

## Gc-MS Analysis of Whole Plant of *Leptadenia Reticulata*

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**Abstract :** The aim of the present study was to evaluate the phyto components present in the ethanolic extract of whole plant of *Leptadenia reticulata* by using GC-MS method. The chemical compounds were investigated using Perkin-Elmer Gas chromatography- Mass spectrometry. GC-MS analysis of ethanol extract of *Leptadenia reticulata* revealed the existence of thirty one compounds.

**Key Words:** Phyto components, *Leptadenia reticulata*, Perkin-Elmer Gas chromatography- Mass spectrometry.

### Introduction

During the twentieth century, when exploring the natural environment, man has made great discoveries that have enabled him to use a considerable number of natural resources.<sup>1</sup> Plants are like natural laboratories where a great number of chemicals are biosynthesized and in fact they may be considered the most important source of chemical compounds. Phyto therapy, the use of plants to medical purposes, is one of the oldest practices in the world. The traditional practice, based on empirical data, is considered as folk medicine and the approach based on scientific studies aims to extract and study active components from plants. Medicinal plants or their derived material, have been widely employed in all cultures, throughout history, for the prevention and treatment of diseases. The significant increase in the use of herbal medicines in recent decades may be attributed to popular wisdom, the costs of synthetic drugs and the resurgence of interest in the development of new drugs and the re-establishment of old ones from plant sources.<sup>2</sup> In many urbanized countries well-liked use of Complementary and Alternative Medicine (CAM) is fuelled by concern about the undesirable effects of synthetic drugs. In budding countries, broad use of Traditional Medicine (TM) is often attributable to its ease of access and availability. For the last few years, there has been a global trend for the regeneration of awareness in the traditional system of treatments. Simultaneously investigation of medicinal plants using indigenous medical systems has become ever more important for speed up better and effective treatment.<sup>3</sup> Herbal medicines are safer than synthetic medicines because the phytochemicals in the plant extract target the biochemical pathway. Traditional systems of medicines are prepared from a single plant or combinations of number of plants. The efficacy depends on the use of proper plant part and its biological potency which in turn depends upon the presence of required quantity and nature of secondary metabolite in a raw drug.<sup>4,5</sup> Screening of active components from plants has unswerving to the development of new medicinal drugs which have well-organized fortification and treatment role against various diseases.<sup>6</sup> *Leptadenia reticulata* (jivanti) is distributed in tropical and sub-tropical parts of Asia and Africa . In India, it is found in Gujarat, sub - Himalayan tracts from Punjab to Sikkim and Khasi hills and throughout peninsular India, ascending up to a altitude of 900 metres. Jivanti is jeevana to nichh at boosts energy level of the body as per according to ayurveda. It is beneficial for the patient for the persons from weak debility or a lack of energy. It also increases longevity, memory enhancement ,immune modulation and adoption.<sup>7</sup> The whole plant ameliorates 'tridoshas' (Vatta, Pitta and Kapha), and is of great value in general debility, involuntary seminal discharge, as a stimulant and snake bite<sup>8-9</sup>, abortifacient, tonic, restorative, bactericidal, antifabrifuge, prostitutes, wound healer and in mouth ulcer.<sup>10</sup>

## Materials and Methods

### Collection and Identification of Plant Materials

Fresh leaves of the selected plant *Leptadenia reticulata* were collected from Thirunelveli district, Tamil Nadu, India. The plant materials were taxonomically identified and authenticated by Dr. V. Chelladurai, Research officer - Botany (scientist C), Central council for research in Ayurveda and Siddha, Govt. of India; Thirunelveli. Whole plant were cleaned, shade dried and pulverized to powder in a mechanical grinder. The powdered materials were stored in air tight polythene bags till use.

### Preparation of Extracts

The dried parts were extracted with ethanol using soxhlet extractor.<sup>11</sup> The extract which is obtained is concentrated with rotary evaporator till dry powder was obtained. The final concentrated extract is analysed by using GC-MS.

