounds were reported from *C. glanduliferum*, with 18 sesquiterpenes and 7 sesquiterpenoids, 2 monoterpenoids, 4 monoterpenes, 1 diterpene, and 1 fatty acid, α-cadinol has been observed from the large area of distribution with an area percentage of 10.7% followed by β-caryophyllene (9.35%), β-elemene (6.31%), *δ*-muurolene (5.11%), and α-cadinene (5.9%) at the retention time of 16.65, 15.95, 21.14, respectively (Table 2 and Fig. 2f). The *C. glanduliferum* EO composed in various geographical regions showed different composition in which, (E)-nerolidol (52.2 %) and caryophyllene oxide (6.0%) was recorded in samples from the Chessa area of Arunachal Pradesh [121], α-terpineol (9.40 %), α-pinene (20.28 %), 1, 8-cineole (41.42 %), and germacrene D-4-ol (6.10 %) from Uttarakhand [122], eucalyptol (59.44%), sabinene (14.99%), a-pinene (5.27%), a-terpineol (6.44%), β-elemene, and β-pinene (3.75%) from Al-Zohria Garden, Cairo, Egypt [123], and a new benzyl benzoate affluent chemotype was reported from Southern Western Ghats, India [36].

C. malabatrum leaf essential oil was reported with a total of fourteen compounds consisting of esters, sesquiterpenes, phenyl propanoids, monoterpene, and sulfone compounds. Isoeugenol acetate (27.95% at retention time 15.14), (E)-cinnamyl acetate (4.5% at retention time 17.075), cinnamaldehyde (4.28% at retention time 13.304), and benzyl acetate (1.02% retention time 10.68) were the major compounds (Table 2. and Fig. 2g). A previous study in *C. malabatrum* essential oil reported cinnamyl acetate and benzyl benzoate which were also detected in the present study [41].

By comparing the present attempt with the previous studies, the principle components of EO of species of *Cinnamomum* have varied due to geographical distribution and season of procurement. Nevertheless, the dissimilarity in the yield of EO and its chemical composition among the plants comes to analogous taxonomic groups anticipated to vary across agro-climatic and geographical conditions. It is as well as influenced by numerous external and internal environmental factors [2, 124, 125].

3.2 Antibacterial activity of EOs

The antibacterial potentialities of EOs from seven species of *Cinnamomum* were evaluated against 6 selected bacterial strains through the agar well method (diffusion). The results indicated that there has been a significant increase in the inhibition of bacterial growth with an increase in oil concentrations. The growth of microorganisms was inhibited and a maximum zone of inhibition (32 ± 0.88 mm) in Gentamicin. The leaf oils of *Cinnamomum* spp. were shown a synergistic effect with an inhibition zone alongside all experienced microorganisms (Fig. 3).

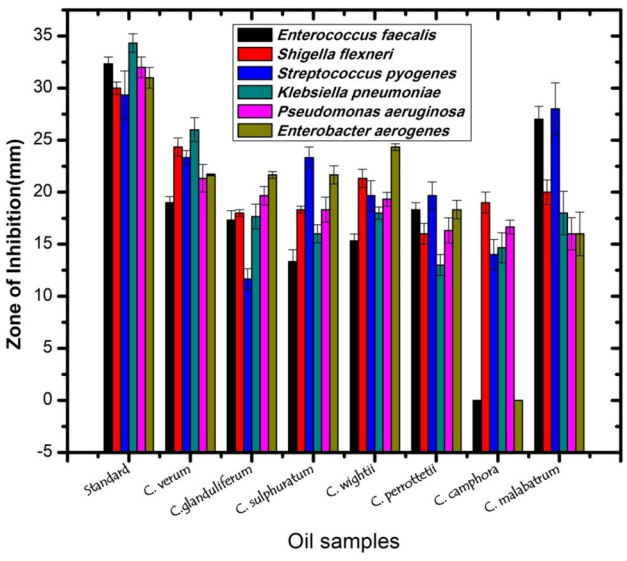

Fig. 3 Antibacterial activities of essential oils at higher concentrations (150µg/ml) of seven species of *Cinnamomum* sp.

Table 3 Antibacterial activities of essential oils at higher concentration (150µg/ml) of seven species of *Cinnamomum*

Essentials from	Control					
	Antibacterial activity with zone of inhibition(mm)					
	Enterococcus faecalis	*Shigella flexneri*	*Streptococcus pyogenes*	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*	*Enterobacter aerogenes*
Gentamicin	32.33 ± 0.666	30 ± 0.577	29.33 ± 2.333	34.33 ± 0.881	32 ± 1	31 ± 1
C.verum	19 ± 0.57	24.33 ± 0.88	23.33 ± 0.66	26 ± 1.15	21.33 ± 1.33	21.66 ± 1.0
C.glanduliferum	17.33 ± 0.88	18 ± 0.33	11.66 ± 1	17.66 ± 1.2	19.66 ± 0.88	21.66 ± 0.33
C.sulphuratum	13.33 ± 1.14	18.33 ± 0.33	23.33 ± 1	16 ± 0.88	18.33 ± 1.2	21.66 ± 0.88
C.wightii	15.33 ± 0.66	21.33 ± 0.88	19.66 ± 1.45	18 ± 0.577	19.33 ± 0.66	24.33 ± 0.33
*C. **perrottetii***	18.33 ± 0.66	16 ± 1	19.66 ± 1.33	13 ± 1	16.33 ± 1.2	18.33 ± 0.88
C.camphora	0.00	19 ± 1	14 ± 1.45	14.66 ± 1.45	16.66 ± 0.66	0.00
C. malabatrum	27 ± 1.15	20 ± 1.18	28 ± 2.5	18±2.08	16 ± 1.56	16 ± 2.1

C. verum has shown notable inhibition zone against *E. faecalis* (19 ± 0.57 mm), *S. flexneri* (24.33 ± 0.88 mm), *S. pyogenes* (23.33 ± 0.66 mm), *K. pneumoniae* (26 ± 1.15 mm), *P. aeruginosa* (21.33 ± 1.33 mm), and *E. aerogenes* (21.66 ± 1.0 mm) at higher concentration (150 µg/ml). Among the screened bacteria, *K. pneumoniae* exhibited higher antibacterial activity with the highest zone of inhibition (26 ± 1.15 mm) (Table 3). *S. pyogenes* showed higher susceptibility to the essential oils isolated from *C. sulphuratum* with a higher zone of inhibition (23.33 ± 1 mm). *E. aerogenes* exhibited a moderate inhibition zone (21.66 ± 0.88 mm) followed by *S. flexneri* (18.33 ± 0.33 mm) and *P. aeruginosa* (18.33 ± 1.2 mm) (Figs. 4 and 5).

