



Phyto - Chemical Studies on the Leaves of Methanol Extract of *Hyptis suaveolens* L (Poit.) from Nagapattinam District, Tamil Nadu, India

KEYWORDS

Hyptis suaveolens L (Poit.), Methanol extract, GC-MS analysis Phenanthrenemethanol, n-hexadecanoic acid, Podocarp

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ABSTRACT *Hyptis suaveolens* L (Poit.) leaves of Methanol Extract obtained by Solvent extraction by maceration process were analyzed by gas chromatography mass spectroscopy (GC-MS). Thirty six components were identified in the leaf extract. The major components were Phenanthrenemethanol (44.74%), n-hexadecanoic acid (12.69%), Podocarp (9.92%), 7-isopropyl-1, 1, 4a-trimethyl-1, 2, 3, 4, 4a, 9, 10, 10a-octahydrophenanthrene (7.41%), Oleic acid (5.53%), 2-(1,4,4-Trimethyl-cyclohex-2-enyl)-ethanol (4.58%), (3,7-Dimethyl-octa-2,6-dienyl)-benzene (4.49%), Androsta-5,7-diene, 4,4-dimethyl (3.03%), phytol (1.52%), 3, 7, 11, 15-tetramethyl-2-hexadecen-o-1 (1.30%). The compositions of Methanol Extract varied qualitatively and quantitatively.

INTRODUCTION

Hyptis suaveolens L (Poit.) and commonly known as American mint belongs to the family Lamiaceae. It is a weed of waste places, plantation crops, forest margins, road sides and becoming abundant in fallow ground. It prefers a wet tropical climate, less common in regions with a seasonal wet/dry regime (Waterhouse and Mitchell, 1998). The genus *Hyptis* comprising more than 300 species, exhibits a major morphological diversity found in various tropical and subtropical regions of the world including Tamil Nadu. Most of them originate from tropical America. This specie is quite aromatic and is frequently used in treatments of gastrointestinal infections, cramps, and pain, as well as skin infections (Correa, 1931). The plant is used in the southern Sahara to treat asthma and malaria, cereals conservation (Adjanohoun et al., 1986) and to repel mosquitoes (Seyoum et al., 2002). The plant showed antibacterial and antifungal activities (Goun et al., 2003; Zollo et al., 1998). Despite those intensive works done to investigate the chemical composition of *Hyptis species* Methanol Extract all over the world. But there is no published report in the literature about the chemical composition of *Hyptis suaveolens* L (Poit.) Methanol Extract from Nagapattinam district of Tamil Nadu, India. So, an attempt has been taken to investigate the chemical components of Methanol Extract obtained from the leaves of *Hyptis suaveolens* L (Poit.) grown widely in Nagapattinam district of Tamil Nadu.

MATERIALS AND METHODS

PLANT MATERIAL

The fresh plant organ (Leaves) of *H. suaveolens*, (L), poit. Were collected from Nagapattinam (10.7906°N and 79.8428°E), district of Tamil Nadu, India. The voucher specimens were preserved in the Department of Botany, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

PREPARATION OF EXTRACT

Solvent extraction by maceration process

25gms of the powder of plant material was transferred into different conical flask (250ml). The conical flask containing 100ml of Methanol. The conical flask containing plant powder and solvent was shaken it well for 48 hours by me-

chanical shaker. The extracts were filtered using Whatmann filter paper No.1. The filtrates were evaporated to dryness using water bath. The obtained extracts were stored at 4°C in air tight bottle until further use.

GC-MS ANALYSIS

The Methanol Extract from leaves of *H. suaveolens* L, (Poit) were analyzed by GC-MS analysis was carried out on Shimadzu 2010 plus comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer instrument employing the following conditions: column RTX 5Ms (Column diameter is 0.32mm, column length is 30m, column thickness 0.50µm), operating in electron impact mode at 70eV; Helium gas (99.999%) was used as carrier gas at a constant flow of 1.73 ml /min and an injection volume of 0.5 µl was employed (split ratio of 10:1) injector temperature 270 °C; ion-source temperature 200 °C. The oven temperature was programmed from 40 °C (isothermal for 2 min), with an increase of 8 °C/min, to 150°C, then 8°C/min to 250°C, ending with a 20min isothermal at 280°C. Mass spectra were taken at 70eV; a scan interval of 0.5 seconds and fragments from 40 to 450 Da. Total GC running time is 51.25min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adopted to handle mass spectra and chromatograms was a TurboMass Ver 5.2.0. (Adams 2009)

IDENTIFICATION OF THE COMPOUNDS

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight and structure of the components of the test materials were ascertained.