### Gas Chromatography-Mass Spectrometry (Gc-Ms) Analysis

GC-MS analysis was carried out on a GC Clarus 500 Perkin Elmer system and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: column Elite-MS fused silica capillary column (30m x 250  $\mu$ m), composed of (5% Phenyl 95%Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 2.0 $\mu$ l was employed (split ratio of 1:10) injector temperature 280°C; ion source temperature 150°C. The oven temperature was programmed at 50°C at 8°C/min to 220°C for 5 mins with an increase of 7°C/min, to 280°C for 15 min .Mass spectra were taken at 70 eV; Mass range is from 40-600 amu is maintained.

### Identification of Components

Interpretation of 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

GC-MS chromatogram of the ethanol extract of whole plant of *Leptadenia reticulata* (Fig. 1) clearly showed thirty one peaks indicating the presence of thirty two phytochemical compounds. The mass spectra of identified compounds were matched with those found in the NIST/NBS spectral database are given in table. 1. The identification of the phytochemical compounds was confirmed based on the peak area, retention time and molecular formula. The medicinal properties of the analyzed phytochemical constituents were listed in table. 2. The individual fragmentation patterns of necessary components were illustrated in Figures A-V.

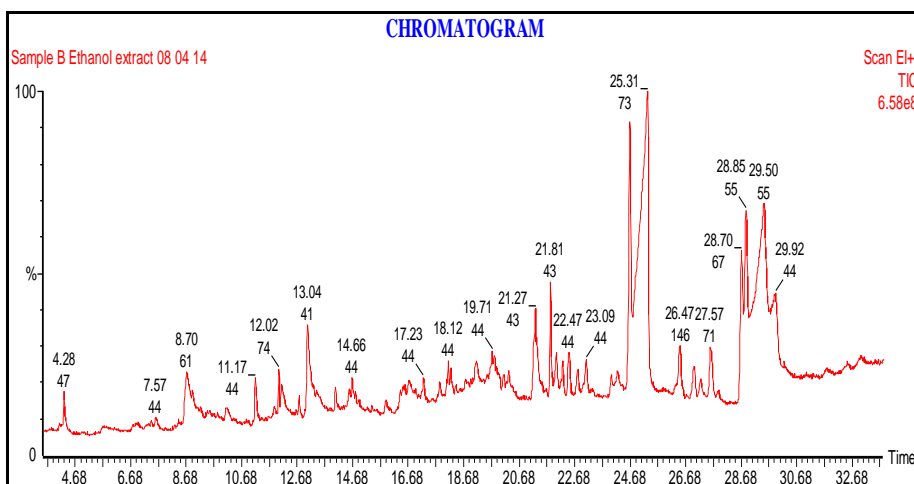


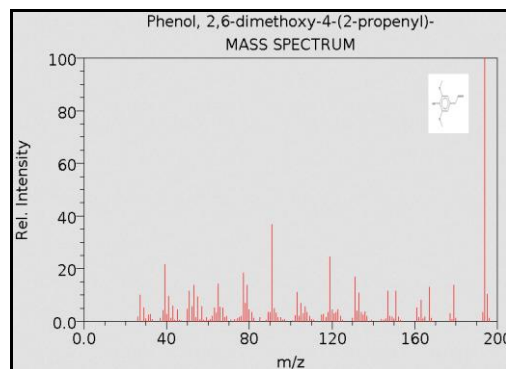
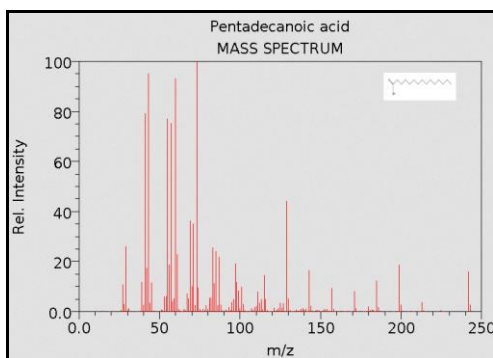
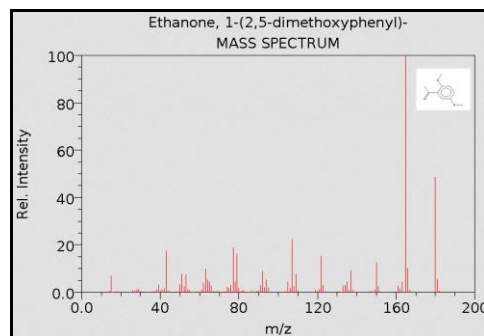
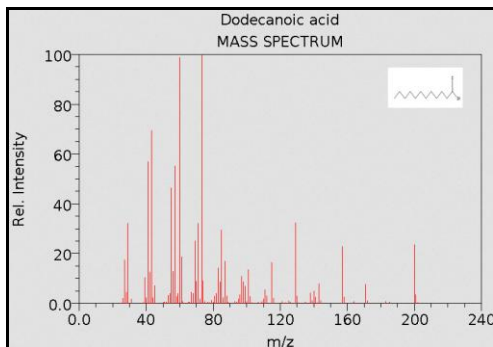
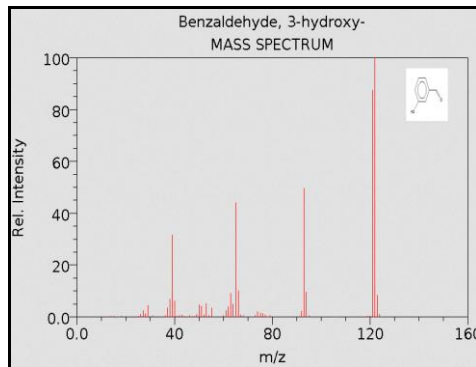
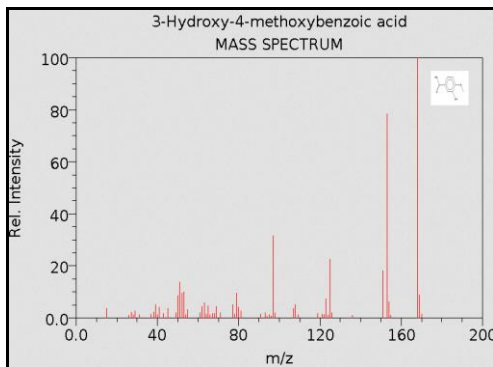
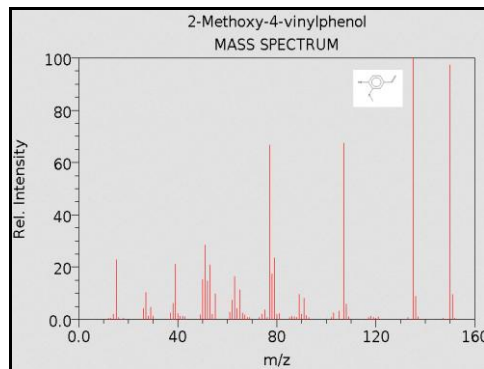
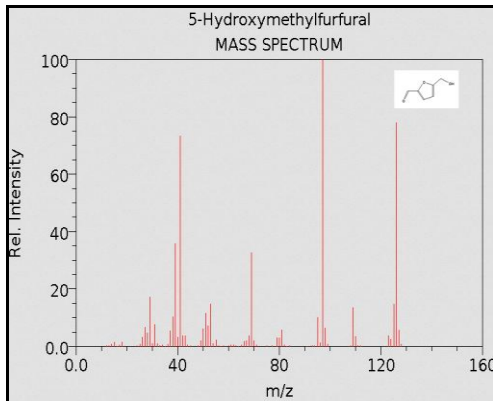
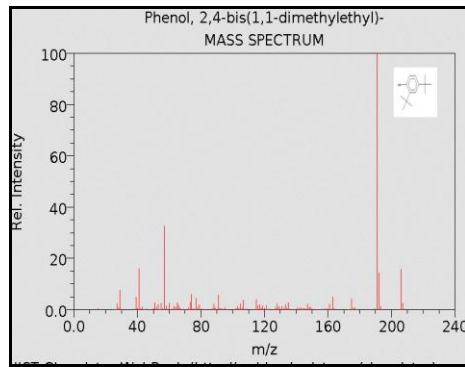
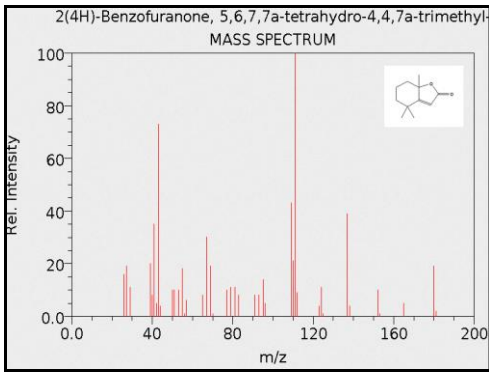
Fig. 1: The GC - MS Chromatogram of ethanol extracts of whole plant of *Leptadenia reticulata*