Cinnamomum perrottetii exhibited very similar trends of antibacterial activities and *S. pyogenes* (19.66 ± 1.33 mm) showed the prevailing zone of inhibition. *E. faecalis* and *E. aerogenes* were recorded with an almost similar zone of inhibition (18 ± 0.57 mm and 18.33 ± 0.88 mm) followed by *P. aeruginosa* and *S. flexneri* (16.33 ± 1.2 mm and 16 ± 1 mm). Among the tested oil samples, *C. wightii* and *C. camphora* exhibited less susceptibility to selected pathogens. *C. wightii* displayed a moderate zone of inhibition for all screened bacteria and the highest resistance was recorded in *E. aerogenes* (24.33 ± 0.33 mm). The essential oil of *C. camphora* showed higher inhibitory activity against *S. flexneri* (19 ± 1 mm) and *P. aeruginosa* (16.66 ± 0.66 mm), *S. pyogenes* and *K. pneumoniae* with an equal value of inhibition at higher concentrations. Among the screened strains, *E. faecalis* and *E. aerogenes* exhibited 100% resistance against essential oil isolated from *C. camphora* (Figs. 6, 7, and 8).

C. malabatrum EO has shown a minimum inhibition zone of about 11 ± 0.58 mm, 11 ± 0.47 mm, and 20 ± 1.15 mm against *S. flexneri*, *K. pneumoniae*, and *S. pyogenes* at initial concentration (50 µg/mL). By increasing the oil concentration to 100 µg/mL, there was an increased antibacterial activity with an inhibition zone. Maximum zone of inhibition was noticed at higher concentration (150 µg/mL) against all microbial species such as *K. pneumoniae* (18 ± 2.08 mm), *S. flexneri* (20 ± 1.18 mm), *S. pyogenes* (28 ± 2.5 mm), *E. faecalis* (27 ± 1.24 mm) *P. aeruginosa*, (16 ± 1.56 mm), and *E. aerogenes* (16 ± 2.1 mm) (Figs. 9 and 10).

The data observed by the agar well dispersion assay of the essential oil suggested that the samples have synergistic antibacterial potentialities alongside gram +ve and −ve bacteria. The results suggested that *S. flexneri*, *K. pneumoniae*, and *E. aerogenes* were found to be more susceptible when compared with other selected strains [107]. *E. faecalis* and *E. aerogenes* showed higher resistance against all essential oils of *Cinnamomum* [108]. Nonetheless, all the oils exhibited antibacterial activity against screened bacteria and the inhibition zones were more eminent in *C. verum* followed by *C. malabatrum*, *C. sulphuratum*, *C. glanduliferum*, *C. perrottetii*, *C. wightii*, and *C. camphora*. Essential oils are a rich repository of volatile constituents such as phenol-derived aromatic and aliphatic compounds, terpenes, and terpenoids responsible for bactericidal activities [109]. Several studies have revealed that cinnamon has a strapping and reliable inhibitory effect against human pathogens [67, 126–132]. Previous studies have also confirmed that the phytocomponents, sesquiterpenes, cinnamaldehyde, and eugenol are accountable for the antibacterial activity of cinnamon oil [132–135]. Also, the bactericidal action mechanisms of cinnamaldehyde and eugenol of essential oils were recorded [136].

Gram -ve organisms are supposed to be less responsive to EOs than gram-positive bacteria [10, 14]. However, the present study has revealed equal inhibitory action of all oil samples with clear zones of inhibition that portray higher antibacterial efficacy. The essential oils are considered antimicrobials due to the hydrophobic character compounds are cause the destruction of cell structures, leading to the escape of the cell membrane and increasing the membrane permeability [132, 137] and thereby in a cascade type of action that affects other cellular structures. The essential oils damage lipids and proteins by getting coagulated in the cytoplasm of the bacterial cell [138, 139].