RESULTS

Methanol Extract from the leaves of *H. suaveolens* L, (Poit) were analyzed by GC-MS. Tables 1, 2 and figure 1 reported the composition of the leaf extract of *H. suaveolens* L, (Poit). The major constituents of the leaves of methanol

extract were Phenanthrenemethanol (44.74%), n-hexadecanoic acid (12.69%), Podocarp (9.92%), 7-isopropyl-1, 1, 4a-trimethyl-1, 2, 3, 4, 4a, 9, 10, 10a-octahydrophenanthrene (7.41%), Oleic acid (5.53%), 2-(1,4,4-Trimethyl-cyclohex-2-enyl)-ethanol (4.58%), (3,7-Dimethyl-octa-2,6-dienyl)-benzene (4.49%), Androsta-5,7-diene, 4,4-dimethyl (3.03%), phytol (1.52%), 3, 7, 11, 15-tetramethyl-2-hexadecen-ol (1.30%).

DISCUSSION

The study reveals that composition of the extract differs from the earlier reports and may, therefore be treated as different chemotypes. On the basis of aforementioned fact, it may be concluded that *H. suaveolens*, growing widely in Nagapattinam in Tamil Nadu, may be utilized as a source for the isolation of natural Phenanthrenemethanol (44.74%), n-hexadecanoic acid (12.69%), Podocarp (9.92%), 7-isopropyl-1, 1, 4a-trimethyl-1, 2, 3, 4, 4a, 9, 10, 10a-octahydrophenanthrene (7.41%), Oleic acid (5.53%), 2-(1,4,4-Trimethyl-cyclohex-2-enyl)-ethanol (4.58%), (3,7-Dimethyl-octa-2,6-dienyl)-benzene (4.49%), Androsta-5,7-diene, 4,4-dimethyl (3.03%), phytol (1.52%), 3, 7, 11, 15-tetramethyl-2-hexadecen-ol (1.30%) respectively. The high concentration of n-hexadecanoic acid (12.69%) and phytol (1.52%) in leaf extract make it potentially useful in

the medicines because they exhibit antioxidant and anti-cancer activities (Dr. Duke online database). However, further study has to be conducted for its confirmation. It is worth noting that the methanol extract of *H. suaveolens* L, (Poit) has been reported to be used in folk medicine in the treatment of asthma and malaria, cereals conservation and to repel, larvicidal and adulticidal activities of mosquitoes.

Figure 1: Chromatogram obtained by the GC/MS with the methanol extract of *Hyptis suaveolens* from Nagapattinam District of Tamil Nadu.

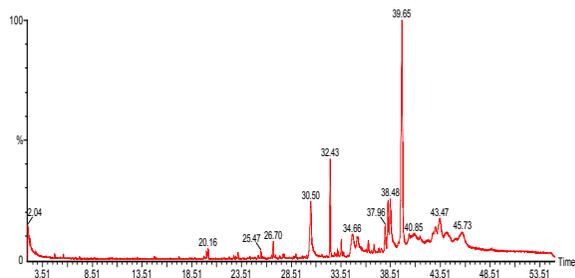


Table 1: Shows the components identified by GC/MS in methanol extract of *Hyptis suaveolens* from Nagapattinam District of Tamil Nadu.

