

ORIGINAL ARTICLE

Spectral Analyses of the Bioactive Compounds Present in the Ethanolic Leaf Extract of *Strobilanthes kunthiana* (Nees) T. Anderson ex. Benth

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ABSTRACT

The present study was carried out to determine the bioactive compounds present in the ethanol extracts of *Strobilanthes kunthiana* (Neela kurinji) leaves. Fourier-transform infrared spectroscopy (FT-IR) and Gas chromatography mass spectrometry (GC-MS) were used for analysis. FT-IR analysis of the plant extract showed the presence of alcohols and phenols, alkanes, aliphatic amines, aromatics and alkyl halides at, 3400.5, 2937.59, 1056.99, 1431.18, and 1265.3cm⁻¹ respectively. There were strong bands at 3400.5, 2937.59 and 1056.99cm⁻¹ due to O-H, C-H and C-N groups respectively. GC-MS analysis revealed the presence of 16 chemical compounds representing about 83.30% of the total ethanol extract. The major compounds were, Azulene (14.74%), 2, 6, 10-Trimethyl, 14-Ethylene-14-Pentadecene (17.34%), 2,Hexaceden-1-ol,3,7,11,15-Tetramethyl (8.11%), 1-Heptacosanol (18.35%), 2,6,10,14,18,22-Tetracosahexaene,2,6 (11.33%). From the results obtained it is evident that *Strobilanthes kunthiana* leaves contains various functional groups and phytochemicals which can further be exploited for phytopharmaceuticals.

Keywords: *Strobilanthes kunthiana* leaves, ethanol extract, GC-MS, and FT-IR.

Received 02/03/2015 Accepted 16/04/2015

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How to cite this article:

Isoe M E, Alagar Y S, Deivamarudachalam T P D. Spectral Analyses of the Bioactive Compounds Present in the Ethanolic Leaf Extract of *Strobilanthes kunthiana* (Nees) T. Anderson ex. Benth. Adv. Biores., Vol 6 [3] May 2015: 65-71. DOI: 10.15515/abr.0976-4585.6.3.6571

INTRODUCTION

In recent years, secondary plant metabolites, previously with unknown pharmacological activities, have been extensively investigated as a source of medicinal agents [1]. The plant *Strobilanthes kunthiana* locally known as *Neela kurinji* belongs to the family Acanthaceae. The plant extensively grows in Tamil Nadu and Kerala states of India mostly for ornamental purpose. It has been used for the treatment of inflammation. It has anti-osteoarthritic properties [2], Analgesic activities [3], Antigiardial activity, central nervous depressant activity, antioxidant and anti-cancer properties [4]. Fourier Transform Infrared Spectroscopy (FT-IR) is the latest accurate technique that offers quantitative and qualitative analysis for organic and inorganic samples. Recently, gas chromatography mass spectrometry has become firmly established as a key technological platform for secondary metabolite profiling in both plant and non-plant species [5]. Owing to the detailed literature survey that was conducted, it was found that so far there is no available literature regarding the functional groups as well as the phytochemicals present in this plant. Therefore, the objective of the present study was to determine the functional groups as well as the phytochemical components of the ethanol extract of the *Strobilanthes kunthiana* leaves.

MATERIALS AND METHODS

Collection of plant material

The leaves of *Strobilanthes Kunthiana* were collected from their natural habitat in Western Ghats Ooty, Tamil Nadu, India, during the month of August 2014 and identified by Dr. S. Rajan field Botanist, the Survey of medicinal plants and collection unit, Government of India, Nilgiri.

Preparation of plant extraction

The leaves were washed thoroughly in tap water, shade dried at room temperature for 7 days and powdered using a grinder. A portion of (40grams) of powdered sample was dissolved in 400 ml of ethanol (1:10) and it was subjected to cold maceration for 48hours then kept on a rotary shaker at 190–220 rpm for 24 h. The extract was centrifuged (3000 X g) and a clear supernatant was collected and filtered using Whatman No. 1 filter paper and was evaporated dryness at 40°C by rotary evaporator (Buchi type, Flawil/Schweiz, Switzeland).⁶ The resulting crude ethanol extract (4g) was stored at 4 °C in airtight bottle for the analyses.