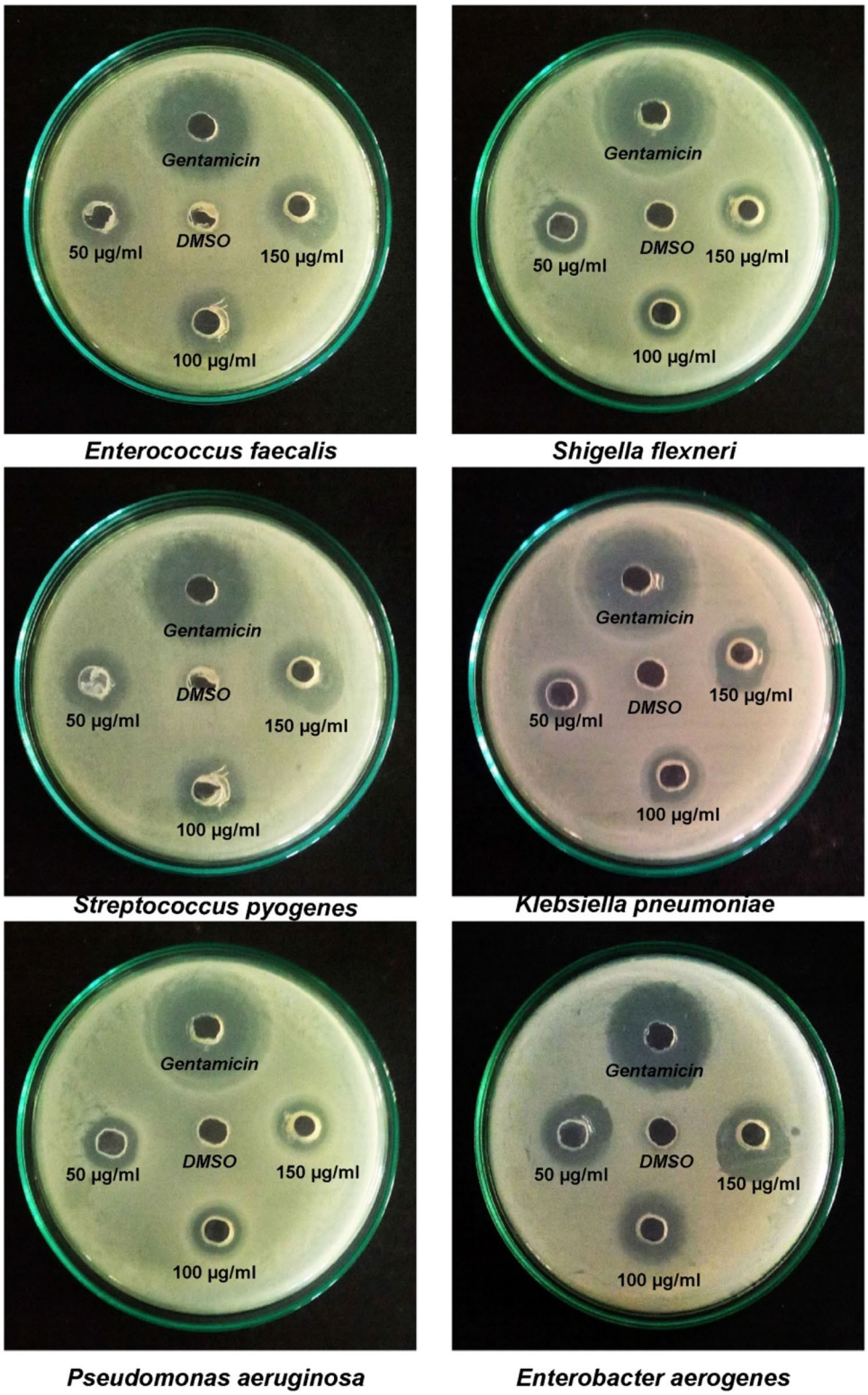

Fig. 4 Antibacterial activity of essential oil of *C. verum*

3.3 Minimal inhibitory concentration (MIC) determination of EOs

C. verum showed a similar MIC value (1.17 ± 0.34 µg/mL) against *S. flexneri*, *S. pyogenes*, and *K. pneumonia* and it was highest against *E. faecalis* (9.375 ± 0.95 µg/ml). *Klebsiella* sp. membrane was destroyed by inducing oxidative damage by *C. verum* bark essential oil [140]. Apart from bark, EOs obtained from flowers and leaves also act as strong antibacterial agents against both gram +ve and −ve bacteria

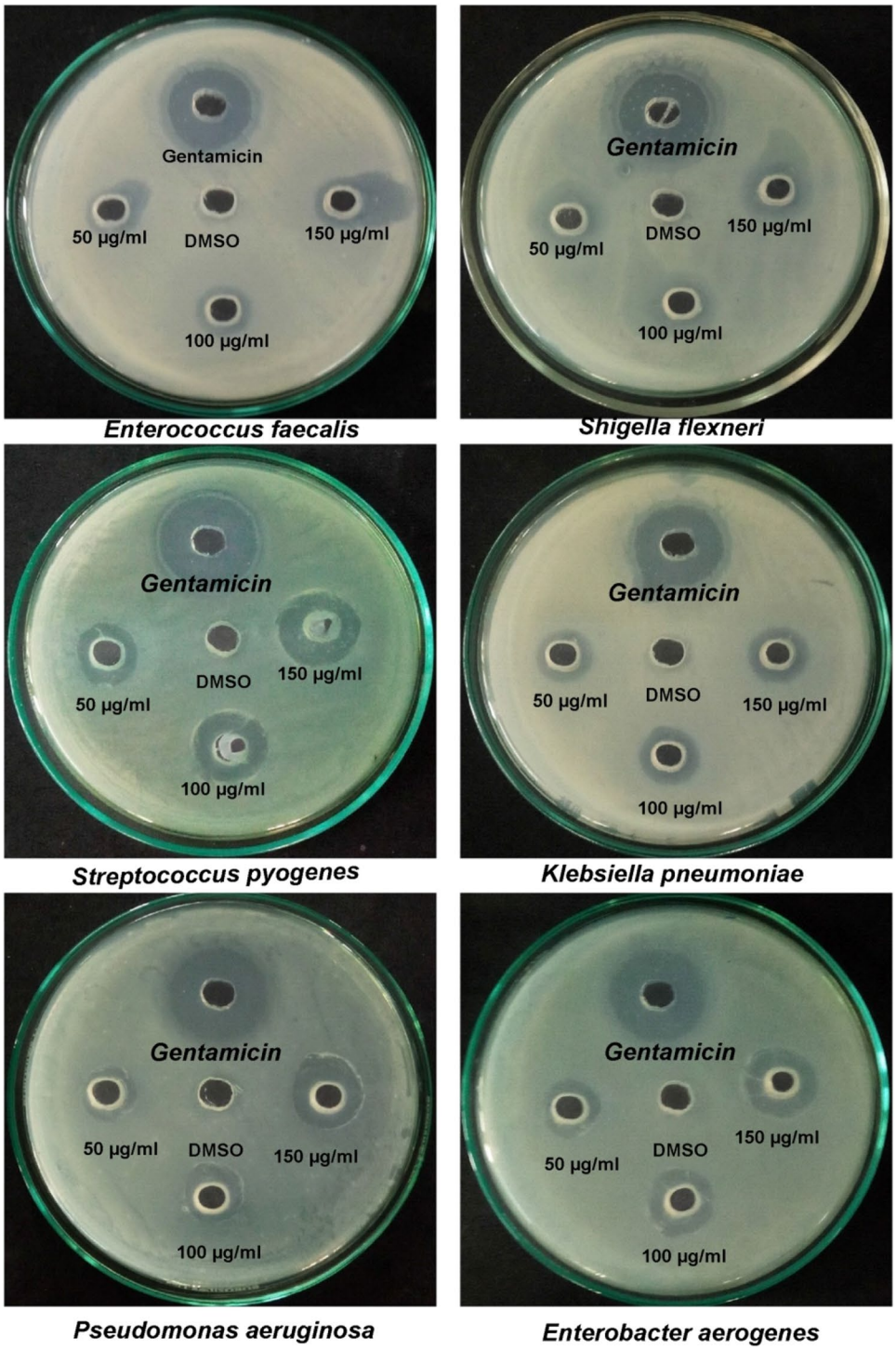

Fig. 5 Antibacterial activity of essential oil of *C. sulphuratum*

[141]. *C. glanduliferum* exhibited the least MIC value of 2.34 ± 0.67 µg/mL against *E. aerogenes. E. faecalis* and *S. pyogenes* exhibited the MIC value of 18.75 ± 0.63 µg/mL and 18.75 ± 0.46 µg/mL. The MIC value (3.43 µg/mL) was observed against *P. aeruginosa* and *C. glanduliferum*. *C. sulphuratum* showed minimum inhibition at concentrations ranging from 1.17 to 18.75 µg/mL against *S. pyogenes* and *E. aerogenes*, *S. flexneri*, *K. pneumonia*, and *P. aeruginosa*. The essential oil of *C. wightii* exhibited MIC values of 9.375 ± 0.87, 1.17 ± 0.97, 9.375 ± 1.23, etc. µg/mL against *E. faecalis*, *S. flexneri*, *S. pyogenes*, etc. Marasini et al. [142] reported MIC value of 49 µg/mL against *S. pyogenes* by *C. camphora* extracts however, the present study showed more inhibitory effect compared to previous studies. *C. camphora*

