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### GC-MS analysis of biologically active compounds in *Canthium parviflorum* Lam. leaf and callus extracts

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**Abstract :** *Canthium parviflorum* is an important medicinal plant widely used in traditional systems of medicine. In the present work, we reported the FTIR and GC-MS analysis by using in vitro grown callus on MS medium supplemented with BA (0.1 to 0.5 mg/l) and NAA (0.5 and 1.0 mg/l) used with leaf explants compared with wild leaf explants. After 2 to 3 subcultures callus was collected and dried at normal temperature. To explore the phyto profile of callus extracts of *Canthium parviflorum*, fluorescence UV-Vis and FT-IR were analysed and GC-MS analysis revealed more phytochemicals comparatively with wild plants. The compounds were identified by comparing their retention time and peak area with that of literature and by interpretation of mass spectra. More than sixty compounds were extracted from the callus extracts. The presence of phyto-components reveals the importance of the plant as medicinally used. So, it is recommended as a plant of phyto - pharmaceutical importance.

**Keywords :** *Canthium parviflorum*, FT-IR, GC-MS analysis, phytochemicals, callus extracts, in vitro, pharmacological importance.

#### Introduction

Medicinal plants have been used as traditional treatments for numerous human diseases for thousands of years and in many parts of the world. The natural products derived from medicinal plants have proven to be an abundant source of biologically active compounds many of which have been the basis for the development of new lead chemicals for pharmaceuticals.<sup>1</sup> The pharmaceutical companies shows that for some complex diseases, natural products still represent an extremely valuable source for the production of new chemical entities.<sup>2</sup> Phyto pharmacological screening of medicinal plants and their extracts will reveal the presence of valuable compounds and provide insight into new ways of treatment with new drugs. The accumulation of phytochemicals in the plant cell cultures has been studied for more than thirty years, and the generated knowledge has helped in the realization of using cell cultures for production of desired phytochemicals.<sup>3</sup> Although very few plant cell processes are operating commercially, the most successful commercial pharmaceuticals produced from undifferentiated cell cultures are anti-biotic compounds. So, cell suspension culturing is considered one of the best approaches for studying the biosynthesis of natural products, and calli are the richest sources of cell mass when establishing such cultures.<sup>4</sup>

*Canthium parviflorum* Lam. (syn: *Plectoria parviflora*) of Rubiaceae is commonly called as Balusu in Telugu. The leaves and roots are astringent, sweet, thermogenic, constipating and tonic. They are astringent and effective against cough and indigestion.<sup>5</sup> Leaves and roots of this plant are used as astringent, diuretic, febrifuge, anthelmintic, anti diarrhoea and for leucorrhoea<sup>6</sup> and its used for the treatment of diabetes among

major tribal groups in South Tamilnadu.<sup>7</sup> Tribes of Orissa state in India use fruits of this plant to treat headache. Traditionally the roots and leaves are used to cure vitiated conditions of Kapha in fever and constipation.<sup>8</sup>

## Material & methods

### Callus culture.

Fresh, young leaf material was collected and washed thoroughly under running tap water to remove dust particles. Leaf explants was excised aseptically and cultured on MS (Murashige & Skoog)<sup>9</sup> medium (1962) supplemented with BA (Benzyl adenine) (0.2 mg/l) and NAA (Naphthalene acetic acid) (0.5 and 1.0 mg/l).

### Extraction from Callus Cultures

6 - 8 week-old callus derived from the leaf cuttings were collected and 25g of wild leaf explants taken after dried in an oven at  $40 \pm 1^\circ \text{C}$  for 5 hours. 25g of leaf callus powder were extracted with 150 ml of solvent ethyl acetate and methanol for 24 h by using Soxhlet apparatus. 100 mg / ml were prepared by redissolving the extracted powder in the same solvent which was used in the extraction. This callus extracts was used for FTIR and GC-MS analysis.

### Spectroscopic analysis

For UV-Vis and FT-IR spectrophotometer analysis, the crude extracts and wild plant leaf extracts were centrifuged at 3000 rpm for 10 min and filtered through Whatmann No. 1 filter paper by using high pressure vacuum pump. The sample is diluted to 1:10 with the same solvent. The extracts were scanned in the wavelength ranging from 1100-3000 nm using Perkin Elmer Spectrophotometer and the characteristic peaks were detected. FT-IR analysis was performed using Perkin Elmer Spectrophotometer system, which was used to detect the characteristic peaks and their functional groups. The peak values of the UV-Vis and FT-IR were recorded. Each and every analysis was repeated twice for the spectrum confirmation.

### Procedure

The GC-MS analyses were carried out in a GC Model: 7890A GC System, MS: 5975C Inert MSD with Triple Axis Detector. Gas chromatograph fitted with a DB1 (methylphenylsiloxane, 30 m  $\times$  0.25 mm i.d.) capillary column. The MS operating parameters were as follows: ionization potential 70 eV; ion source temperature 200°C; quadrupole 100°C, solvent delay 6.0 min, scan speed 2000 amu/s, total MS running time 35min. and Mass Scan Range: 30 to 600m/z, eV voltage 3000 volts. The concentrated extract is injected into the GC/MS instrument (HP-5MS 30m\*0.25um\*0.25 Agilent Technologies Part No: 19091S-433).

The sample is volatilized at the injection port and eluted through a capillary column under increasing temperature. As the sample moves through the column, various components are separated due to their affinity for the stationary phase of the column and can be identified by retention time. Each chemical component in a sample has a distinct retention time measured in minutes, shown in a peak on a graph. The integrated peak is correlated to the concentration of the chemical. A mass selective detector breaks up each chromatographic component into fragment ions, which are shown by their abundance, with each ion represented as a vertical line in increasing molecular weight. The height of each line corresponds to the abundance of that ion. The resulting mass spectrum is unique to that chemical. This mass spectrum forms a "fingerprint" that can identify the compound by a computer search of mass spectra. A computer search of the mass spectra corresponding to all the chromatographic peaks for a sample should yield a statistical match for nicotine at a 12.9 min retention time value. First, there is a "Scan" mode which looks at all the constituents of a sample, listing whatever chemical components are present

### Compound Identification

Components of the ethyl acetate, methanol extracts were identified by comparison of their mass spectra and retention indices with those published in the literature and contained in The National Institute of Standard and Technology (NIST) library database. Library Version: 2.0 MS computer library.

