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Research

Gas Chromatography-Mass Spectrometry Analysis of Hydroalcoholic extract of *Calliandra haematocephala* leaves

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	Abstract
Published on: 22 July 2025	<p><i>Calliandra haematocephala</i> belongs to Fabaceae, is an ornamental shrub. It is native to Bolivia, but widely cultivated in India. It is traditionally used for treating infections, inflammation, neurological disorders and metabolic diseases. Phytochemical survey revealed the presence of flavanoids, terpenoids, saponins, phenolic compounds, glycosides, and alkaloids. These compounds exhibit a wide range of pharmacological activities such as antimicrobial, antioxidant, anti-inflammatory, neuroprotective, antidiabetic, anticancer, and hepatoprotective effects. Leaves were collected, washed with water, shade dried, powdered and extracted with 70% hydroalcohol. The extract was concentrated and stored in air tight container for further use. This study aims to identify the bioactive constituents present in the hydroalcoholic extract of <i>Calliandra haematocephala</i> (HAECH_L) using Gas Chromatography-Mass Spectrometry (GC-MS). GC-MS chromatogram showed phytoconstituents which may attribute to eighteen phytoconstituents, which may contribute to the plant's diverse pharmacological activities such as, treating infections, pain, inflammatory and neurological disorders.</p>
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	Keywords: Gas Chromatography-Mass Spectrometry (GCMS), <i>Calliandra haematocephala</i> .

INTRODUCTION

Calliandra haematocephala, belongs to Fabaceae, is an ornamental shrub commonly known as Red Powder Puff plant. It is native to Bolivia, but widely cultivated in India[1]. Its traditional use in treating infections, pain, inflammatory conditions, and neurological disorders, particularly in southern India[2] In Tamil Nadu and Kerala leaf and flower preparations for treating skin infections, wounds, fever, and inflammatory disorders[3]. In

some parts of northeastern India, indigenous users rely on aqueous leaf extracts for gastrointestinal issues and as a general tonic [4]. In central and south America, Nigeria, Colombia people used leaf decoction to treat diabetics [5]. In Nigeria Yoruba, Nupe people decoction of leaf is used to treat malaria [6]. In south western Nigeria Tude people used antioxidant and blood purifier [7]. In China, Jinjiang, Yunnan people leaf, flower and bark used by tranquilizing effect [8]. Phytochemical survey revealed the presence of flavanoids, terpenoids, saponins, phenolic compounds, glycosides and alkaloids [9]. It exhibited pharmacological activities such as, antimicrobial [10], antioxidant and anti-inflammatory [11], antidiabetic [12], hepatoprotective [13]. The study is to analyse the phytoconstituents in hydroalcoholic extract of *C. haematocephala* leaves using Gas Chromatography - Mass Spectrometry (GC-MS).

Collection of leaves and authentication

Leaves were collected from the Alagar foot Hills, Madurai district, Tamil Nadu during the March 2025. It was authenticated by Dr. Stephen, Professor of Botany, The American College, Madurai-625002. A herbarium of this specimen was deposited in the department for future reference.

Preparation of hydroalcoholic extract of *Calliandra haematocephala* (HAECH_L)

The collected leaves were rinsed with water, shade-dried and grounded into a fine powder. 5g of powder was macerated in 70% hydroalcohol until exhaustive extraction, after which the mixture was filtered and the filtrate concentrated under reduced pressure to dryness and the percentage was found to be 12 %w/w and stored in air tight container.

Gas chromatography- mass spectroscopy analysis

The chemical profile of the hydroalcoholic leaf extract of *Calliandra haematocephala* was elucidated by Gas Chromatography–Mass Spectrometry (GC–MS) using a Shimadzu QP-2020. This hyphenated technique, which marries the separation power of gas chromatography with the compound-identification capability of mass spectrometry, is widely employed for both qualitative and quantitative analysis of volatile and semi-volatile phytoconstituents in medicinal plants.

Column

A fused-silica column (SH-Rxi-5 Sil MS, 30 m × 0.25 mm i.d., 0.25 µm film thickness) was employed, with helium as the carrier gas at a constant flow rate of 1 mL/min, and the injector temperature maintained at 280 °C.

Condition

A 1 µL aliquot of the hydroalcoholic extract was injected, and the oven was programmed at 50 °C for 3 minutes, then ramped to 180 °C at 15 °C min⁻¹.

Mass detector

The mass spectrometer was operated with a transfer line temperature of 290 °C and an ion source temperature of 230 °C, using 70 eV electron impact ionization. Scans were recorded from m/z 50 to 600 Da with a scan time of 0.2 s and an inter-scan interval of 0.1 s. Component spectra were identified by comparison with the NIST 2017 GC–MS library.

