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GC-MS ANALYSIS OF OIL FROM *LAVANDULA LATIFOLIA* L. AND ITS REPELLENT ACTIVITY AGAINST MOSQUITO

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ABSTRACT: Mosquitoes are one of the important vectors of various diseases. Repellent activity significantly reduces contact with disease-transmitting mosquitoes. Essential oils have repellent activity that is being potentially developed as an alternative to commercial chemical pesticides. In this study, *Lavandula angustifolia* was used to evaluate its repellent activity against *Ae. Aegypti*. The repellent activity of *L. angustifolia* oil was tested with the 100, 200, 300, 400, and 500 ppm concentrations against the adult mosquitoes of *Ae. Aegypti*. *L. angustifolia* showed 100% protection up to 150 minutes against the bite of *Ae. aegypti*. After that, the repellency was found reduced by the increased exposure periods against *Ae. Aegypti*. The presence of phytochemicals in oils could be responsible for different mosquitocidal agents. Lavender oil was subjected to GC-MS analysis. It showed the presence of 37 compounds, including, 1R- α -Pinene, Bicyclo[2.2.1]heptane, 2,2-dimethyl-5methylene-, Tricyclo[2.2.1.0 (2,6)]heptane, 1,7,7-trimethyl-, Ocimene, Camphene, α -Pinene, Cyclohexene, 4-methylene-1-(1-methylethyl)-, Eucalyptol, 1,4-Cyclohexadiene, 1-methyl-4-(1-methylethyl)-, Bicyclo [2.2.1]heptan-2-one, 1,3,3-trimethyl-, 3-Oxatricyclo[4.1.1.0(2,4)] octane, 2,7,7-trimethyl-, Acetaldehyde, (3,3-dimethylcyclohexylidene)-, (E)-, Bicyclo[2.2.1]heptan-2-one, 1,7,7-trimethyl-, (1S)-, Isoborneol, Borneol, Cyclohexanol, 1-methyl-4-(1-methylethylidene)-, Tetra-cyclo[6.3.2.0(2,5).0(1,8)]tridecan-9-ol, 4,4-dimethyl-, 1,6,10,14-Hexadecatetraen-3-ol, 3,7,11,15-tetramethyl-, (E,E)- and 13-Hexyloxacyclotridec-10-en-2-one.

INTRODUCTION: Plant essential oils are fashioned commercially from plant families. Generally, these plant oils possess the complex mixtures of several groups of phytoconstituents, such as monoterpenes biogenetically related phenols and sesquiterpenes. The use of oil in India is already there from time immemorial.

Certain plant oils confirm that some plant oils not only repel insects but have contact and fumigant insecticidal actions against a variety of pests, pathogens, and target organisms¹. Essential oils possess a broad spectrum of biological activities, including anti-microbial, insect repellent, and herbicidal activity².

Plant essential oils have been used on various species of mosquitoes, including *Culex*, *Anopheles*, and *Aedes*. They have been used as larvicides, repellents, adulticides, and ovicides³. Many plant families found larvicides against *Ae. Aegypti*, including Cupressaceae⁴, Piperaceae⁵, Lauraceae⁶, Asteraceae⁷ and Labiatae⁸.

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Several plant oils possess several phytochemicals, which can act as mosquitocidal agents. The present investigation was aimed to assess the chemical components of Lavender, *Lavandula latifolia* L. oil and its repellent activity against *Ae. Aegypti*.

MATERIALS AND METHODS:

The Essential Oil from *Lavandula latifolia*: In this study, *Lavandula latifolia* oil was extracted from the sample as described previously. The extracted oil was stored in an amber-colored bottle for further analysis⁹.

Repellent Activity: The repellent activity of three-days-old blood-starved female mosquitoes, *Ae. aegypti* (100 nos.) was kept in an adult mosquito rearing cage (40 cm × 30 cm × 40 cm). The volunteer had no contact with lotions, perfumes, or perfumed soaps on the day of the assessment. The arms of volunteers, only 25 cm² dorsal side of the skin on each arm, were exposed, and the remaining area covered by rubber gloves. The crude extract was applied at 100 - 500 mg/cm² separately in the exposed area of the forearm. Only methanol served as control. The time of the test depended on whether the target mosquitoes bite in the day or night. *Ae. aegypti* was tested from 07.00 to 17.00 h. The control and treated arm were introduced concurrently into the experimental cage, and gently tapping the sides on the experimental cages, the mosquitoes were activated. Each test concentration replicated five times. The volunteer conducted their test of each concentration by inserting the treated and control arm into the same cage for one full minute for every five minutes. The mosquitoes that landed on the hand were recorded and then shaken off before imbibing any blood, making out a 5 minutes protection. The percentage (%) of repellency was calculated by the following formula¹⁰.

$$\% \text{ Repellency} = [(T_c - T_t)/T_a] \times 100$$

Where T_c is the number of adults in the control group, and T_t is the number of adults in the treated group.