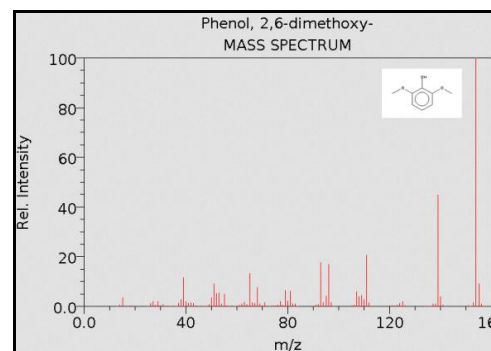
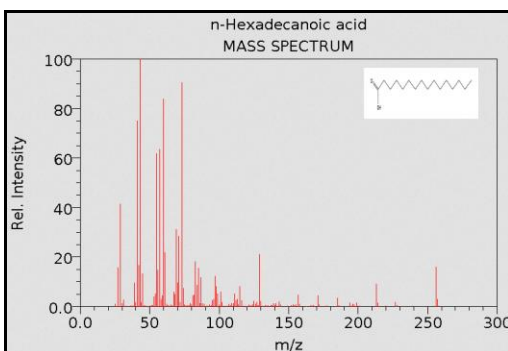
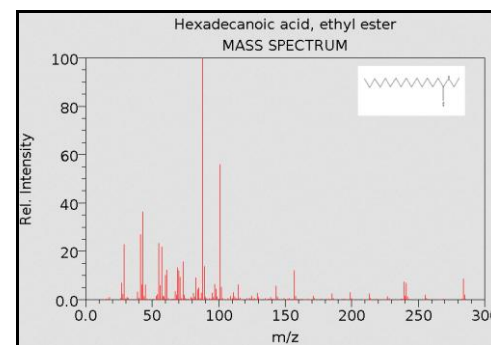
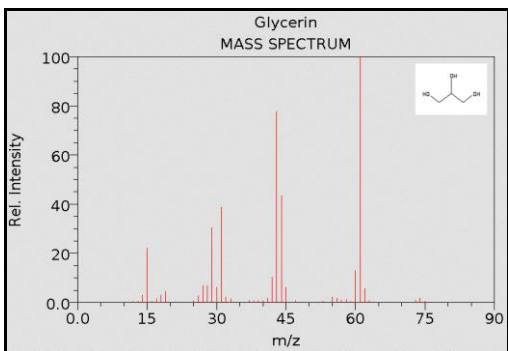
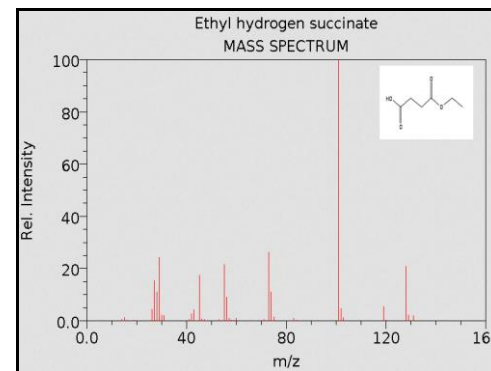