## Results

The FTIR (Fourier Transform Infrared Spectrometer) was performed from the plant callus extract of *Canthium parviflorum* to analyse the functional group. In *Canthium parviflorum* extract IR spectrum shows strong absorption peaks at 1651.07 cm<sup>-1</sup>, 1112.93 cm<sup>-1</sup>, 1735.93cm<sup>-1</sup>, 2040.69cm<sup>-1</sup>, 2216.21cm<sup>-1</sup>, 2524.82 cm<sup>-1</sup>, 2835.36 cm<sup>-1</sup>, 2947 cm<sup>-1</sup>, 3302 cm<sup>-1</sup> which corresponds to alkene (C=C) and ether (C-O) groups (Table. 1 and Fig. 1).

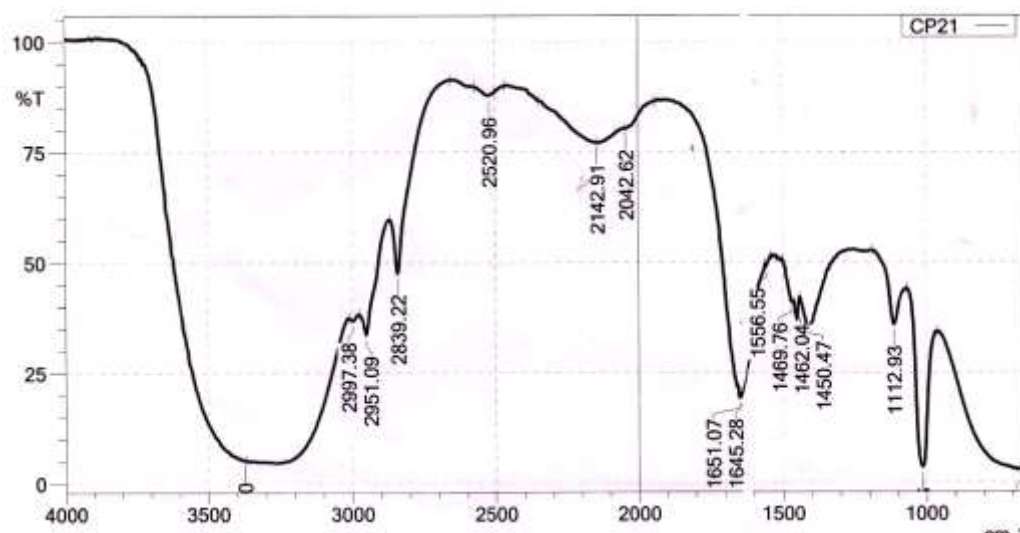


Fig 1 FTIR spectrum of callus extracts *Canthium parviflorum*.

Table 1: FT-IR peak values with functional groups in callus extract of *Canthium parviflorum*.

Peak	Functional Group
1112.93	Aliphatic amines (C-N Stretch) Alcohols, carboxylic acid, esters, ethers (C-O Stretch)
1450.47	Aromatics (C-C Stretch (in-ring))
1556.55	Primary amines
1645.28	Alkenes(-C=C-Stretch), Aldehydes, saturated aliphatic (C=O Stretch)
2042.62	Alkanes (C-H Stretch); Alkanes (terminal) (-triple bond) (C-H:C-H stretch)
2520.96	Primary, secondary amines, amides (N-H Stretch)
2839.22	Alkanes (C-H Stretch); Alkanes (terminal)(-triple bond) (C-H:C-H stretch); Primary, secondary amines, amides
2951.09	Alkanes (C-H Stretch)