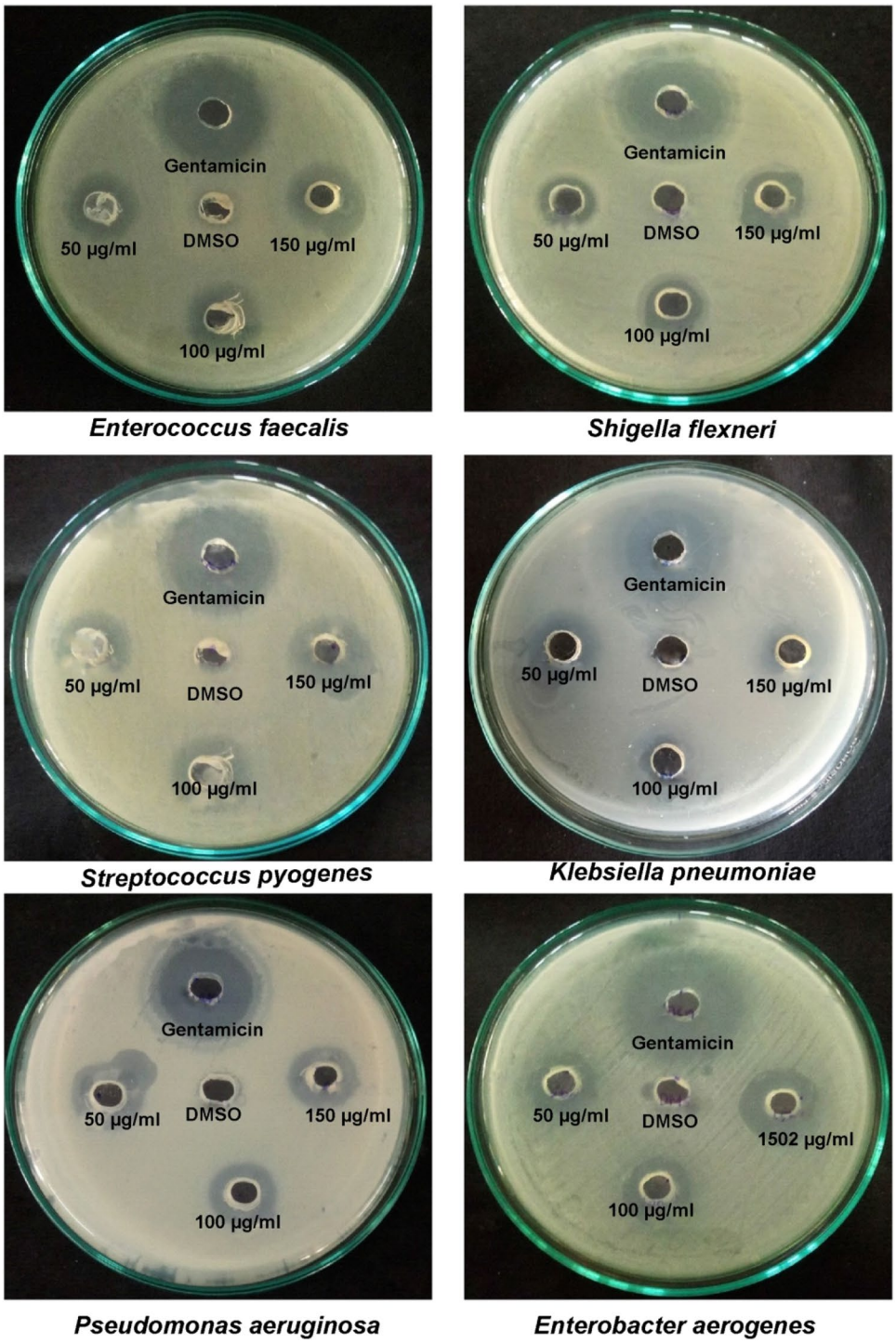

Fig. 6 Antibacterial activity of essential oil of *C. perotteti*

essential oil showed less inhibitory activity against *E. faecalis* and *Enterobacter aerogenes* compared to the other seven essential oils at higher concentrations (150 µg/mL). *C. perrottetii* exhibited varying minimum inhibitory concentrations against bacterial species with higher efficiency against *S. pyogenes* (2.34 µg/mL) and low efficiency against *K. pneumonia* with MIC value of 18.75 µg/mL. The most effective microbial inhibitory activity was observed in *C. malabatrum* leaf essential oil against *E. faecalis* and *S. pyogenes* with MIC value of about 1.17 µg/mL.

3.4 Free radical scavenging (antioxidant) activity of EOs

The antioxidant properties of the EOs were found promising, highly comparable with Standard (Ascorbic acid),