Fourier transform infrared (FT- IR) analysis

FT-IR analysis was used to determine the functional groups in the ethanol extract and it was performed on SHIMADZU, Tokyo Japan, Fourier Transform Infrared spectrometer 800series, between 4000-400 cm⁻¹

Gas chromatography mass spectrometry analysis

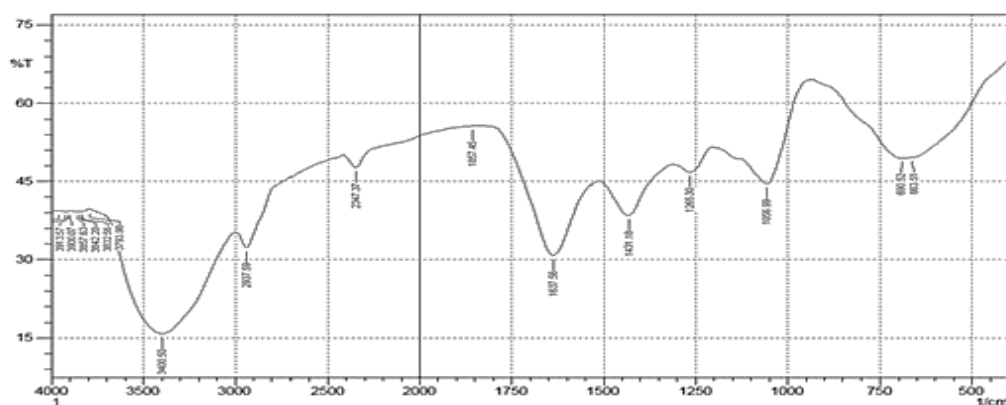
The Phytochemical investigation of ethanol extract was performed on a GC-MS equipment (Thermo Scientific Co.) Thermo GC-TRACE ultra ver.: 5.0, Thermo MS DSQ II. Experimental conditions of GC-MS system were as follows: TR 5-MS capillary standard non-polar column, dimension: 30Mts, ID: 0.25mm, Film thickness: 0.25µm. Flow rate of mobile phase (carrier gas: He- 99.99%) was set at 1.0 ml/min. In the gas chromatography part, temperature programme (oven temperature) was 50° raised to 260°C at 5°C/min and injection volume was 1µl. Samples dissolved in ethanol were run fully at a range of 40-600 m/z and the total running time was 38 minutes. The unknown compounds were identified by comparison of their retention indices with known compound spectrum in WILEY8 and NISTO8 (National Institute Standard and Technology) libraries for interpretation of the spectrum. This ascertained the molecular weight, peak area as well as the structure of the compounds.

RESULTS

Fourier transforms infrared spectroscopy

The result of the FT-IR peak values, chemical bonds and functional groups are represented in (Table 1). The FT-IR spectrum profile is illustrated in (Figure1). The FT-IR analysis of the plant extract showed the presence of alcohols and phenols, alkanes, aliphatic amines, aromatics and alkyl halides at, 3400.5, 2937.59, 1056.99, 1431.18, and 1265.3cm⁻¹ respectively.

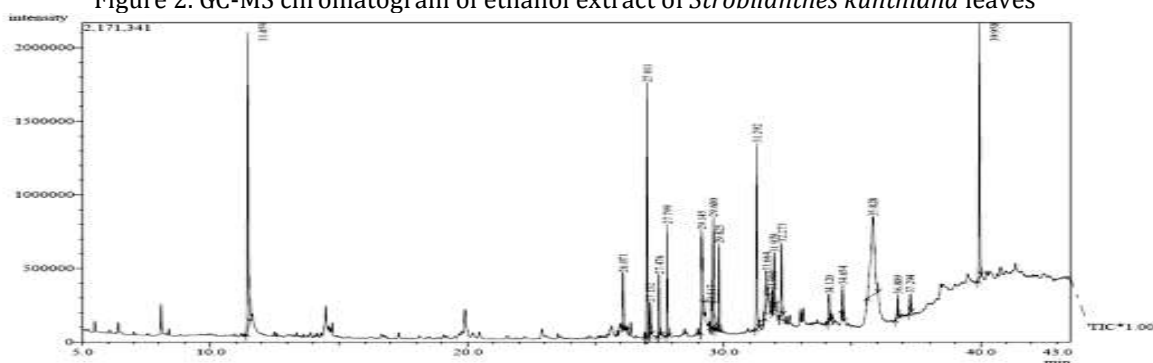
Figure 1. The FT-IR spectrum of the ethanol extract of *Strobilanthes kunthiana*