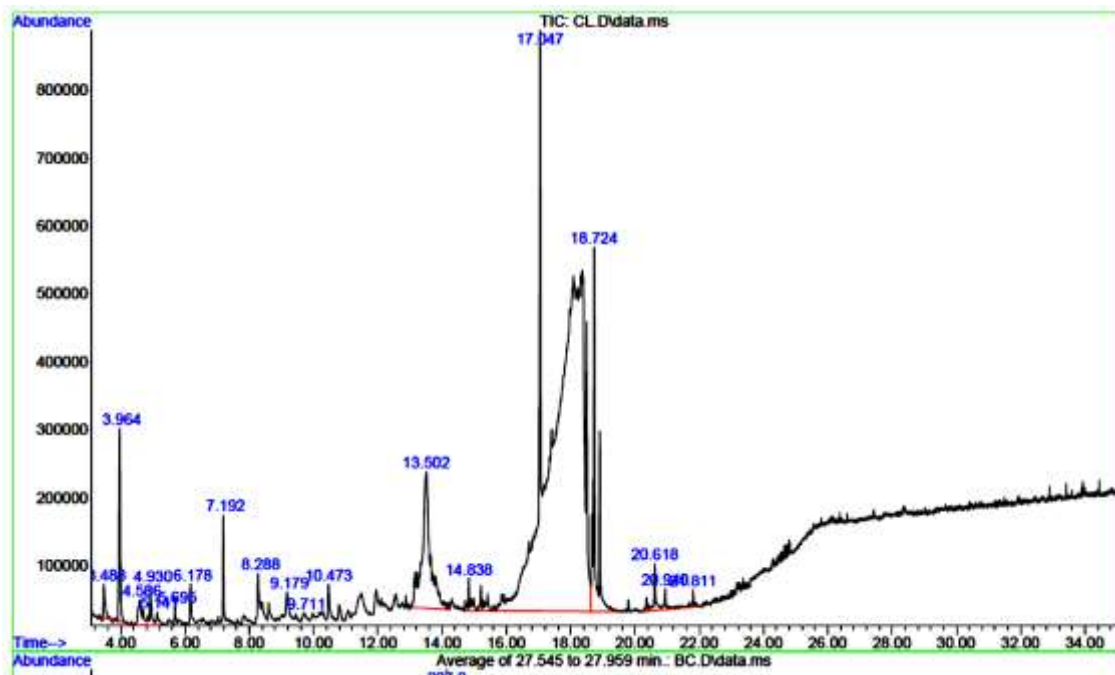
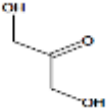
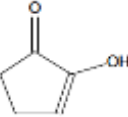
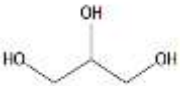
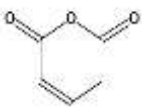
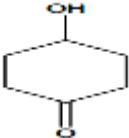
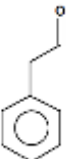
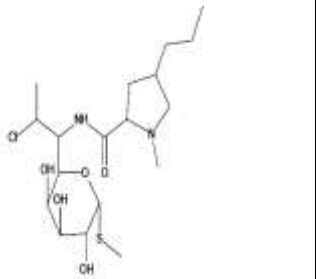
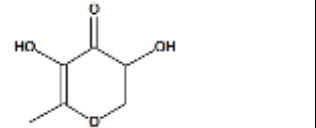
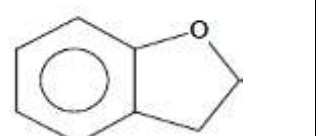
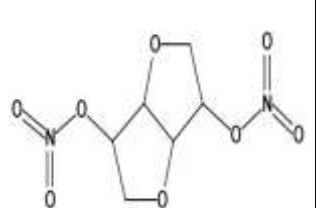
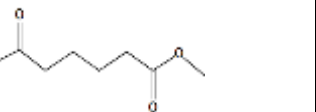
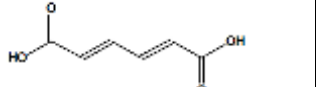
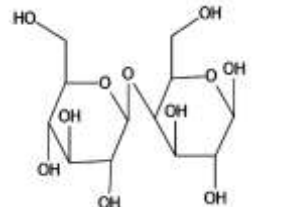
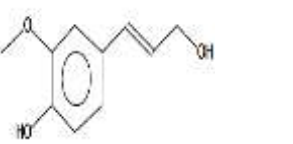
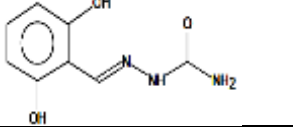
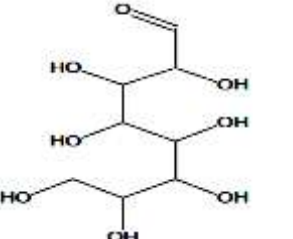
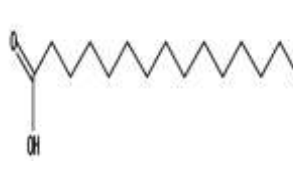
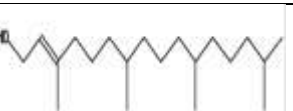


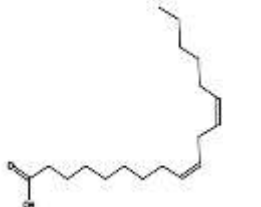
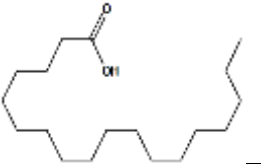

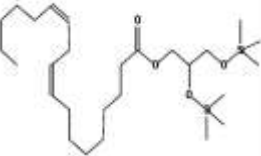
Fig 2 GC MS analysis of leaf methanol extract of *Canthium parviflorum*

Table: 2GC MS analysis of leaf methanol extract of *Canthium parviflorum*

No.	RT	Name of the compound	Molecular formula	Molecular Weight	Peak area %	Structure	Biological activity
1.		2-Propanone, 1,3-dihydroxy	C3H6O3	90			cholinesterase inhibitor drugs
2.		2-Cyclopenten-1-one, 2-hydroxy-	C5H6O2	98			Antimicrobial, Anti-inflammatory, Anticancer, Dieruretic
3.		Glycerin	C3H8O3	92			cough syrups, elixirs and expectorants, toothpaste, mouthwashes, skin care products,
4.		2H-Pyran-2,6(3H)-dione	C5H4O3	112			Antiallergic activity
5.		Cyclohexanone, 4-hydroxy-	C6H10O2	114			antioxidant capacities
6.		Benzeneacetaldehyde	C8H8O	120			Antimicrobial

7.	Clindamycin	C <sub>18</sub> H <sub>33</sub> ClN <sub>2</sub> O <sub>5</sub> S	424		antibacterial
8.	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	C <sub>6</sub> H <sub>8</sub> O <sub>4</sub>	144		antioxidant
9.	Benzofuran, 2,3-dihydro	C <sub>8</sub> H <sub>8</sub> O	120		Neuroinflammation
10.	IsosorbideDinitrate	C <sub>6</sub> H <sub>8</sub> N <sub>2</sub> O <sub>8</sub>	236		chest pain, headaches
11.	Methyl 6-oxoheptanoate	C <sub>8</sub> H <sub>14</sub> O <sub>3</sub>	158		Anti cancer
12.	2,4-Hexadienedioic acid	C <sub>6</sub> H <sub>6</sub> O <sub>4</sub>	142		preservatives in food and drinks to prevent the growth of mold, yeast, and fungi