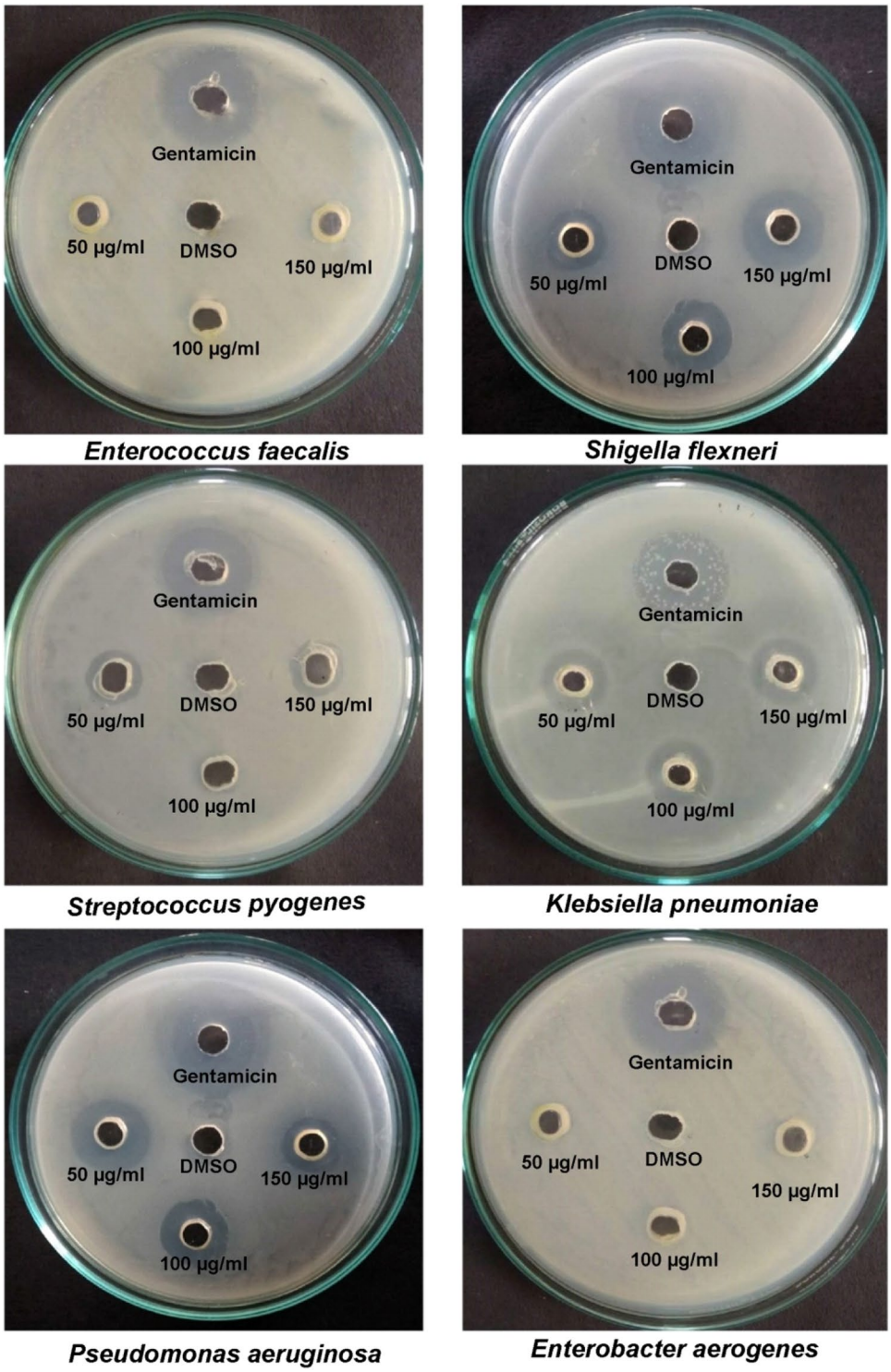

Fig. 7 Antibacterial activity of essential oil of *C. camphora*

and found to be significantly augmented with the augment in attentiveness. The antioxidant activity of EOs was recorded in terms of IC_{50} value and the minimum IC_{50} value corresponds to a greater potency [143]. All the EOs exhibited appreciable inhibition with IC_{50} values ranging 10.83–15.06 µg/mL which was secure to IC_{50} value (18.33 µg/mL) of the standard. *C. verum* was observed as a prospective foundation of antioxidants and recorded with the lowest IC_{50} value (10.83 µg/mL) followed by *C. perrottetii* (11.63 µg/mL), *C. glanduliferum* (13.02 µg/mL), *C. camphora* (13.04 µg/mL), *C. sulphuratum*(13.58 µg/ml), and *C. wightii* (15.06 µg/mL) (Fig. 11). However,