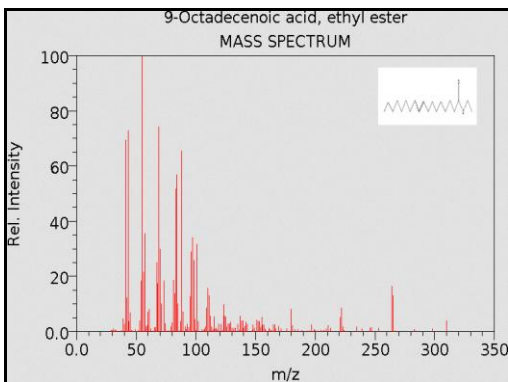
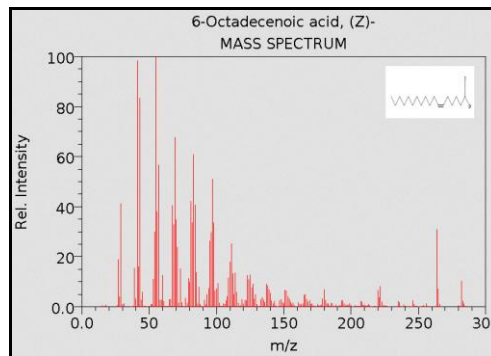
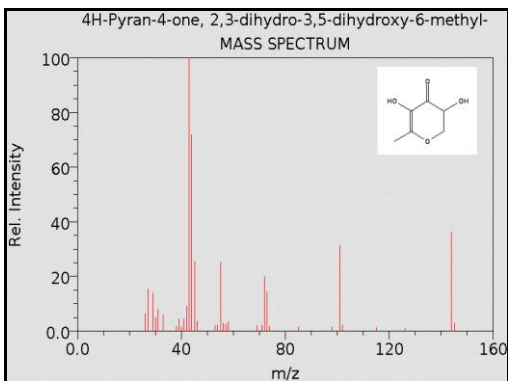
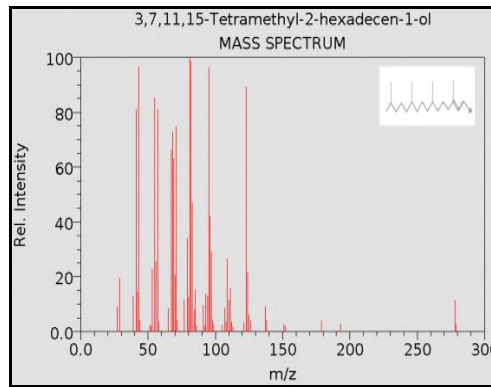
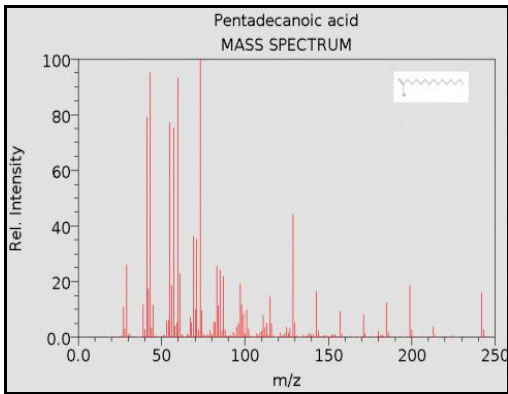
**Table 1: List of Compounds**