13.	$\beta$ -D-Glucopyranose, 4-O- $\beta$ -D-galactopyranosyl	C <sub>12</sub> H <sub>22</sub> O <sub>11</sub>	342		sweeten food, beverages, medications, arthritis. <sup>16</sup>
14.	4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol	C <sub>10</sub> H <sub>12</sub> O <sub>3</sub>	180		Antioxidant, Antimicrobial Anti inflammatory
15.	2,6-Dihydroxybenzaldehyde, carbamoylhydrazone	C <sub>8</sub> H <sub>9</sub> N <sub>3</sub> O <sub>3</sub>	195		lipid peroxidation
16.	$\beta$ -Gala-1-ido-octose	C <sub>8</sub> H <sub>16</sub> O <sub>8</sub>	240		<b>Used for memory Drugs production</b>
17.	n-Hexadecanoic acid	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	256		Antioxidant, Hypochloesterolemic, Nematicide, Pesticide, Lubricant, Antiandrogenic, Hemolytic,5- Alpha reductase inhibitor
18.	Phytol	C <sub>20</sub> H <sub>40</sub> O	296		Anticancer, antioxidant, antiinflammatory and diuretic

19.	9,12-Octadecadienoic acid (Z,Z)-	C <sub>18</sub> H <sub>32</sub> O <sub>2</sub>	280			Anti-inflammatory, Hypercholesterolemic, Cancer preventive, Hepatoprotective, Nematicide, Insectifuge, Antihistamine, Anti-eczemic, Anti-acne, 5-Alpha reductase inhibitor, Antiandrogenic, Anti-arthritis, Anti-coronary
20.	Octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284			Antimicrobial and anti-inflammatory, hepatoprotective, hypercholesterolemic, anticancer
21.	1-Heptatriacotanol	C <sub>37</sub> H <sub>76</sub> O	536			anticancer, antineoplastic and anti-HIV
22.	1-Monolinoleoylglycerol trimethylsilyl ether	C <sub>27</sub> H <sub>54</sub> O <sub>4</sub> Si <sub>2</sub>	498			Antimicrobial Antioxidant Antiinflammatory Antiarthritic Antiasthma, Diuretic



The most prevailing compounds from *Canthium parviflorum* leaf were listed methanolic extracts 2-Propanone, 1,3-dihydroxy, 2-Cyclopenten-1-one, 2-hydroxy-, Glycerin, 2H-Pyran-2,6(3H)-dione, Cyclohexanone, 4-hydroxy-, Benzene acetaldehyde, Clindamycin, 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-, Benzofuran, 2,3-dihydro, IsosorbideDinitrate, Methyl 6-oxoheptanoate, 2,4-Hexadienedioic acid,  $\beta$ -D-Glucopyranose, 4-O- $\beta$ -D-galactopyranosyl, Phytol, Octadecanoic acid, 4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol, 2,6-Dihydroxybenzaldehyde, n-Hexadecanoic acid, Carbamoylhydrazone, l-Gala-l-ido-octose, 9,12-Octadecadienoic acid (Z,Z)-, 9,12,15-Octadecatrienoic acid, (Z,Z,Z)-, 1-Heptatriacotanol, 1-Monolinoleoylglycerol trimethylsilyl ether. The GC-MS chromatogram shows the peak area separation.

Based on the previous reports Biphenyl, 2-Methyl-4-heptanone, 1,2,4,5-Tetroxane, 3,3,6,6-Tetraphenyl-, 3-Oxo-Alapha-, Ionol, Methyl 7-hydroxy-2-methyl-3,5-octadienoate, 4-(2-Hydroxy-2,6,6-Trimethylcyclohexyl)-3-buten-2-one, n-Hexadecanoic acid, E-11- Hexadecanoic acid, Ethylester, EthylHexadecanoate, Phytol, Ethyl(9Z,12Z)-9,12- Octadecanoate, Ethyl Linolenate, 1-Hexadecanol, 2-Phenoxy-2-phenylpropanoic acid, All-trans-squalene, Methyl Linolenate, Gamma-Tocopherol, DEPH; 1,2-Benzenedicarboxylic acid, bis (2-hylhexyl)ester, Stigmasterol, Gamma-stigmasterol, Methyl cis-11,14,17-Icosatrienoate.<sup>10</sup>

Based on the previous reports on the *Canthium coromandelicum* leaf extract revealed Thirteen phytochemicals by GC-MS analysis. The components are 1,6,10-odecatriene, 7,11-dimethyl -3-methylene-, (E), 3,7,11,15-Tetramethyl-2-Hexadecan-1-ol, 2-Tridecen-1-ol, E-2-Tetradecen-1-ol, Phytol, 2-Aminononadecane, Octadecane, 6-methyl-, Didodecyl phthalate, 1-Nonodecanol, Valeric acid, 2-Pentadecyl ester, Squalene, Z-8-Methyl-9-tetradecenoic acid, Heptadecanoic acid.<sup>11</sup> *Canthium parviflorum* leaf with methanol extracts presence of the phytol compound exhibits more biological properties like Antimicrobial, Anticancer, Cancer preventive, Diuretic Antimicrobial, Anticancer, Cancer preventive, Diuretic, Anti-inflammatory.<sup>9</sup> In our results we observed the phytol compound present in all areas of *Canthium parviflorum* plants including Guntur district plants also, comparatively with Tamilnadu and Chennai are plants revealed the compounds also phytol n-Hexadecanoic acid.<sup>10,11</sup> Octadecanoic acid possesses antiinflammatory, hepatoprotective, hypercholesterolemic, anticancer and many other properties<sup>12,13</sup> and Antimicrobial activity of hexadecanoic acid was discussed by Bergsson.<sup>14</sup> The l-gala idooctose used for the production of Drugs used to specifically facilitate learning or memory, particularly to prevent the cognitive deficits associated with dementias.<sup>15</sup> In the presence of the compound is 6- dihydroxybenzaldehyde used in the lipid peroxidation.<sup>16</sup> So comparatively with these results more compounds revealed the Guntur district area plants of *Canthium parviflorum* than the Tamilnadu area plants.