RESULTS AND DISCUSSION

GC–MS analysis revealed eighteen bioactive constituents are given Table 1 and their corresponding biological activities are presented in Table 2, with the chromatograms Fig. 1.

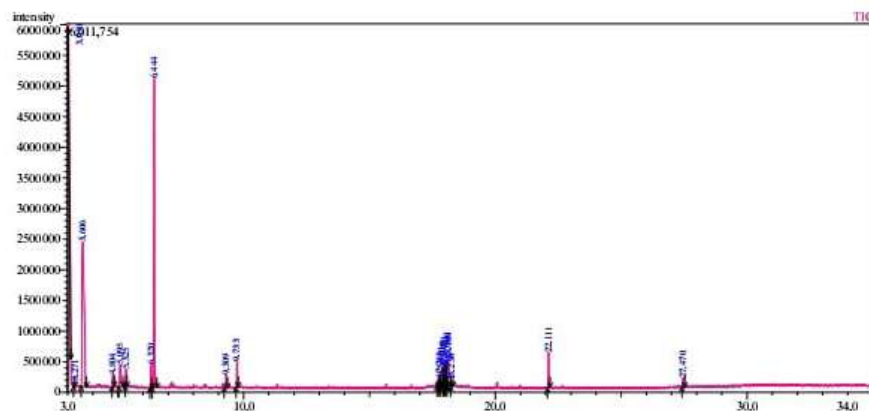


Fig 1: Gas Chromatography - Mass Spectrometry of HAECHL

Table 1: Identification of bioactive compounds in HAECHL by GC-MS

Peak	Rtime	Area	Area%	Height	Height %	Molecular formula	Molecular weight	Bioactive constituents
1	3.030	4020460	536666	536666	4.16	C ₅ H ₁₄ OSi	118	(Methoxymethyl)trimethyl silane
2	3.271	220240	0.53	97063	0.75	C ₅ H ₁₂ O	88	1-Butanol, 3-methyl
3	3.606	13991583	33.36	2356925	18.25	C ₇ H ₁₆ O ₂	132	Propane, 2,2-diethoxy
4	4.804	453560	1.08	195112	1.51	C ₇ H ₁₆ O	116	3-Hexanol,3-methyl
5	5.095	1220090	2.91	371180	2.87	C ₆ H ₁₀ O ₂	114	2-Butanoic acid, Ethyl ester
6	5.325	507430	1.21	277115	2.15	C ₇ H ₁₆ O ₂	132	Propane, 1,1-diethoxy
7	6.320	806758	1.92	5036243	39.01	C ₄ H ₅ NO ₂	99	Pyrolidine-2,4-dione
8	9.309	387016	0.92	209276	1.62	C ₉ H ₂₀ O ₃	176	Propane, 1,1,3-triethoxy-
9	17.755	567520	1.35	136715	1.06	C ₁₀ H ₁₆ O ₃	184	1,4-Butanediol, 1-(2-furyl)-2,3-dimethyl
10	17.820	59633	1.42	195164	1.51	C ₁₃ H ₂₂ O ₄	242	3-(Tetrahydropyran-2-yloxy)-9-oxa-bicyclo [3.3.1]nonan-2-ol
11	17.910	1358795	3.24	298695	2.31	C ₈ H ₁₈ O ₂	146	1,3-Octanediol
12	17.945	649244	1.55	318756	2.47	C ₈ H ₁₄ O ₄	174	Octanedioic acid
13	17.970	1481013	3.53	357197	2.77	C ₆ H ₈ ClF	134	6-Chloro-2-fluorohexa-1,3-diene
14	18.060	1254838	2.99	436020	3.38	C ₁₀ H ₂₀ O ₂	172	Octanoic acid, 4-methyl-, methyl ester
15	18.104	2485376	5.93	443576	3.44	C ₇ H ₁₄ O ₆	194	Mome Inositol
16	18.230	324815	0.77	97759	0.76	C ₇ H ₁₄ O ₆	194	3-O-Methyl-d-glucose
17	22.111	1002386	2.39	581068	4.50	C ₂₀ H ₄₀ O	296	2-Hexadecen-1-ol, 3,7,11,15-Tetramethyl-Hexadec-2-en-1-ol
18	27.470	415677	0.99	156175	1.21	C ₂₄ H ₃₈ O ₄	390	Bis(2-ethylhexyl)phthalate