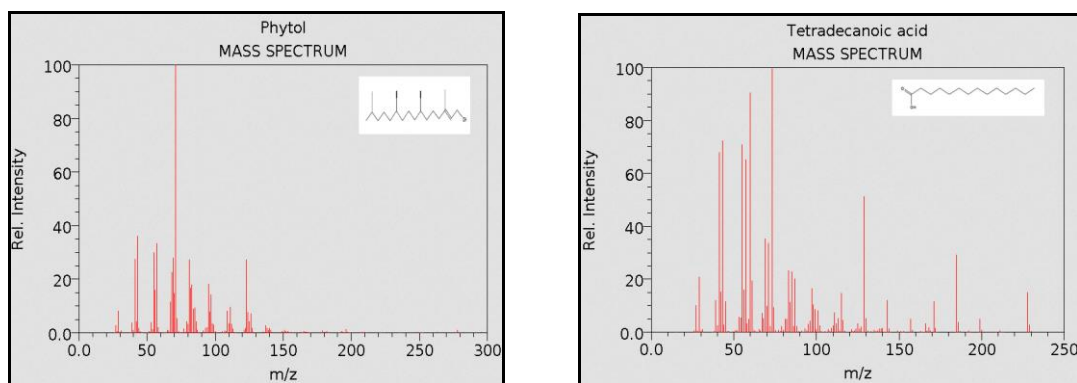
S.No.	Peak Name	Molecular Formula	Molecular Weight	Retention Time	% Peak area
1.	2-Propanone, 1-(ethylthio)-	C <sub>5</sub> H <sub>10</sub> OS	118	4.28	0.9849
2.	Glycerin	C <sub>3</sub> H <sub>8</sub> O <sub>3</sub>	92	8.70	2.4203
3.	1-Penten-4-one, 2-acetyl-1-dimethylamino- ((Z)- or (E)-)	C <sub>9</sub> H <sub>15</sub> NO <sub>2</sub>	169	10.11	0.8084
4.	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	11.17	1.8528
5.	Ethyl hydrogen succinate	C <sub>6</sub> H <sub>10</sub> O <sub>4</sub>	146	11.84	0.4658
6.	á-d-Allopyranoside, methyl 6-deoxy-2-O-methyl-	C <sub>8</sub> H <sub>16</sub> O <sub>5</sub>	192	12.02	0.9721
7.	Benzene carboxylic acid	C <sub>7</sub> H <sub>6</sub> O <sub>2</sub>	122	12.11	1.1104
8.	2-Furancarboxaldehyde, 5-(hydroxymethyl)-	C <sub>6</sub> H <sub>6</sub> O <sub>3</sub>	126	13.04	3.6765
9.	2-Methoxy-4-vinylphenol	C <sub>9</sub> H <sub>10</sub> O <sub>2</sub>	150	14.05	0.8089
10.	Phenol, 2,6-dimethoxy-	C <sub>8</sub> H <sub>10</sub> O <sub>3</sub>	154	14.66	0.5354
11.	Benzaldehyde, 3-hydroxy-4-methoxy-	C <sub>8</sub> H <sub>8</sub> O <sub>3</sub>	152	15.88	0.8500
12.	Phenol, 2,4-bis(1,1-dimethylethyl)-	C <sub>14</sub> H <sub>22</sub> O	206	17.23	0.4185
13.	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-	C <sub>11</sub> H <sub>16</sub> O <sub>2</sub>	180	17.82	0.6981
14.	Dodecanoic acid	C <sub>12</sub> H <sub>24</sub> O <sub>2</sub>	200	18.12	1.2661
15.	3',5'-Dimethoxyacetophenone	C <sub>10</sub> H <sub>12</sub> O <sub>3</sub>	180	18.22	0.9888
16.	3-Hydroxy-4-methoxybenzoic acid	C <sub>8</sub> H <sub>8</sub> O <sub>4</sub>	168	19.15	.3717
17.	Phenol, 2,6-dimethoxy-4-(2-propenyl)-	C <sub>11</sub> H <sub>14</sub> O <sub>3</sub>	194	20.31	0.4260
18.	Tetradecanoic acid	C <sub>14</sub> H <sub>28</sub> O <sub>2</sub>	228	21.27	0.9418
19.	7,11,15-Tetramethyl-2-hexadecen-1-ol	C <sub>20</sub> H <sub>40</sub> O	296	21.81	2.8479
20.	2,6,8-Trimethylbicyclo[4.2.0]oct-2-ene-1,8-diol	C <sub>11</sub> H <sub>18</sub> O <sub>2</sub>	182	22.02	0.1151
21.	2H-Pyran, 6-heptyltetrahydro-2,2-dimethoxy-	C <sub>14</sub> H <sub>28</sub> O <sub>3</sub>	244	22.24	1.1455
22.	Pentadecanoic acid	C <sub>15</sub> H <sub>30</sub> O <sub>2</sub>	242	22.80	0.9175
23.	2,6-Di-O-methyl-d-galactopyranose	C <sub>8</sub> H <sub>16</sub> O <sub>6</sub>	208	24.01	0.8619
24.	Hexadecanoic acid, ethyl ester	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	24.67	8.9910
25.	n-Hexadecanoic acid	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	256	25.31	36.5259
26.	4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester	C <sub>13</sub> H <sub>15</sub> NO <sub>3</sub>	233	26.47	2.0761
27.	Phytol	C <sub>20</sub> H <sub>40</sub>	296	27.57	2.9637
28.	9,12-Octadecadienoic acid, ethyl ester	C <sub>20</sub> H <sub>36</sub> O <sub>2</sub>	308	28.70	5.3488
29.	9-Octadecenoic acid, ethyl ester	C <sub>20</sub> H <sub>38</sub> O <sub>2</sub>	310	28.85	6.4691
30.	6-Octadecenoic acid, (Z)-	C <sub>18</sub> H <sub>34</sub> O <sub>2</sub>	282	29.50	9.6053
31.	Z-2-Octadecen-1-ol	C <sub>18</sub> H <sub>36</sub> O	268	34.63	1.0956