Gas Chromatography Analysis: The volatile oil was analyzed using PerkinElmer Clarus 500GC equipped with a Flame Ionization Detector (FID). The test was carried out on a capillary column Elite-5 (Column length 30mm; Column id: 250 μ m;

Crossband 5% Phenyl 95% dimethylpolysiloxane). Helium was employed as a carrier gas at a flow rate of 1 mL/min. Injector and detector temperatures were set at 250 °C and 280 °C, respectively. The oven temperature was kept at 60 °C then gradually raised to 240 °C at 3 °C / min and finally held isothermally for 54.5 min. One microliter of the diluted samples (1/100 in hexane, v/v) were injected manually (split mode, split ratio 1:20). Calculation of peak area percentage was performed on the basis of the FID signal using the Turbomass software (ver 5.2.0).

Identification of Compounds: The identification of the compounds was done based on the comparison of retention indices and mass spectra of most of the compounds with data generated under identical experimental conditions by applying a two-dimensional search algorithm, considering the retention index, as well as mass spectral similarity¹¹, or with those of authentic compounds available in NIST 2005 libraries. Moreover, special software, namely TurboMass software (Ver5.2.0) was used for the processing and interpretation of mass spectra with several commercially available libraries included.

RESULTS AND DISCUSSION:

Repellent Activity of *Lavandula angustifolia*: The repellent activity of *L. angustifolia* oil was tested with the 100, 200, 300, 400, and 500 ppm concentrations against the adult mosquitoes of *Ae. aegypti*, the data pertaining to the experiments are represented in the table. It was observed that *L. angustifolia* showed 100% protection up to 150 minutes against the bite of *Ae. aegypti*. After that, the repellency was found reduced by the increased exposure periods against the selected mosquitoes **Table 1**. The presence of phytochemicals in oils could be responsible for different mosquitocidal agents. Furthermore, while experimenting, the application of oil with the respective concentration formed a thin film over the surface of the experimental cups, suggesting that the oil film might prevent the further exchange of gases in the medium of experimental cups, thereby causing the death of the larvae. Our present findings are going in hand in hand with the earlier findings of several authors. Jude et al.,¹² evaluated the larvicidal and repellent effect of the essential oil from the seeds and leaves of *Chenopodium ambrosioides* against

the larvae and adults of *An. gambiae* mosquitoes. Chen et al.,¹³ determined the larvicidal activity of essential oil derived from *Clinopodium gracile* aerial parts against the larvae of *Ae. Albopictus*. Liu¹⁴ determined the larvicidal activity of the essential

oil derived from *Illicium henryi* leaf and steamed against the larvae of *Ae. albopictus*. Eliningaya et al.,¹⁵ evaluated the larvicidal efficacy of eight volatile components of essential oils against third instar larvae of *An. gambiae*.

TABLE 1: REPELLENT ACTIVITY OF LAVENDULA ANGUSTIFOLIA TESTED AGAINST THE ADULTS OF SELECTED VECTOR MOSQUITO

Conc. ($\mu\text{g}/\text{cm}^2$)	% of repellency							
	Post application of repellent (min)							
	30	60	90	120	150	180	210	240
100	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	41.2 \pm 1.7	36.4 \pm 1.3	23.7 \pm 1.3
200	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	47.3 \pm 1.4	39.6 \pm 1.1	30.1 \pm 1.5
300	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	61.3 \pm 1.6	51.3 \pm 2.7	45.7 \pm 2.7
400	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	73.7 \pm 1.7	68.6 \pm 2.8	51.1 \pm 2.4
500	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	100.0 \pm 0.0	81.5 \pm 1.1	70.3 \pm 2.5	65.1 \pm 2.3