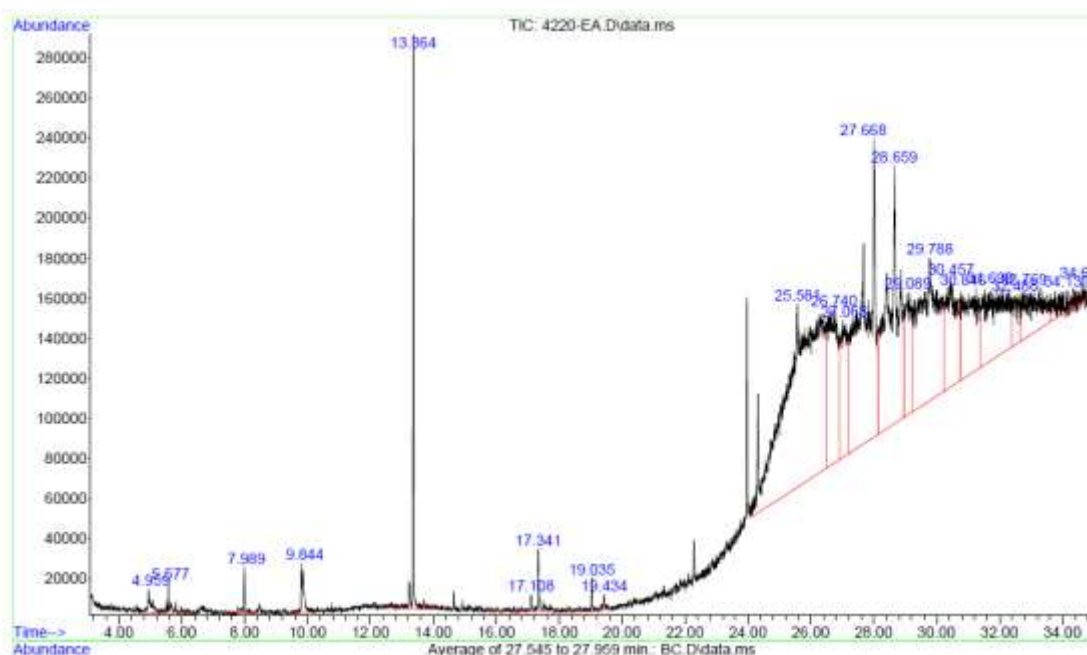

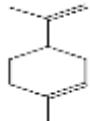
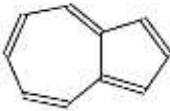
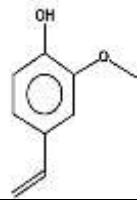
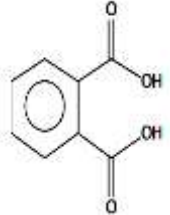

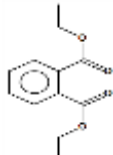

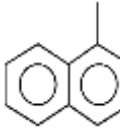
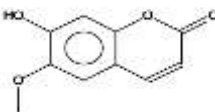
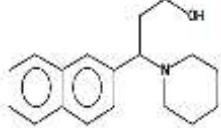
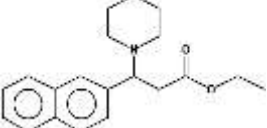
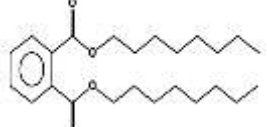
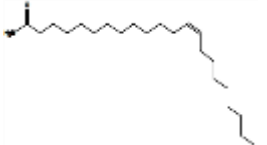
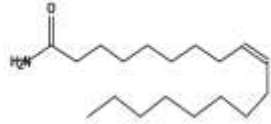
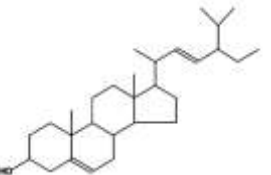


Fig 4 GC MS analysis of callus extract with ethyl acetate of *Canthium parviflorum*

Table: 3 GC MS analysis of callus extract with ethyl acetate of *Canthium parviflorum*