Gas chromatography mass spectrometry

Sixteen chemical compounds were identified through mass spectrometry attached with gas chromatography (GC) as illustrated in figure 2. The various compounds present in the crude ethanol extract of the leaves of *Strobilanthes kunthiana* that were detected by GC-MS are shown in (Table 2 and figure 2). They included; Azulene, n-Nonadecanol-1, 2,6,10-Trimethyl,14-ethylene-1, 2-Pentadecanone, n-Hexadecanoic acid, 2-Hexadecene, 3,7,11,15-tetramethyl-, [4-(3,5-Di-tert-butyl-4-hydroxyphenyl)]b, 2-Hexadecen-1-OL, 3,7,11,15-Tet, 8,11,14-docosatrienoic acid, methyl (9z,12z)-9,12-octadecad, (R)-(-)-14-Methyl-8-hexadecyn-1-ol, 1-Heptacosanol, Cyclohexane, 2-(1-decylunde, 1-Dodecanol, n-Pentadecanol, 2,6,10, 14, 18, 22-Tetracosahexaene. There were five prominent phytocomponents (Figure 3 to figure 7).

Figure 2. GC-MS chromatogram of ethanol extract of *Strobilanthes kunthiana* leaves



The major compounds represented are shown below (Figure 3 to figure 7) with their two dimensional structures;

Figure 3. Azulene (RT: 11.459)

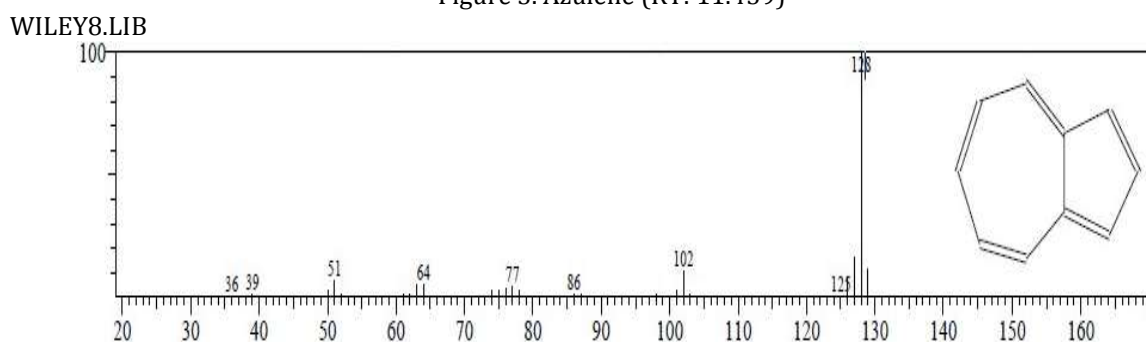


Figure 4. 2, 6, 10-Trimethyl, 14-Ethylene (RT: 27.011)