Values are mean \pm S.D of five replications

GC-MS Analysis of Essential Oils: Lavender oil was injected into the GC-MS spectrogram column. The column was washed according to the manual. It showed the presence of 37 different compounds, including 1R- α -Pinene, Bicyclo[2.2.1]heptane, 2,2-dimethyl-5methylene-, Tricyclo [2.2. 1.0 (2,6)]heptane, 1, 7, 7-trimethyl-, Ocimene, Camphene, α -Pinene, Cyclohexene, 4-methylene-1-(1-methylethyl)-, Eucalyptol, 1,4-Cyclohexadiene, 1-methyl-4-(1-methylethyl)-, Bicyclo[2.2.1]heptan-2-one, 1, 3,3-trimethyl-, 3-Oxatricyclo[4.1.1.0(2,4)]octane, 2,7,7 trimethyl-, Acetaldehyde, (3,3-dimethylcyclohexylidene)-, (E)-, Bicyclo[2.2.1]heptan-2-one, 1,7,7-trimethyl-, (1S)-, Isoborneol, Borneol, Cyclohexanol, 1-methyl-4-(1-methylethylidene)-, Tetracyclo[6.3.2.0(2,5).0(1,8)]tridecan-

9-ol, 4,4-dimethyl-, 1,6,10,14-Hexadecatetraen-3-ol, 3,7,11,15-tetramethyl-, (E,E)- and 13-Hexyloxacyclotridec-10-en-2-one **Fig. 1, Table 2.** The presence of 1,6-Octadien-3-ol, 3,7-dimethyl in *L. latifolia* belongs to terpenes. This finding is in agreeing with the earlier findings of several authors. The oil is effective and possesses strong antibacterial, antifungal, carminative, sedative, anti-depressive, and useful for burns and insect bites. Essential oils are versatile of many different aroma compounds. The analysis of Lavandula oils with typical scents has demonstrated that due to the level of linalool, linalyl acetate, and various sesquiterpenes. These essential oil components showed antimicrobial and larvicidal activity¹⁶⁻¹⁹.

TABLE 2: CHEMICAL COMPOSITION OF LAVANDULA ANGUSTIFOLIA OIL

Peak	Compound name	Formula	Retention Time	%Peak Area
1	1R- α -Pinene	C ₁₀ H ₁₆	4.61	0.0091
2	Bicyclo[2.2.1]heptane, 2,2-dimethyl-5methylene-	C ₁₀ H ₁₆	5.12	0.0074
3	Tricyclo[2.2.1.0(2,6)]heptane, 1,7,7-trimethyl-	C ₁₀ H ₁₆	5.59	0.6032
4	Ocimene	C ₁₀ H ₁₆	6.08	10.7845
5	Camphene	C ₁₀ H ₁₆	6.48	7.9248
6	α -Pinene	C ₁₀ H ₁₆	7.29	5.4178
7	α -Pinene	C ₁₀ H ₁₆	7.65	2.8714
8	Cyclohexene, 4-methylene-1-(1-methylethyl)-	C ₁₀ H ₁₆	8.06	0.2562
9	Eucalyptol	C ₁₀ H ₁₈ O	9.24	33.8367
10	1,4-Cyclohexadiene, 1-methyl-4-(1-methylethyl)-	C ₁₀ H ₁₆	10.09	0.5678
11	Bicyclo[2.2.1]heptan-2-one, 1,3,3-trimethyl-	C ₁₀ H ₁₆ O	11.22	0.0915
12	3-Oxatricyclo[4.1.1.0(2,4)]octane, 2,7,7trimethyl-	C ₁₀ H ₁₆ O	11.65	0.0283
13	Acetaldehyde, (3,3-dimethylcyclohexylidene)-, (E)-	C ₁₀ H ₁₆ O	11.96	0.4926
14	Bicyclo[2.2.1]heptan-2-one, 1,7,7-trimethyl-, (1S)-	C ₁₀ H ₁₆ O	14.23	10.8711
15	Isoborneol	C ₁₀ H ₁₈ O	14.60	17.6978
16	Borneol	C ₁₀ H ₁₈ O	15.22	3.0625
17	Bicyclo[2.2.1]heptan-2-one, 1,7,7-trimethyl-, (1S)-	C ₁₀ H ₁₆ O	15.94	0.1567
18	trans-2-Pinanol	C ₁₀ H ₁₈ O	16.14	0.6746
19	Cyclohexanol, 1-methyl-4-(1-methylethylidene)-	C ₁₀ H ₁₈ O	16.34	0.2057