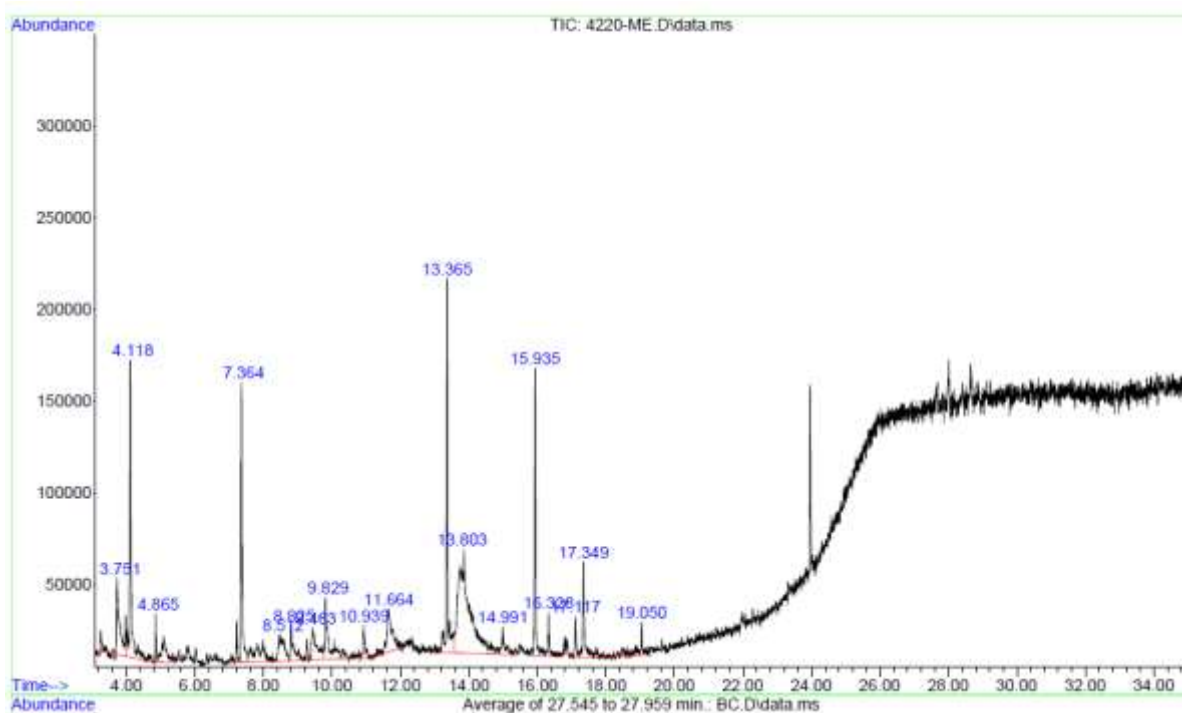
No.	RT	Name of the compound	Molecular formula	Molecular Weight	Peak area %	Structure	Biological activity
1.	3.76	2-Nonenol	C <sub>9</sub> H <sub>16</sub> O	140	10.18		Lipid peroxidation
2.	4.12	Cyclohexane	C <sub>10</sub> H <sub>16</sub>	136	15.82		Antimicrobial agents
3.	7.36	Azulene	C <sub>10</sub> H <sub>8</sub>	128	14.44		Antiulcer Antimicrobial Antiallergic Antiinflammatory, Antipyretic Antiseptic Anticancer
4.	9.43	2-Methoxy-4-vinylphenol	C <sub>9</sub> H <sub>10</sub> O <sub>2</sub>	128	2.295		Antioxidant Antimicrobial Anti-inflammatory
5.	13.70	1,2-Benzenedicarboxylic acid	C <sub>8</sub> H <sub>6</sub> O <sub>4</sub>	166	13.90		Used as Softeners, perfumes and cosmetics, plasticized vinyl seats on furniture and in cars, and clothing including jackets, raincoats and boots. Used in textiles, as dyestuffs, cosmetics and glass making
6.	15.93	2-Naphthalenemethanol	C <sub>11</sub> H <sub>10</sub> O	158	13.39		anti-micro-organism

7.	13.35	Diethyl phthalate	C <sub>12</sub> H <sub>14</sub> O <sub>4</sub>	222	13.39		Antimicrobial Antifouling
8.	13.41	Dodecanol	C <sub>12</sub> H <sub>24</sub> O	184	4.80		Antioxidant
9.	19.04	Naphthalene, 1-methyl	C <sub>11</sub> H <sub>10</sub>	142	1.86		Anti-tumor, analgesic antibacterial, anti-inflammatory sedative, fungicide
10.	18.34	7-Hydroxy-6methoxy-2H-1-benzopyran-2-one	C <sub>10</sub> H <sub>8</sub> O <sub>4</sub>	192	1.42		Antimicrobial
11.	16.22	3-Naphthalene-2yl-3-piperidin-1yl-propan-1-ol	C <sub>18</sub> H <sub>23</sub> N O	269	1.22		anti- insect, anti- microbial
12.	23.96	1-Piperidinepropanoic acid	C <sub>22</sub> H <sub>29</sub> N O <sub>2</sub>	339	1.86		Urinary track infections, Antiviral, antidepressant, cytotoxic and antimalarial activity.
13.	20.28	Di-n-octyl phthalate	C <sub>24</sub> H <sub>38</sub> O <sub>4</sub>	390	1.26		Antivenom
14.	18.22	13-Docosamide	C <sub>22</sub> H <sub>43</sub> N O	337	1.44		antimicrobial, antioxidant and antiinflammatory properties

15.	16.24	9-Octacenamide	C <sub>18</sub> H <sub>35</sub> N O	281	1.68		Anti-inflammatory activity, antibacterial activity, and anti-cancer properties
16.	18.66	Stigmasterol	C <sub>29</sub> H <sub>48</sub> O	412	1.22		Antioxidant, hypoglycemic and thyroid inhibiting properties, precursor of progesterone, antimicrobial, anticancer, antiarthritic, antiasthama, anti inflammatory, diuretic

The most prevailing major compounds were revealed from *Canthium parviflorum* callus with ethyl acetate extracts 2-Nonenol, Cyclohexane, Azulene, 2-Methoxy-4-vinylphenol, 1,2-Benzenedicarboxylic acid, 2-Naphthalenemethanol, Diethyl phthalate, 2-Decen-1-ol, Naphthalene, 1-methy, 7-Hydroxy-6methoxy-2H-1-benzopyran-2-one, 3-Naphthalene-2yl-3-piperidin-1yl-propan-1-ol, 1-Piperidinepropanoic acid, Di-n-octyl phthalate, 13-Docosenamide, 9-Octacenamide, Stigmasterol.

Prabhu *et al.*, 2013 reported the Cyclohexane,1,2-Benzenedicarboxylic acid, Stigmasterol phyto compounds are present in the ethyl acetate leaf extract of *Canthium parviflorum*, these compounds matched with our ethylacetate leaf callus extract results. In the presence of the callus extracts Stigmasterol, it has been reported to induce apoptosis in Ehrlich's ascites carcinoma in mice through the activation of protein phosphatase 2A via ceramide.<sup>17</sup> Most of these compounds have been associated with several biological or pharmaceutical properties; it's responsible for medicinal potential of the plants.