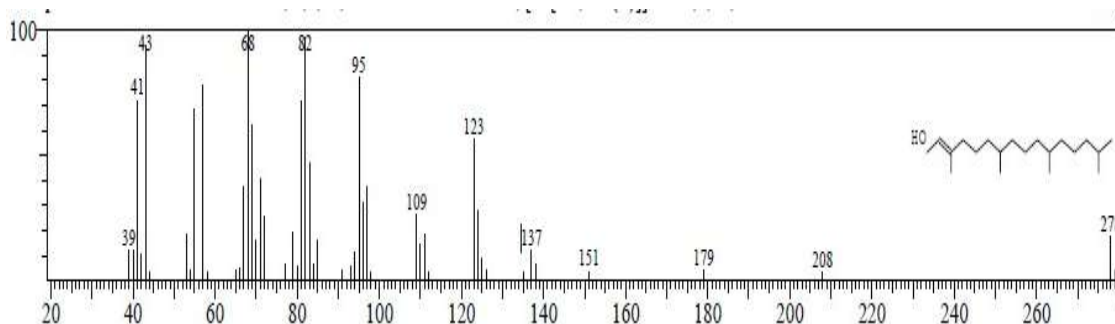
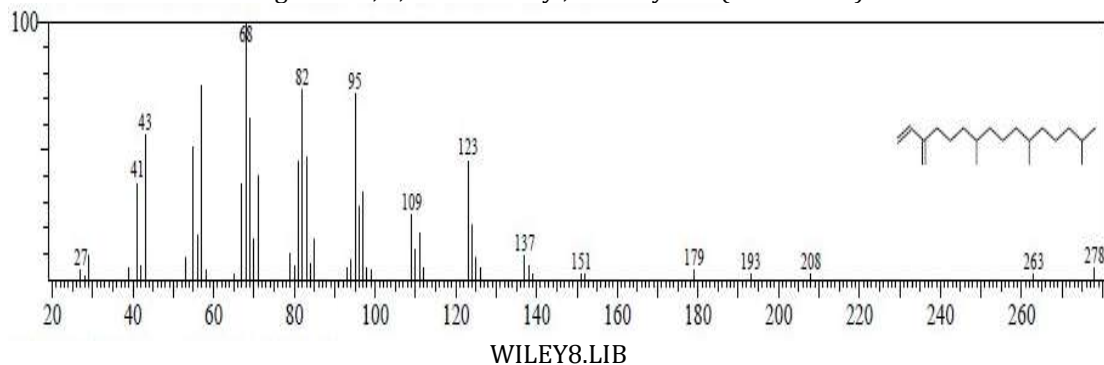


Figure 5. 2-Hexadecen-1-ol, 3, 7, 11, 15-Tetramethyl-, [r-[r*, r*-(e)] (RT: 31.292) WILEY8.LIB

Figure 6. 1-Heptacosanol (RT: 34.654)

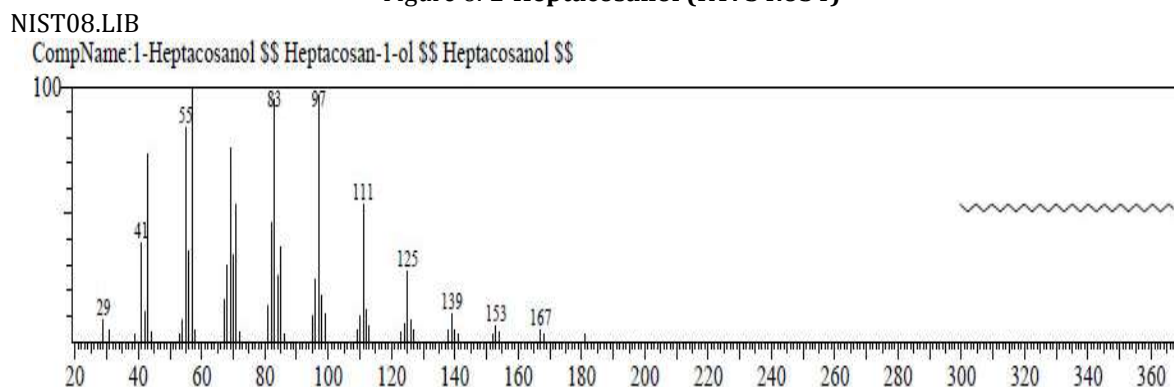
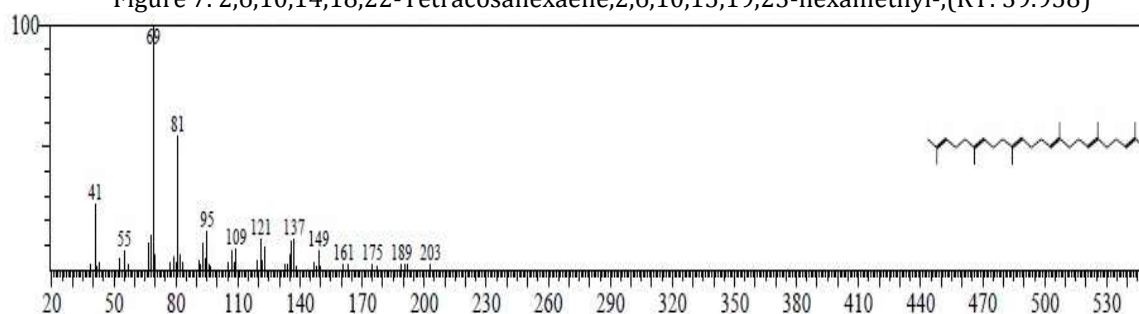