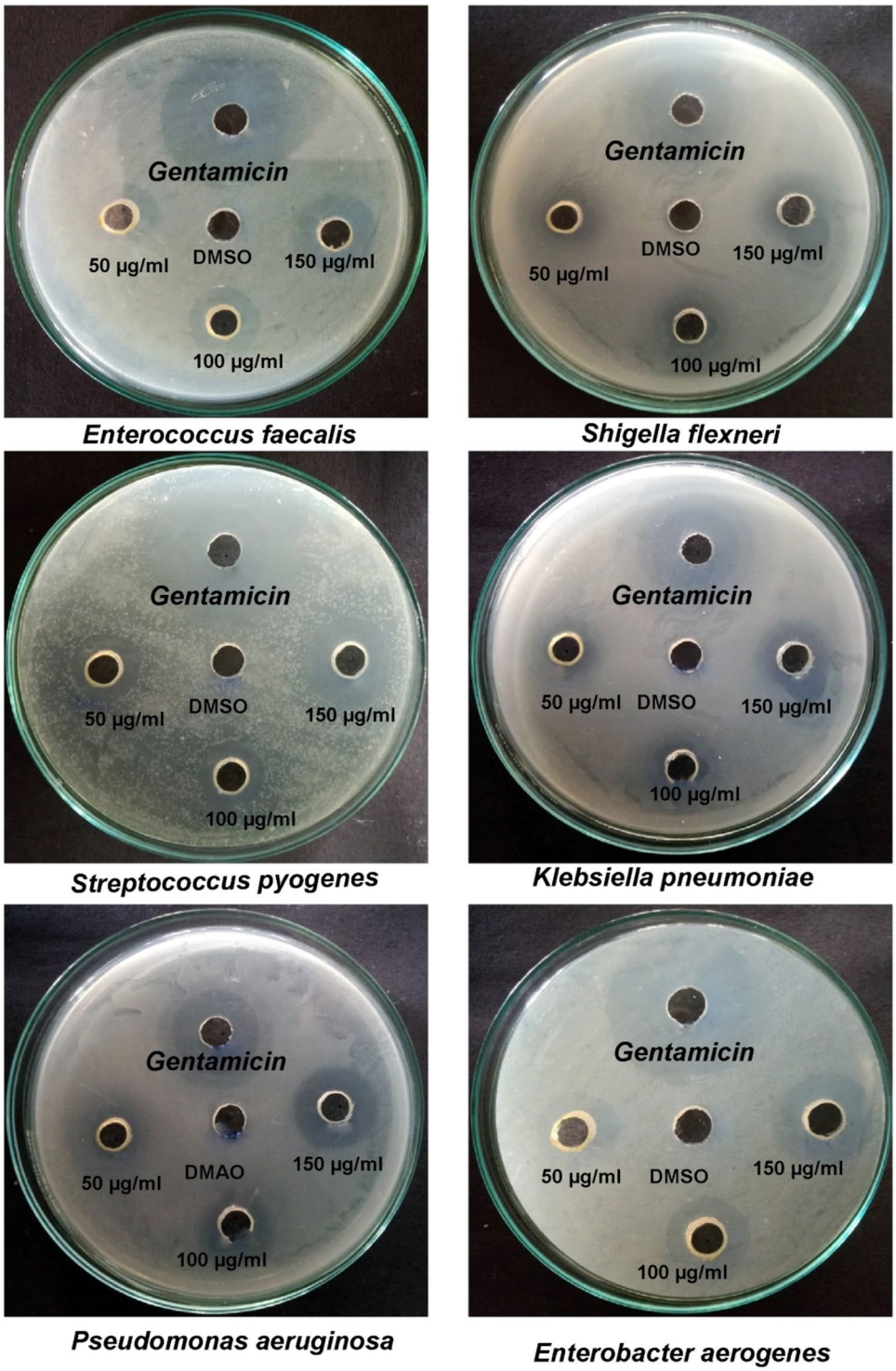

Fig. 8 Antibacterial activity of essential oil of *C.wightii*

C. malabatrum has recorded a higher IC_{50} value of about 15.86 µg/mL, since it was lower than standard (ascorbic acid).

The magnitude of antioxidant activity of volatile oils is generally attributed to the effects caused through the interaction of all phytoconstituents of the oil and also by the number of compounds that have been involved. Among all the oil samples studied, *C. verum* exhibited maximum antioxidant activity. Terpenes are considered to be a potential natural antioxidant and the main components of the aromatic plants Eos [144]. Thus, this movement could be accredited to the terpene oil content which is in agreement with the

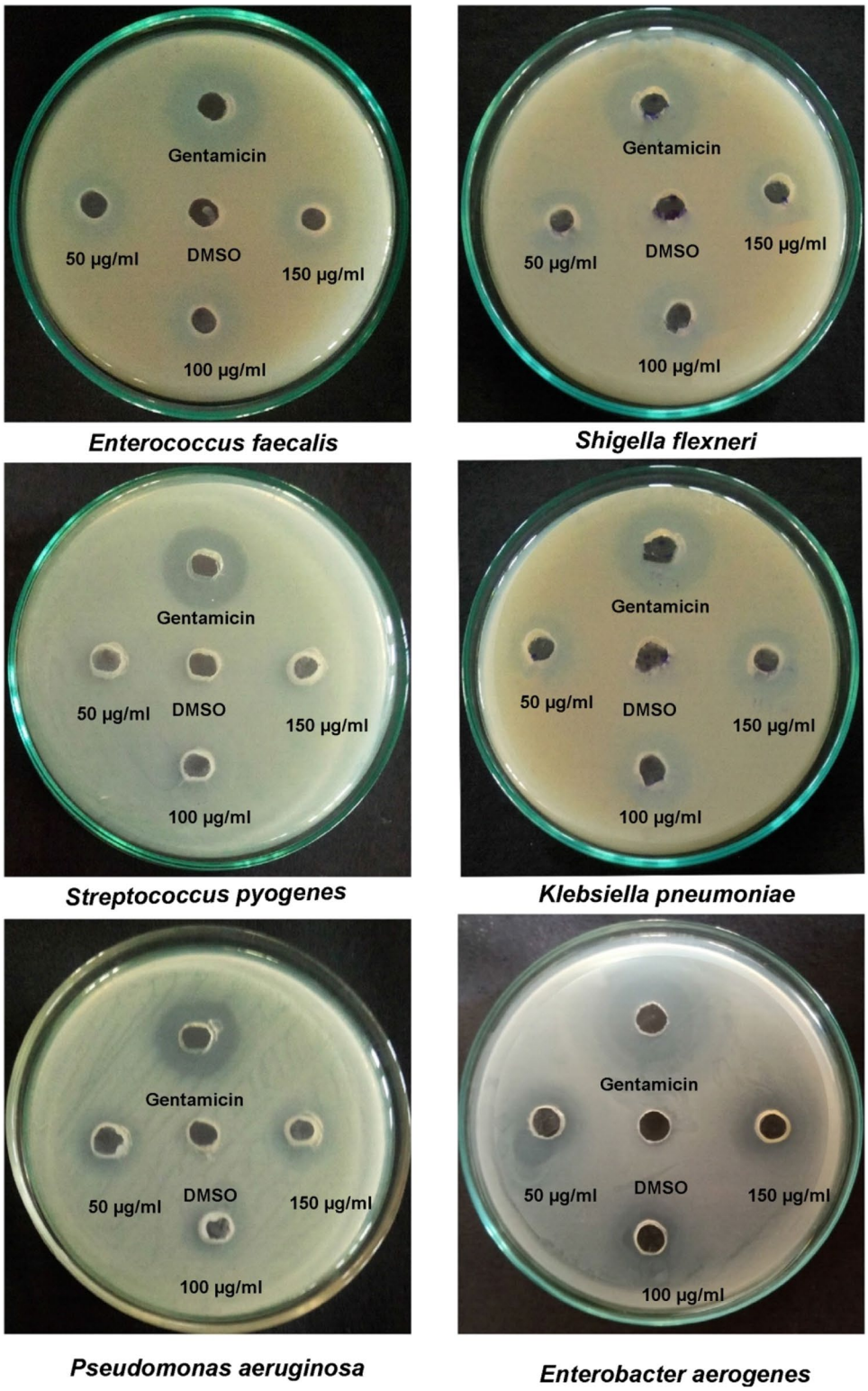

Fig. 9 Antibacterial activity of essential oil of *C. glanduliferum*

reports of Palozza and Krinsky [145]. GC-MS results of oils have also confirmed that sesquiterpenes were the principal compound of all the species studied, whereas, monoterpenes and phenylpropanoids were predominantly reported in *C. verum*.

