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Short Communication

GC-MS analysis of ethanolic extract of *Canthium parviflorum* Lamk Leaf

Purushoth Prabhu.T[1], Panneerselvam.P[2], Suresh.R[3], Clement Atlee.W[4], Balasubramanian.S[1]
[1]Dept of pharmacognosy, C.L.Baid Metha College of Pharmacy, Thoraipakkam, Chennai.
[2]Dept of pharmaceutical chemistry, C.L.Baid Metha College of Pharmacy, Thoraipakkam, Chennai.
[3]Dept of pharmaceutical chemistry, Jadavpur University, Calcutta
[4]Dept of pharmacology, C.L.Baid Metha College of Pharmacy, Thoraipakkam, Chennai.

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ABSTRACT

Canthium parviflorum Lamk(Rubiaceae) is a shrubby and woody plant found throughout the Western Ghats. *Canthium parviflorum* have reported to possess a number of pharmacological activities such as antioxidant, wound healing activity and antitumor acitivity. D-mannitol, phenolic acid, phenolic compounds, carbohydrates, proteins were found from *Canthium parviflorum.* The current study was carried out to analyze the active constituents present in the leaf of *Canthium parviflorum.* Twenty two constituents in ethanolic extracts were identified by GC-MS analysis. D-Mannitol and Squalene which was present in this plant considered to have anticancer activity.

INTRODUCTION

A knowledge of the chemical constituents of plants is desirable not only for the discovery of therapeutic agents, but also because such information may be of great value in disclosing new sources of economic phytocompounds for the synthesis of complex chemical substances and for discovering the actual significance of folkloric remedies.

Hence a thorough validation of the herbal drugs has emerged as a new branch of science emphasizing and prioritizing the standardization of the natural drugs and products because several of the phytochemicals have complementary and overlapping mechanism of action. Mass spectrometry, coupled with chromatographic separations such as Gas chromatography (GC/MS) is normally used for direct analysis of components existing in traditional medicines and medicinal plants. In recent years GC-MS studies have been increasingly applied for the analysis of chinese medicinal plants as this technique has proved to be a valuable method for the analysis of non polar components and volatile essential oil, fatty acids and lipids Khare C.P (2007). *Canthium parviflorum* Lamk(Rubiaceae) is a shrubby and woody plant found throughout the Western Ghats. Members of Rubiaceae are distributed more widely all pans of the earth. All the genus of the family are economically important. *Canthium* is a genus of about 230 species of shrubs or small trees. Plant pacifies vitiated kapha, diarrhea, fever, leucorrhea, worm infestation and general debility. In siddha system of medicine the plant was used in respiratory disorder, diuretic, diabetic, obesity. In Ayurvedha system of medicine the plant was used in cough, diuretic, tumor and as anthelmintic Anonyms, 1992. An antioxidant, wound healing activity and antitumor acitivity were reported. D-mannitol, phenolic acid, phenolic compounds, carbohydrates, proteins were found from *Canthium parviflorum.* Sathish kumar et al (2008). Pharmacological activities such as antimicrobial, antioxidant, antidiabetic, wound healing, diuretic, anti-inflammatory, antinociceptive, antitumor and antipyretic from various species of *Canthium* has been reported. Elayaraj et al (2007).

* *Corresponding Author*
Purushoth Prabhu.T,
Assitant Professor, Dept of pharmacognosy,
C.L.Baid Metha College of Pharmacy, Thoraipakkam, Chennai, India

MATERIALS AND METHODS

Collection of plant materials

The leaf *Canthium parviflorum* was collected from surroundings of Tambaram, Chennai. They were identified and authenticated by Prof Jayaraman, PARC , Tambaram, Chennai, Tamilnadu, India.

Sample Preparation

The leaf *Canthium parviflorum* were shade dried and pulverized well. About 20g of the powdered leaves were soaked in 100 ml of ethanol. It was left for 24 hours so that terpenoids, and other constituents if present will get dissolved. The ethanolic extract was filtered using Whatmann (number 1) filter paper and the residue was removed.