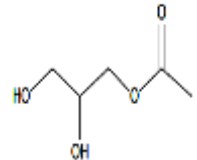
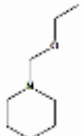
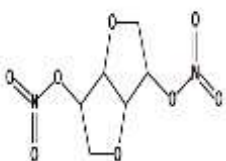
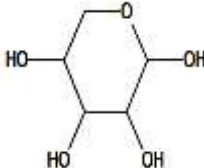
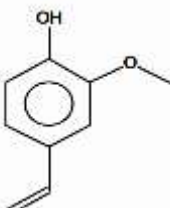



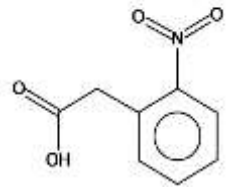
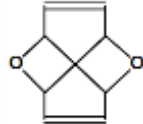
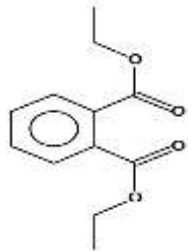
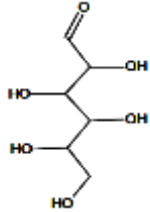
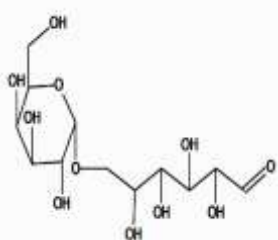
**Fig 6 GC MS analysis of callus extract with methanol of *Canthium parviflorum***

The most prevailing major compounds were revealed from callus with methanol extracts 2-Propanone, 1,3-dihydroxy, 1,2-Ethandiol, 3-Buten-1-ol, Butyrolactone, 2-Cyclopenten-1-one, 2-hydroxy-, Cyclohexanone, 2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one, 1,3-Dioxane, 2,4-dimethyl-, Urea, N-butyl-N-nitroso-, Formic acid, butyl ester, 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-, Isosorbide Dinitrate, 1,2,3-Propanetriol, monoacetate, Piperidine, 1-(ethoxymethyl, 1-(5-Hydroxypentyl) piperidine, Isosorbide Dinitrate, DL-Arabinose, 2-Methoxy-4-vinylphenol, 4-Hydroxy-2-methylacetophenone, Undecanoic acid, 11-amino-, 1-Ethyl-1-propoxy-1-silacyclopentane, Benzeneacetic acid, 2-nitro-, 2,7-Dioxa-tricyclo[4.4.0.0(3,8)]deca-4,9-diene, Diethyl Phthalate, d-Mannose, 5-(1-Hydroxy-1-methyl-ethyl)-3-(4-methoxy-phenyl)-isoxazolidin-5-ol,  $\alpha$ -D-Glucopyranoside, O- $\alpha$ -D-glucopyranosyl-(1.fwdarw.3)- $\beta$ -D-fructofuranosyl, D-Glucose, 6-O- $\alpha$ -D-galactopyranosyl-, Phenol, 4-(3-hydroxy-1-propenyl)-2-methoxy-, 4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol, Acetamide, N-(4-ethoxy-3-hydroxyphenyl)-, Piperidine, 1-cyclohexyl-, 1,2-Benzene dicarboxylic acid, butyl octyl ester, 1-Naphthaleneacetamide, Naphthalene, 1-methyl-, 5-Ethyl-3,4-dimethyl-1H-pyran[2,3-c]pyrazol-6-one, 5,6-Dimethoxy-1-indanone, Naphthalene, 1H-Purine-2,6-dione, 3,7-dihydro-8-(hydroxymethyl)-1,3,7-trimethyl-, 3-Naphthalen-2-yl-3-piperidin-1-yl-propan-1-ol, 13-Docosenamide, (Z)-.

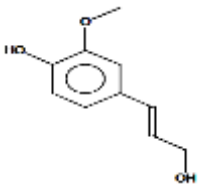
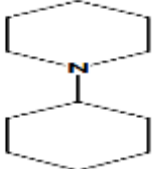
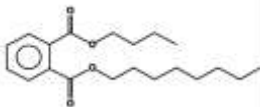
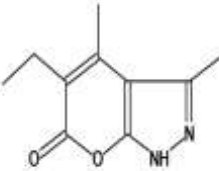
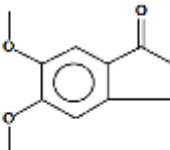
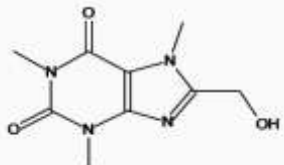
Table: 4 GC MS analysis of callus extract with methanol of *Canthium parviflorum*

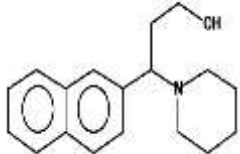
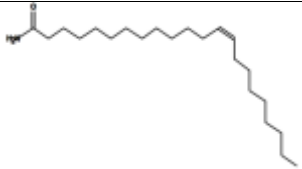
No.	RT	Name of the compound	Molecular formula	Molecular Weight	Peak area %	Structure	Biological activity
1.	3.766	2-Propanone, 1,3-dihydroxy-	C <sub>3</sub> H <sub>6</sub> O <sub>3</sub>	90	1.85		It is used to make plastic, fibers, drugs, and other chemicals.
2.	7.362	Butyrolactone	C <sub>4</sub> H <sub>6</sub> O <sub>2</sub>	86	2.92		antioxidant and analgesic activities
3.	4.282	Cyclohexanone	C <sub>6</sub> H <sub>10</sub> O	98	4.24		Anti-microbial agents
4.	7.362	2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one	C <sub>6</sub> H <sub>8</sub> O <sub>4</sub>	144	1.48		flavor compounds in many fruit
5.	4.622	Formic acid, butyl ester	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub>	102	3.22		antibacterial agent
6.	4.282	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	C <sub>6</sub> H <sub>8</sub> O <sub>4</sub>	144	5.34		antibacterial activity
7.	5.262	IsosorbideDinitrate	C <sub>6</sub> H <sub>8</sub> N <sub>2</sub> O <sub>8</sub>	236	5.22		therapy of heart failure