DPPH scavenging activity was observed to be increased with an augment in the phenolic components such as flavonoids, phenolic diterpenes, and phenolic acids. Essential oils contain relatively higher amounts of phenolic compounds, which play a significant

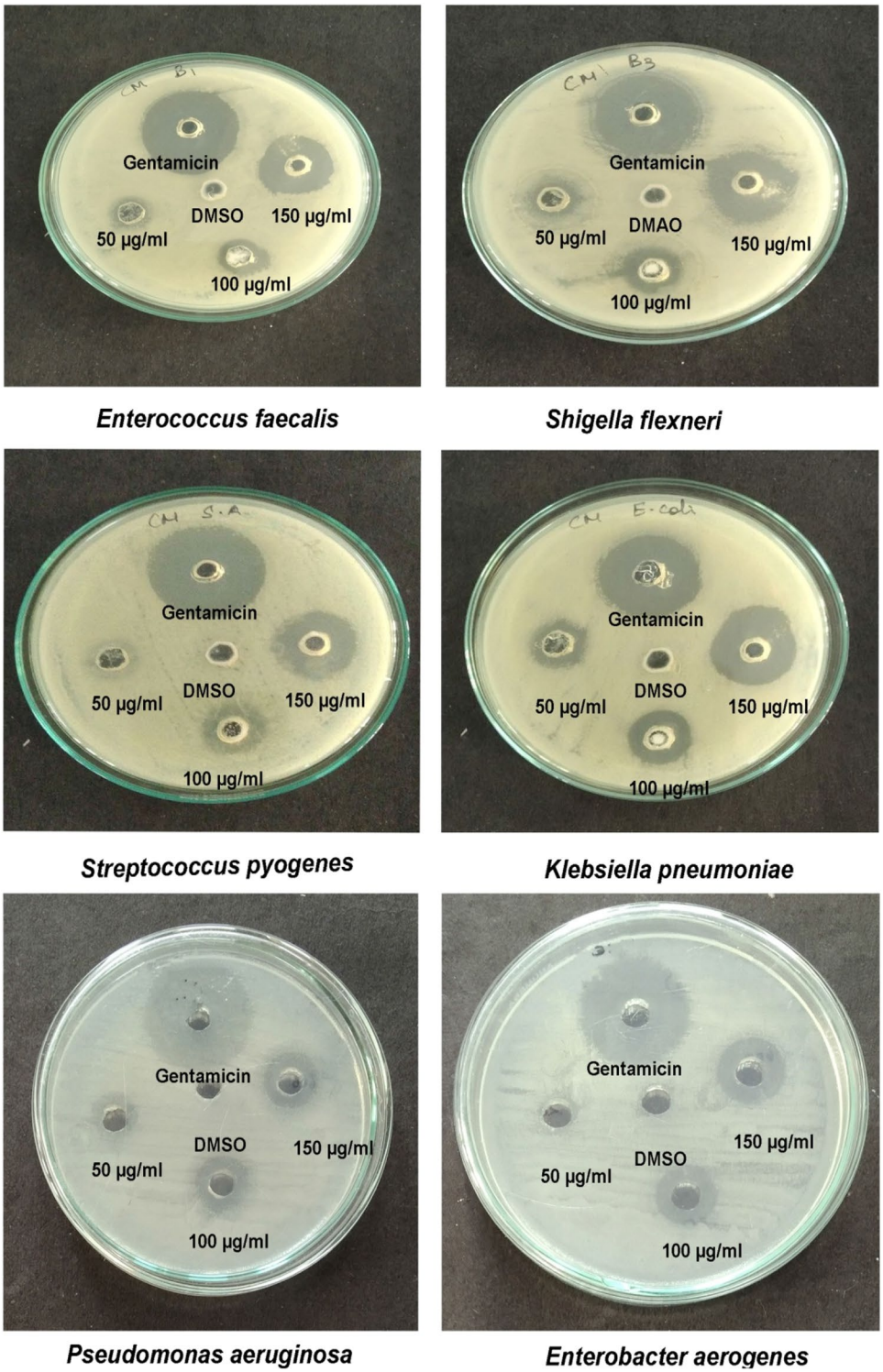

Fig. 10 Antibacterial activity of essential oil of *C.malabatrum*

responsibility in antioxidant activity by acting as H donors [146, 147] In vitro studies have confirmed the antioxidant effects of EOs. However, previous reports suggested that synergistic interaction between major and minor compounds attributed to the hydrogen-donating capacity which results in the antioxidant activity of monoterpenes [148, 149] sesquiterpenes [150], phenolic compound, Eugenol [74, 151–154] reported with significant concentrations. Several studies have proven the potency of the phenolic derivative in which eugenol has the most powerful antioxidant activity and radical-scavenging activity [155, 156].