Figure 7. 2,6,10,14,18,22-Tetracosahexaene,2,6,10,15,19,23-hexamethyl-,(RT: 39.958)



NIST08.LIB

DISCUSSION

The present study is the first report on the GC-MS analysis as well as the FT-IR analysis in *Strobilanthes kunthiana* species. The FT-IR analysis of the plant extract showed the presence of alcohols and phenols, alkanes, aliphatic amines, aromatics and alkyl halides at, 3400.5, 2937.59, 1056.99, 1431.18, and 1265.3 cm^{-1} respectively. There were strong bands at 3400.5, 2937.59 and 1056.99 cm^{-1} due to OH, CH and C-N groups respectively. GC-MS analysis revealed the presence of different phytochemical constituents in the ethanol extract of *Strobilanthes kunthiana* leaves. A total of 16 compounds were identified representing about 83.30% of the total ethanol extract. The nature of the compounds represented, aromatic hydrocarbons, straight chain primary alcohols, Fatty acids, alkenes, phenolic compounds, titerpene alcohols and triterpenes, primary aliphatic alcohols and trienoic fatty acids. The major compounds were, Azulene (14.74%), 2, 6, 10-Trimethyl, 14-Ethylene-14-Pentadecne (17.34%), 2,Hexaceden-1-ol,3,7,11,15-Tetramethyl also known as phytol (8.11%), 1-Heptacosanol (18.35%), 2,6,10,14,18,22-Tetracosahexaene,2,6 also known as squalene (11.33%). Rahman reported antimicrobial components from the ethanolic extract of *Psidium guajava* of which the major components included Squalene (11.63%), Phytol (10.30%), and Azulene (5.19%).¹⁵ Comparatively our study reveals higher percentage of azulene and almost same percentage of phytol and squalene in addition to this *Strobilanthes kunthiana* contains an even higher percentage of an additional antimicrobial component (1-Heptacosanol 18.35%). Besides the antimicrobial activity of these compounds, they also possess antioxidant activities, antiseptic properties and anticancer properties. This implies the phytopharmaceuticals importance of the plant *Strobilanthes kunthiana*.

Table1. Fourier Transform Infrared spectral peak values and functional groups obtained from the ethanol extract of *Strobilanthes kunthiana* leaves

Peak Values in cm^{-1}	Chemical Bonds	Functional groups
663.51	C-CL	Alkyl halides
690.25	C-CL	Alkyl halides
1056.99	C-N Stretch	Aliphatic amines
1265.3	C-H Wag (-CH ₂)	Alkyl halides
1431.18	C-C Stretch (n ring)	Aromatics

1637.56	N-H Bend	Primary amines
1857.45	-	Unknown
2347.37	-	Unknown
2937.59	C-H Stretch	Alkanes
3400.5	O-H Stretch	Alcohol phenols

Table 2. Phytochemical compounds identified in the ethanol extract of *Strobilanthes kunthiana* leaves by GC-MS