Gas Chromatography—Mass Spectroscopy

The ethanolic extract was subjected to GC-MS analysis on the instrument GC-MS SHIMADZU QP2010 with Elite – DB-5M column and the GC-MS solution version 2.53 software. Initially oven temperature was maintained at 70^{o}C for 2.0 minutes, and the temperature was gradually increased up to 300^{o}C at 10.0/35.0 min and 4.0 µl of sample was injected for analysis. Helium gas 99.995% of purity was used as a carrier gas as well as a eluent.

The flow rate of helium gas was set to 1.5 ml/min. The sample injector temperature was maintained at 260º C and the split ratio is 20 throughout the experiment periods. The ionization mass spectroscopic analysis was done with 70 eV. The mass spectra were recorded for the mass range 40-1000 *m/z* for about 35 minutes. Identification of components was based on comparison of their mass spectra. As the compounds separated, on elution through the column, were detected in electronic signals. As individual compounds eluted from the Gas chromatographic column, they entered the electron ionization detector where they were bombarded with a stream of electrons causing them to break apart into fragments. The fragments were actually charged ions with a certain mass. The m/z ratio obtained was calibrated from the graph obtained which was called as the mass spectrum graph which is the fingerprint of the molecule. The identification of compounds was based on the comparisons of their mass spectra with NIST Library 2008 WILEY8, FAME (Sriranmsridharan, 2011).

RESULTS AND DISCUSSION

GC-MS Analysis

GC-MS analysis was carried out on the ethanolic extract of *Canthium parviflorum* and 22 compounds were identified. The GC-MS analysis was done using the instrument GC-MS SHIMADZU QP2010 with GCMS solution version 2.53 software. The sample volume was 4.0 µL. The sample of ethanolic extract was run for 35 minutes. The Chromatogram (Figure.1) shows 6 prominent peaks in the retention time range 18.080 - 27.910. The peak at 18.080 retention time is having the peak area 49.82. This largest peak is due to the presence of Phytol. The Second less prominent peak at 27.910 retention time has the peak area 7.21 is due to the presence of Gamma- stigmasterol. The third less significant peak at 23.703 retention time with the peak area 6.32 is characteristic of All trans squalene. The Fourth less prominent peak at 26.008 retention time with the peak area 4.58 denotes DEPH; 1,2-Benzenedicarboxylicacid, bis (2-hylhexyl)ester. Ganesh S.et al (2011). The other less prominent peaks at other retention times are given in Table 1. The total ion chromatograph (TIC) showing the peak identities of the compounds identified have been given in Figure 1.