The suite of eighteen compounds shows several pharmacological activities

1-Butanol, 3-methyl, 3-hexanol-3-methyl, 1,3-octanediol and caprylic acid serve both as mild antimicrobials and in topical settings, barrier-enhancers or penetration facilitators; 2,2-diethoxypropane, 1,1-diethoxypropane and ethyl 2-butanoate stabilize antiepileptic activity; the pyrrolidine-2,4-dione selectively inhibits T-type calcium currents, it has antiepileptic and neuroprotective effects; 1,4-Butanediol, 1-(2-furyl)-2,3-dimethyl, the 3-(Tetrahydropyran-2-yloxy)-9-oxa-bicyclo[3.3.1]nonan-2-ol, and the halogenated conjugated diene reported anticancer, anti inflammatory and ion-channel modulation activities; metabolic and signaling probes like myo-inositol and 3-O-methyl-D-glucose regulate insulin pathways and GLUT transport respectively; the 2-Hexadecen-1-ol, 3,7,11,15-Tetramethyl-Hexadec-2-en-1-ol exhibited antioxidant, anti inflammatory and mild sedative actions while enhancing photosynthesis in plants; and finally, bis(2-ethylhexyl) phthalate stands apart as a PPAR-activating environmental toxin linked to oxidative stress and metabolic dysregulation.

Table 2: Biological activity of phytocompounds identified in HAECHL by GC-MS

S.no	Bioactive compound	Derivatives	Biological activity
1	1-Butanol, 3-methyl (Isoamylalcohol)	Hydroxyl group	Antimicrobial, Antifungal, Antibacterial [14]
2	Propane, 2,2-diethoxy	2,2-Diethoxypropane	reducing neuronal excitability [15]
3	3-Hexanol,3-methyl	Tertiary alcohol	Antiepileptic activity [16]
4	2-Butanoic acid, Ethyl ester	α,β Saturated ester	reducing neuronal excitability [17]
5	Propane, 1,1-diethoxy	Acetal	Antiepileptic activity [18]
6	Pyrrolidine-2,4-dione	Succinamide	Neuroprotective potential, Anti microbial, antiinflammatory properties [19]
7	Propane, 1,1,3-triethoxy-	Tri-ethoxy-substituted propane derivative (a trialkoxyalkane)	Antibacterial and Antifungal [20]
8	1,4-Butanediol, 1-(2-furyl)-2,3-dimethyl	Furan derivative	Antibacterial, antifungal antioxidant, Anticancer [21]
9	3-(Tetrahydropyran-2-yloxy)-9-oxa-bicyclo[3.3.1]nonan-2-ol	bicyclic ether core and a tetrahydropyran (THP) protected alcohol	Antibacterial, Antifungal, Antioxidant, Anti-inflammatory[22]
10	1,3-Octanediol	Ester and ether	Antibacterial, antifungal ; used in skin care. [23]
11	Octanedioic acid	Caprylic acid (medium-chain dicarboxylic acid)	Antimicrobial and antiviral [24]
12	6-Chloro-2-fluorohexa-1,3-diene	conjugated diene	Antibacterial, antifungal and anticancer. [25]
13	Octanoic acid, 4-methyl-, methyl ester	Fattyacid ester	Antibacterial, Antifungal, Antioxidant; Potential influence on energy metabolism [26]
14	Mome Inositol (Myo-inositol)	Cyclohexanehexol scaffold	Roles in anxiety, depression, fertility (PCOS), metabolic disorders; Antioxidant, Anticancer, Insulin signal [27]
15	3-O-Methyl-d-glucose	Monosaccharide derivative	Brain metabolism, glucose transport; Used in tumor uptake and glucose transport inhibition studies. [28]
16	2-Hexadecen-1-ol, 3,7,11,15-Tetramethyl-Hexadec-2-en-1-ol	Diterpenoid alcohol	Antioxidant, Anti-inflammatory, Anticancer, Antibacterial, Antifungal; Larvicidal and pesticidal [29]

17	Bis(2-ethylhexyl) phthalate	Phthalate ester	Hepatotoxicity; Oxidative stress; Activates PPARs; Linked to obesity, insulin resistance, type 2 diabetes [30]
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CONCLUSION

The present research article provide information of bioactive constituents present in hydroalcoholic extract of *Calliandra haematocephala* (HAECH_L). It revealed seventeen bioactive compounds from the chromatogram. pharmacological use of phytochemicals may serve as a valuable source for the development of therapeutic agents, exhibiting a wide range of activities including antimicrobial, anti inflammatory, antioxidant, modulation of neural excitability, hepatoprotective, gastroprotective, antifungal, antibacterial, antiviral, antidiabetic, anticancer, and other related effects. Therefore current article highlights the essential phytoconstituents it may be used for pharmaceutical purpose.

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