RT	COMPOUND NAME	MOLECULAR FORMULA	MOL. Wt.	PEAK AREA%	COMPOUND NATURE	ACTIVITY
11.459	Azulene	C10H8	128	14.74	Polycyclic aromatic hydrocarbon	ANTIOXIDANT, REDUCE INFLAMMATION IN THE SKIN TISSUE, ANTIFUNGAL, ANTIBACTERIAL AND ANTISEPTIC. ⁷
26.071	n-Nonadecanol-1	C19H40O	284	8.31	Straight-chain primary alcohols (1°)	FLAVOR AND FRAGRANCE AGENT, ANTIMICROBIAL AND CYTOTOXICITY. ⁸
27.011	2,6,10-Trimethyl, 14-Ethylene-14-Pentadecne (Neophytadiene)	C18H36O	278	10.58		ENZYME INHIBITOR
27.132	2-Pentadecanone,6,10,14-Trimethyl (Hexahydrofarnesyl acetone) hexahydrofarnesyl acetone	C18H36O	268	1.59		
29.145	n-Hexadecanoic acid	C16H32O2	256	5.28	Fatty acid	ARTIFICIAL FLAVORS AND ANTI-INFLAMMATORY. ⁹ ANTIOXIDANT, HYPOCHOLESTEROLEMIC5-ALPHA REDUCTASE INHIBITOR. ¹⁰
29.517	2-Hexadecene,3,7,11,15-Tetramethyl-,	C20H40	280	1.25	Alkene	ANTIBACTERIAL ACTIVITY (ISMAEL 2011)
29.825	4-(3,5-Di-Tert-Butyl-4-Hydroxyphenyl	C21H32O3	332	3.43	Phenolic compound	ANTIOXIDANT
31.292	2,Hexadecan-1-ol,3,7,11,15-Tetramethyl (phytol)	C20H40O	296	8.11	Diterpene alcohol	COSMETICS, SHAMPOOS, (ANTIMICROBIAL) TOILET SOAPS, HOUSEHOLD CLEANERS, AND DETERGENTS. ¹¹
31.664	8,11,14-Docosatrienoicacid,Methyl ester	C32H40O2	348	3.02	-	-
31.900	Ethyl(9z,12z)-9,12-Octadecadienoate	C20H36O2	308	0.80	trienoic fatty acid	-

31.979	(R)-(-)-14-Methyl-8-hexadecyn-1-ol	C17H32O	252	2.65	-	-
35.828	1-Heptacosanol	C27H56O	396	14.24	Straight chain primary alcohol	FLAVOR AND FRAGRANCE AGENT, LOWER CHOLESTEROL, ANTIMICROBIAL AND CYTOTOXICITY. ¹²
34.120	Cyclohexane,2-(-1-Decylunde	C29H58	406	1.72	-	-
36.809	1-Dodecanol	C12H26O			Fatty alcohol	USED IN DETERGENT INDUSTRY, EMOLLIENT, EMULSIFIER, FRAGRANCE, FLAVOR AND ANTIBACTERIAL ACTIVITY. ¹³
37.294	n-Pentadecanol	C15H32O	228	0.78	Primary aliphatic alcohol	USED FOR SKIN
39.958	2,6,10,14,18,22-Tetracosahexaene,2,6 (Squalene)	C30H50	410	11.33	Triterpene	ANTIMICROBIAL. SYNTHESIZE CHOLESTEROL STEROID HORMONES AND VITAMIN D, ANTICANCER AND PROTECTS THE SKIN AGAINST UV. ¹⁴

Note: Table 2. Contains retention time, molecular formula, molecular weight and peak area percentage as obtained from the results. Compound nature and activity were ascertained from a detailed literature survey.

CONCLUSION

The present study reveals that *Strobilanthes kunthiana* is a rich reservoir of medicinally useful phytoconstituents. Further investigation is being carried out to ascertain the pharmacological activities of the concerned compounds individually which will be useful for further drug development.

CONFLICT OF INTEREST

None declared.

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