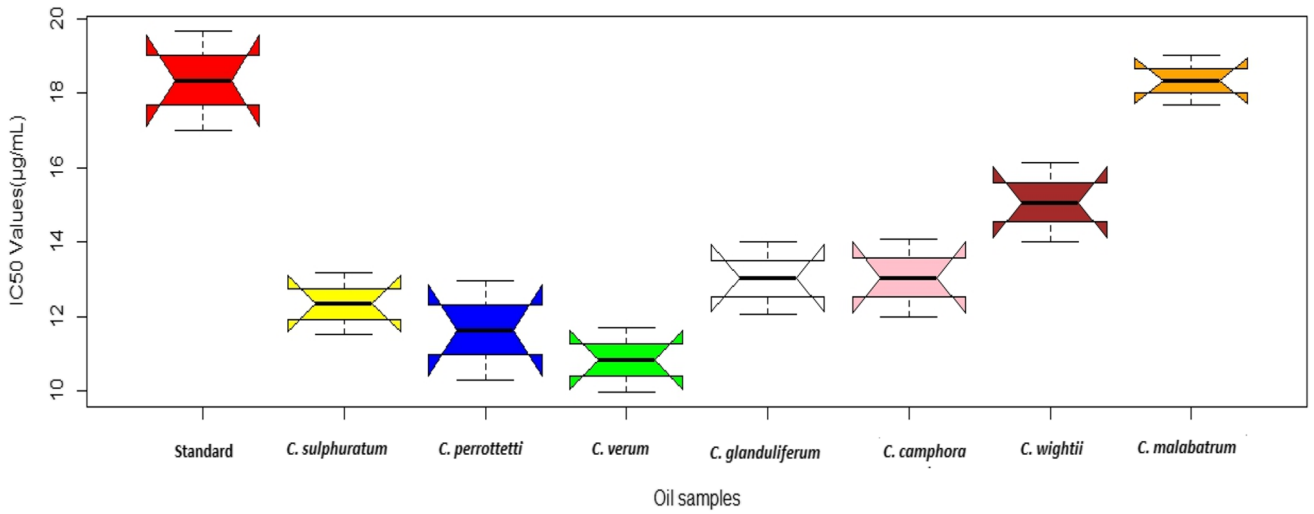

Fig. 11 Total antioxidant activity of essential oils based on IC-50 value

3.5 Larvicidal activity of EOs of *Cinnamomum* spp.

The toxicity effects of EOs of seven *Cinnamomum* species were analyzed beside fourth instar larvae of *Aedes aegypti*. All the oil samples exhibited promising larvicidal activity and the results were recorded with statistical data regarding LC_{50}, LC_{90}, and confidence limits by probit analysis using PROBIT software SPSS. The mortality percentages of third instar larvae of *A. aegypti* tested against essential oils were recorded as dose-dependent by increased with increasing concentrations. The lethal efficiency of oils in the concentration with 90% mortality (LC_{90}), 50% death (LC_{50}), and 95% confidence period values were calculated. The positive control temephos showed LC_{50} and LC_{90} values at 0.021 µg/mL (0.020–0.023) and 0.042 µg/mL (0.035–0.05). Negative control (DMSO) has not shown mortality and is active at ≤ 10 h exposure. The strong larvicidal activity was observed in *C. verum* with the most effective LC_{50} and LC_{90} values of 101.87 µg/mL (56.21–147.54 µg/mL) and 266.34 µg/mL (181–351.78 µg/mL) respectively. *C. sulphuratum* exhibited 104.579 µg/mL (66.39–141.05 µg/mL) and 242.61 µg/mL (159.544–325.687 µg/mL) LC_{90} and LC_{50} values respectively (Table 4).

The oils of *C. perrottetii* and *C. camphora* showed an almost similar susceptibility rate against the larvae of *A. aegypti*. *C. perrottetii* has shown moderate activity whereas *C. wightii* showed mild larvicidal activity with LC_{50} and LC_{90} values. Among the evaluated samples, *A. aegypti* exhibited more susceptibility to essential oils of *C. glanduliferum* with higher LC_{50} and LC_{90} values. The oil of *C. verum* has shown more sensitive to larvicidal activity with a high percentage of mortality. The consequence of the differentiations between the control and test studies was evaluated through one-way ANOVA, and p-value < 0.05 which were considered highly important. *C. malabatrum* oil had an LC_{50} value of 105.54 µg/mL with a lower assurance boundary (LCC) of 81.945 µg/mL and an higher assurance limit (UPL) of 128.545 µg/mL concentrations. LC_{90} value of 253.977 µg/mL (180.196–631.575) were obtained (Table 4).

There were only a few studies conducted on the larvicidal efficiency of EO in selected *Cinnamomum* species [141, 157, 158]. The effectiveness of these oils with a larvicidal property must be emphasized by oils the chemical compositions. In most of the oils of *Cinnamomum*, the dominant constituents were found as terpene derivatives

Table 4 Mosquito larvicidal potentiality of essential oils of *Cinnamomum* spp. against *A. aegypti*

Sl. No	Name of the species	LC 50 (µg/mL) (LCL–UCL)	LC 90 (µg/mL) (LCL–UCL)
1.	***C. sulphuratum***	104.579 (66.39–141.05)	242.61 (159.544–325.687)
2.	***C. perrottetii***	138.228 (103.98–172.47)	282.03 (233.24–330.82)
3.	***C. verum***	101.87 (56.21–147.54)	266.34 (181–351.78)
4.	***C.wightii***	253.45 (154.814–352.08)	615.11 (27502–955.02)
5.	***C.camphora***	138.83 (104.33–173.32)	263.64 (195.47–331.81)
6.	***C.glanduliferum***	689.34 (216.09–1162.59)	1510.79 (320.29–2701.29)
7.	***C. malabatrum***	105.54 (81.945–128.545)	253.977 (180.196–631.575)
7	**Temephos**	0.021 (0.020–0.023)	0.042 (0.035–0.05)

and phenylpropanoids, which are responsible for larvicidal potentiality. Similar results were reported in essential oils with a higher content of phenylpropanoids [159], eugenol [160–163] and sesquiterpenes [164]. The larvicidal activity of a few species of *Cinnamomum* was corroborated with the present findings [158, 165–173].

Efficient larvicidal activity of larval and adult individuals of *A. aegypti* essential oil-based terpene compounds. Moreover, the structural characteristics of essential oils also contributed considerably to the consideration of larvicidal activity. The structural and activity associations of monoterpenes [174] and eugenol [162] derivatives alongside *A. aegypti* were supported by present findings. The results of our current study have demonstrated the potential alternative sources of mosquito larvicides, therefore, the plant-based essential oil compounds are competent when compared to existing synthetic chemicals which can be used to control the population of *Aedes aegypti*. Primarily essential oils affect the midgut epithelium and secondarily the malpighian tubules and gastric caeca in the mosquito larvae [175, 176] and also through physiology paralyzing and osmoregulation systems of the organisms [177].

4 Conclusion

The current study recorded the list of phytocompounds from the leaf EOs of seven species of *Cinnamomum*, *C. sulphuratum, C. perotettii, C. verum, C. wightii, C. camphora, C. glanduliferum*, and *C. malabatrum*. The GC-MS analysis of Eos indicated that the highest amount of eugenol and shyobunol was recorded in *C. verum* and *C. perrottetii*. In antibacterial activity, the higher zone of inhibition of Eos of *C. malabatrum* showed against *S. pyogenes* and *E. faecalis* followed by *C. verum* has shown a notable inhibition zone against *K. pneumoniae* . The total antioxidant assessed by IC50 values and larvicidal activities against *Aedes aegypti* indicated the biomedical efficiency of all seven species studied. Due to its higher efficiency, the multipurpose tree *C. verum* can be cultivated as a commercial crop instead of utilizing natural resources for the pharmaceutical industry for the development of drugs. Further, this research can be extended with in vivo and in vitro animal studies to confirm the potentiality of seven species of *Cinnamomum* in the field of biomedicine to detect novel medicines.

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Author contribution Saranya Surendran; execution of laboratory studies, preparation of the manuscript; Smija, K.P: execution and assistance of laboratory studies; Raju Ramasubbu: planning of research problem, work designed, lead, review and editing of the manuscript, Azhagu Madhavan Sivalingam and Arjun Pandian reviewed, edited revised article.

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Declarations

Conflict of interest The authors declare no competing interests.

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