8.	2.264	1,2,3-Propanetriol, monoacetate	C <sub>5</sub> H <sub>10</sub> O <sub>4</sub>	134	4.48		Anti fungal
9.	3.766	Piperidine, 1-(ethoxymethyl)-	C <sub>8</sub> H <sub>17</sub> NO	143	5.45		anticoagulant activity
10.	3.766	IsosorbideDinitrate	C <sub>6</sub> H <sub>8</sub> N <sub>2</sub> O <sub>8</sub>	236	2.662		decrease of blood pressure, angina pectoris
11.	8.726	DL-Arabinose	C <sub>5</sub> H <sub>10</sub> O <sub>5</sub>	150	2.46		Antitumor
12.	4.762	2-Methoxy-4-vinylphenol	C <sub>9</sub> H <sub>10</sub> O <sub>2</sub>	150	6.82		Antioxidant Antimicrobial Anti-inflammatory, Anticancer,
13.	4.882	Undecanoic acid, 11-amino-	C <sub>11</sub> H <sub>23</sub> NO <sub>2</sub>	201	2.428		No activity reported

14.	4.624	Benzeneacetic acid, 2-nitro-	C <sub>8</sub> H <sub>7</sub> NO <sub>4</sub>	181	4.62		No activity reported
15.	8.811	2,7-Dioxatricyclo[4.4.0.0(3,8)]deca-4,9-diene	C <sub>8</sub> H <sub>8</sub> O <sub>2</sub>	136	3.69		color cosmetic composition
16.	13.359	Diethyl Phthalate	C <sub>12</sub> H <sub>14</sub> O <sub>4</sub>	222	13.90		Antimicrobial Antifouling
17.	19.045	d-Mannose	C <sub>6</sub> H <sub>12</sub> O <sub>6</sub>	180	1.86		Sugar moiety and Presevative
18.	3.766	D-Glucose, 6-O-α-D-galactopyranosyl-	C <sub>12</sub> H <sub>22</sub> O <sub>11</sub>	342	2.29		Sugar moiety and Presevative



19.	19.045	Phenol, 4-(3-hydroxy-1-propenyl)-2-methoxy-	C <sub>10</sub> H <sub>12</sub> O <sub>3</sub>	180	1.86		antimicrobial activity antiviral, anti-inflammatory, cytotoxic activity, antimutagenic and anticarcinogenic activities, Antioxidant .
20.	4.220	Piperidine, 1-cyclohexyl-	C <sub>11</sub> H <sub>21</sub> N	167	1.86		allergic or inflammatory disorders such as asthma
21.	4..288	1,2-Benzenedicarboxylic acid, butyloctyl ester	C <sub>20</sub> H <sub>30</sub> O <sub>4</sub>	334	4.24		antimicrobial cytotoxicity antioxidant
22.	6.224	Naphthalene, 1-methyl-	C <sub>11</sub> H <sub>10</sub>	142	4.60		fungicidal, antimicrobial
23.	3.7661	5,6-Dimethoxy-1-indanone	C <sub>11</sub> H <sub>12</sub> O <sub>3</sub>	192	2.24		Antioxidative
24.	18.22	1H-Purine-2,6-dione, 3,7-dihydro-8-(hydroxymethyl)-1,3,7-trimethyl-	C <sub>9</sub> H <sub>12</sub> N <sub>4</sub> O <sub>3</sub>	224	4.62		antitubercular, fungicidal, antiallergic, antimicrobial, antitumor, antihistamic

25.	20.34	3-Naphthalen-2-yl-3-piperidin-1-yl-propan-1-ol	C <sub>18</sub> H <sub>23</sub> NO	269	6.99		fungicidal, antimicrobial
26.	23.967	13-Docosamide, (Z)-	C <sub>22</sub> H <sub>43</sub> NO	337	7.69		antimicrobial, antioxidant and antiinflammatory properties

1,2-Benzenedicarboxylic acid Used as Softeners, Used in preparation of perfumes and cosmetics.<sup>18</sup> Naphthalene is well known for its antimicrobial activity.<sup>19,20</sup> The phenolic compounds are known to be synthesized by plants in response to microbial infection. It is therefore possible that they can act as effective antimicrobial substances against a wide array of microorganisms. Among the identified compounds phenol, 4H-pyran-4-one, 1,2-benzene dicarboxylic acid, octadecanoic acid have the antibacterial activity as reported by earlier workers.<sup>21</sup>

## Discussion

In recent years, the interest for the study of the organic compounds from plant callus extracts and their activity has increased. The combination of an ideal separation technique (GC) with the best identification technique (MS) made GC-MS an ideal technique for qualitative and quantitative analysis for volatile and semi-volatile compounds. The aim of the present study was to develop a rapid method for the quantitative determination of organic compounds in plant callus extracts and to confirm the phytochemical present in the wild plant extracts and co relation of the presence of compounds and to know the differentiation.

*Canthium parviflorum* ethyl acetate callus extracts are contained 1,2-Benzenedicarboxylic acid and its used in synthesis of dyes and perfumes; neurodegenerative disorders. The result of the present investigation reveals that the methanolic extracts of *Canthium parviflorum* possessed significant anticancer activity which was analyzed by GC-MS analysis. Different phytochemicals have been found to have a broad range of activities, which may help in protection against chronic diseases.

## Conclusion

*Canthium parviflorum* callus extracts are contained various phytochemicals with biological activity can be of valuable therapeutic key. The GC-MS analysis of the ethyl acetate and methanol callus extract of *Canthium parviflorum* reveals the presence of phytoconstituents. The presence of such a variety of phytochemicals may be attributed to the medicinal characteristics of this plant *Canthium parviflorum* callus extract. The presence of phyto-components reveals the importance of the plant as medicinally used. Further investigations are planned to conduct the pharmacological studies to know the potency of these extracts. So, it is recommended as a plant of phyto - pharmaceutical importance.

## Acknowledgements

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