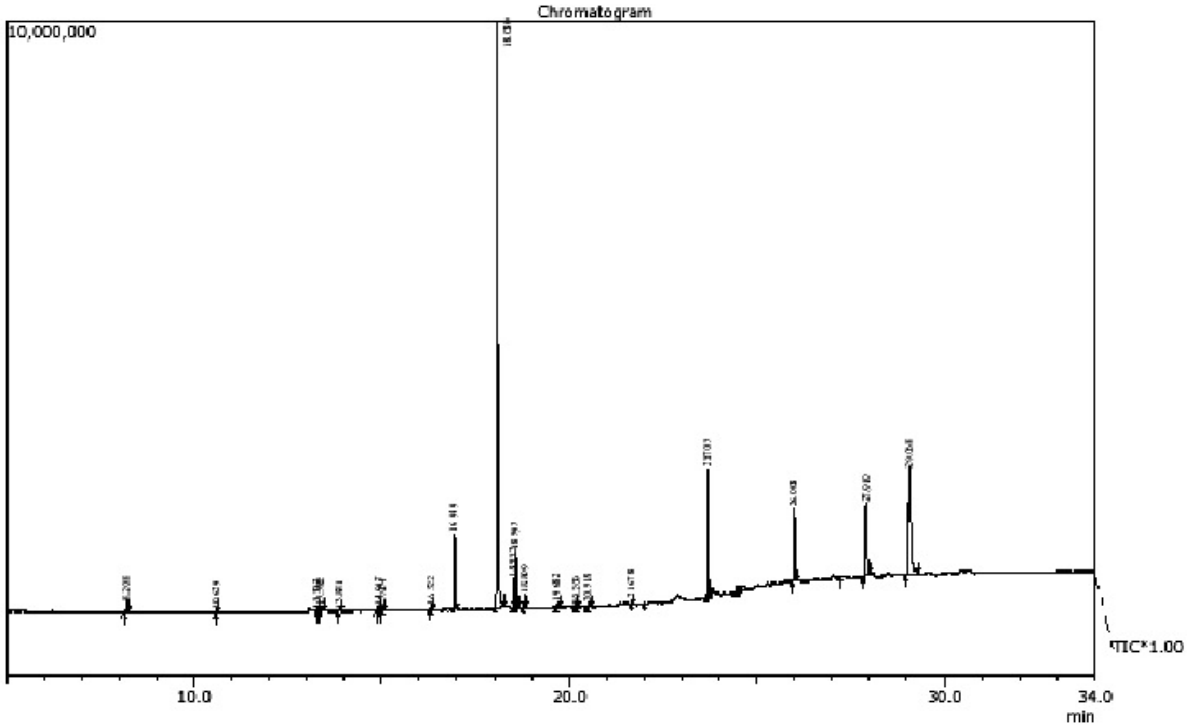

Fig. 1: GC-MS analysis of *Canthium parviflorum* leaf.

Table. 1: Phytoconstituents present in ECP.

S.No	RT (min)	Peak Area (%)	Name of the Phytoconstituents
1.	8.208	0.57	Biphenyl
2.	10.209	0.03	2-Methyl-4-heptanone
3.	12.716	0.07	Di-Isodectyl Phthalate
4.	13.302	0.06	1,2,4,5-Tetroxane,3,3,6.6-Tetraphenyl-
5.	13.382	0.37	3-Oxo-Alapha,-Ionol
6.	14.917	0.33	Methyl 7-hydroxy-2-methyl-3,5-octadienoate
7.	16.322	0.25	4-(2-Hydroxy-2,6,6-Trimethylcyclohexyl)-3-buten-2-one
8.	16.674	0.76	n-Hexadecanoic acid
9.	16.878	0.13	E-11- Hexadecanoic acid,Ethylester
10.	16.949	3.18	EthylHexadecanoate
11.	18.080	49.82	Phytol
12.	18.532	1.21	Ethyl(9Z,12Z)-9,12- Octadecanoate
13.	18.593	2.74	Ethyl Linolenate
14.	19.682	0.32	1-Hexadecanol
15.	22.043	1.65	2-Phenoxyl-2-phenylpropanic acid
16.	23.703	6.32	All-trans-squalene
17.	24.455	0.33	Methyl Linolenate
18.	25.416	1.48	Gamma-Tocopherol
19.	26.008	4.58	DEPH;1,2-Benzenedicarboxylicacid,bis (2-hylhexyl)ester
20.	27.260	0.68	Stigmasterol
21.	27.910	7.21	Gamma-stigmasterol
22.	29.068	0.38	Methyl cis-11,14,17-Icosatrienoate

CONCLUSION

The result of the present investigation reveals that the alcoholic extracts of *Canthium parviflorum* possessed significant anticancer activity which was analyzed by GC-MS analysis. Mannitol and Squalene which was present in *Canthium parviflorum* may be responsible for anticancer activity. The GC-MS analysis of the ethanolic extract of *Canthium parviflorum* reveals the presence of phytoconstituents belonging to the type-acids, esters, alcohols, ethers, *etc*. Thus, the medicinal plant *Canthium parviflorum* is found to possess significant phytoconstituents. The presence of such a variety of phytochemicals may be attributed to the medicinal characteristics of this plant *Canthium parviflorum.*

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