20	Fenchyl acetate	C ₁₂ H ₂₀ O ₂	16.78	1.1904
21	Cyclohexanone, 4-hydroxy-4-methyl-	C ₇ H ₁₂ O ₂	19.05	0.0164
22	Acetic acid, 1,7,7-trimethyl-bicyclo[2.2.1]hept-2-yl ester	C ₁₂ H ₂₀ O ₂	19.60	0.5745
23	trans-Z- α -Bisabolene epoxide	C ₁₅ H ₂₄ O	21.08	0.0137
24	Copaene	C ₁₅ H ₂₄	23.40	0.0057
25	Caryophyllene	C ₁₅ H ₂₄	25.40	2.0544
26	α -Caryophyllene	C ₁₅ H ₂₄	26.75	0.1536
27	cis-Z- α -Bisabolene epoxide	C ₁₅ H ₂₄ O	30.84	0.0119
28	Caryophyllene oxide	C ₁₅ H ₂₄ O	32.12	0.1921
29	12-Oxabicyclo[9.1.0]dodeca-3,7-diene,1,5,5,8-tetramethyl-, [1R (1R*,3E,7E,11R*)]-	C ₁₅ H ₂₄ O	33.19	0.0096
30	Tetracyclo[6.3.2.0(2,5).0(1,8)]tridecan-9-ol, 4,4-dimethyl-	C ₁₅ H ₂₄ O	34.48	0.0177
31	exo-2-Hydroxycineole	C ₁₀ H ₁₈ O ₂	36.36	0.0083
32	Cyclopentanecarboxylic acid, 3-isopropylidene-, bornyl ester	C ₁₉ H ₃₀ O ₂	42.16	0.0055
33	2,6,11,15-Tetramethyl-hexadeca-2,6,8,10,14-pentaene	C ₂₀ H ₃₂	43.19	0.0060
34	1,6,10,14-Hexadecatetraen-3-ol, 3,7,11,15-tetramethyl-, (E,E)-	C ₂₀ H ₃₄ O	43.87	0.0036
35	2,6,11,15-Tetramethyl-hexadeca-2,6,8,10,14-pentaene	C ₂₀ H ₃₂	45.00	0.1269
36	1,6,10,14-Hexadecatetraen-3-ol, 3,7,11,15-tetramethyl-, (E,E)-	C ₂₀ H ₃₄ O	48.19	0.0068
37	13-Hexyloxacyclotridec-10-en-2-one	C ₁₈ H ₃₂ O ₂	48.60	0.0432
	Total percentage of chemical compositions			100.0000

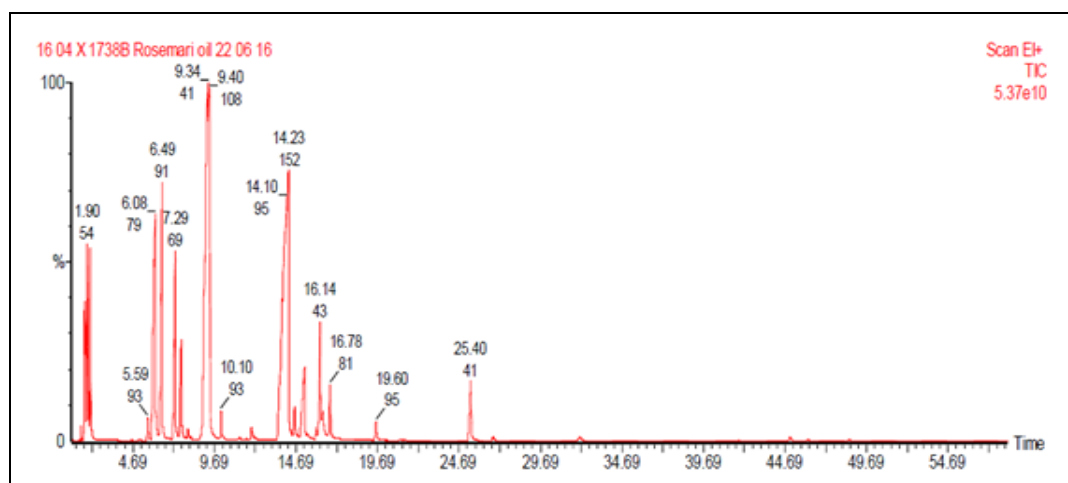


FIG. 1: GC-MS CHROMATOGRAM OF LAVANDULA ANGUSTIFOLIA OIL

CONCLUSION: In conclusion, the oil of *Lavandula angustifolia* showed potential repellent activity against *Ae. Aegypti*. Essential oils from *L. angustifolia* showed activity at deficient concentrations and is the most promising activity. Results revealed that Ocimene, Eucalyptol, Bicyclo[2.2.1]heptan-2-one, 1,7,7-trimethyl-, (1S)- are the significant components significantly responsible for the repellent activity. Essential oil *L. angustifolia* repellent activity is being potentially developed as an alternative to commercial chemical pesticides.

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CONFLICTS OF INTEREST: None of the conflict of interest.

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