S.No.	Peak Name	Retention time	Peak area	%Peak area	MolecularFormula	Molecular Weight
1	1,3-Cyclohexadiene, 1-methyl-4-(1-methylethyl)-	7.23	2324825	0.0731	C ₁₀ H ₁₆	136
2	1,3-Cyclohexadiene, 1-methyl-4-(1-methylethyl)-	7.37	2324825	0.0258	C ₁₀ H ₁₆	136
3	Ethanone, 1-(1,3-dimethyl-3-cyclohexen-1-yl)-	8.69	1557035	0.0212	C ₁₀ H ₁₆ O	152
4	cis-Hept-4-enol	13.28	31319364	0.0214	C ₇ H ₁₄ O	114
5	E-14-Hexadecenal	14.75	1501346	0.1368	C ₁₆ H ₃₀ O	238
6	Caryophyllene	15.62	2422736	0.0224	C ₁₅ H ₂₄	204
7	Eicosane	17.22	2859936	0.1181	C ₂₀ H ₄₂	282
8	1-Penten-3-one, 1-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	18.34	1549830	0.1252	C ₁₄ H ₂₂ O	206
9	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-	18.82	2324825	0.1265	C ₁₁ H ₁₆ O ₂	180
10	Dodecanoic acid	19.75	2324825	0.2736	C ₁₂ H ₂₄ O ₂	200
11	2-Methyl-6-methylene-octa-1,7-dien-3-ol	21.73	1791654	0.1190	C ₁₀ H ₁₈ O	152
12	Ethanone, 1-(octahydro-7a-methyl-1H-inden-1-yl)-, (1à,3aà,7aà)-	22.25	1328335	0.2089	C ₁₂ H ₂₀ O	180
13	1-Cycloheptene, 1,4-dimethyl-3-(2-methyl-1-propene-1-yl)-4vinyl-	22.72	2485758	0.2368	C ₁₅ H ₂₄	204
14	Pentadecane, 2,6,10-trimethyl-	22.96	964935	0.1725	C ₁₈ H ₃₈	254
15	1-Cyclohexanone, 2-methyl-2-(3-methyl-2-oxobutyl)-	24.52	2196184	0.0358	C ₁₂ H ₂₀ O ₂	196
16	15-Eicosene, (E)-	25.47	3127642	0.3977	C ₂₀ H ₄₀	280
17	Hexadecane	22.62	340007	0.0614	C ₁₆ H ₃₄	226
18	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	26.70	1680942	1.3023	C ₁₈ H ₃₆	254
19	2-Pentadecanone, 6,10,14-trimethyl-	26.93	23972436	0.2295	C ₁₉ H ₃₈ O	270
20	Hexadecane, 2,6,11,15-tetramethyl-	28.73	340007	0.1127	C ₁₈ H ₃₈	254
21	Hexadecanoic acid, methyl ester_	28.97	1680942	0.2344	C ₁₇ H ₃₄ O ₂	270
22	Resibufogenin	29.63	1936521	0.1012	C ₁₈ H ₃₆ O	268
23	Heptadecane	29.78	1936521	0.1037	C ₁₇ H ₃₆	240
24	n-Hexadecanoic acid	30.50	20711342	12.6965	C ₁₆ H ₃₂ O ₂	256
25	7-Isopropyl-1,1,4a-trimethyl-1,2,3,4,4a,9,10,10a-ctahydrophenanthrene	32.43	33090166	7.4170	C ₂₀ H ₃₀	270
26	1,3,6,10-Cyclotetradecatetraene, 3,7,11-trimethyl-14-(1-methylethyl)-, [S-(E,Z,E,E)]-	33.02	2583766	0.1305	C ₂₀ H ₃₂	272
27	Phytol	33.54	14259571	1.5214	C ₂₁ H ₄₂	296
28	Octadecanoic acid, methyl ester	33.78	4181217	0.2853	C ₁₉ H ₃₈ O ₂	298
29	Oleic Acid	34.66	42458432	5.5358	C ₁₈ H ₃₄ O ₂	286

30	1-Phenanthrenemethanol, 1,2,3,4,4a,9,10,10a-octahydro-1,4a-dimethyl-7-(1-methylethyl)-, [1S-(1à,4aà,10aà)]- or Androst-5,7-dien-3-ol-17-one	36.84	8478155	0.9188	C ₁₉ H ₂₆ O ₂	286
31	Androsta-5,7-diene, 4,4-dimethyl-	37.96	2843629	3.0307	C ₁₉ H ₂₆	284
32	(3,7-Dimethyl-octa-2,6-dienyl)-benzene	38.25	1848176	4.4999	C ₁₃ H ₁₈	214
33	Podocarp-7-en-3á-ol, 13á-methyl-13-vinyl-	38.48	45133100	9.9231	C ₂₀ H ₃₂ O	288
34	1-Phenanthrenemethanol, 1,2,3,4,4a,9,10,10a-octahydro-1,4a-dimethyl-7-(1-methylethyl)-, [1S-(1à,4aà,10aà)]-	39.65	136137648	44.7408	C ₂₀ H ₃₀ O	286
35	Palustric acid	40.85	41869200	0.4575	C ₁₁ H ₁₈ O	302
36	2-(1,4,4-Trimethyl-cyclohex-2-enyl)-ethanol	43.47	42889338	4.5850	C ₁₁ H ₂₀ O	168

Table 2: Activity of phyto-components identified in Hyptis suaveolens by GC/MS from Nagapattinam District of Tamil Nadu.

S.No	R/T	Name of the Compound	Activity**
1	33.54	Phytol	Antimicrobial, anticancer, Cancer- Preventive, diuretic antiinflammatory
2	15.62	Caryophyllene	Anti-tumor, Analgesic Antibacterial, Anti-inflammatory, Sedative, Fungicid.
3	19.75	Dodecanoic acid	Antioxdant, antibacterial, COX-1 & COX-2 inhibitor, antiviral, hypocholerolemic, candidicide.
4	28.97	Hexadecanoic acid, methyl ester	Antioxidant, Hypocholesterolemic, Antiandrogenic, Hemolytic, Alpha Reducatase inhibitor.
5	30.50	n-Hexadecanoic acid	Antioxidant, Hypocholesterolemic nematicide, pesticide, Antiandrogenic flavor, hemalytic, 5- Alpha reductase inhibitor

****Source: Dr.Duke's phytochemical and ethnobotanical databases [Online database].**

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