**Table 2: Biological Activities of Phytochemical Compounds Identified in Ethanol Extract of Whole Plant *Leptadenia Reticulata***

No.	Name of the compound	**Biological Activity
1.	2-Propanone, 1-(ethylthio)-	No Activity reported
2.	Glycerin	Flavor <sup>12</sup>
3.	1-Penten-4-one, 2-acetyl-1-dimethylamino-((Z)- or (E)-)	No Activity reported
4.	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	Antimicrobial, Anti-inflammatory, Antiproliferative Antioxidant, Automatic nerve activity <sup>13</sup>
5.	Ethyl hydrogen succinate	No Activity reported
6.	á-d-Allopyranoside, methyl 6-deoxy-2-O-methyl-	No Activity reported
7.	Benzenecarboxylic acid	Antifungal agents, Food preservatives <sup>14</sup>
8.	2-Furancarboxaldehyde, 5-(hydroxymethyl)-	Antimicrobial , Preservative <sup>12</sup>
9.	2-Methoxy-4-vinylphenol	Antioxidant, Antimicrobial, Anti inflammatory <sup>12</sup>
10.	Phenol, 2,6-dimethoxy-	No Activity reported
11.	Benzaldehyde, 3-hydroxy-4-methoxy-	No Activity reported
12.	Phenol, 2,4-bis(1,1-dimethylethyl)-	Antioxidant <sup>15</sup>
13.	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl	No Activity reported
14.	Dodecanoic acid	Antibacterial <sup>12</sup>
15.	3',5'-Dimethoxyacetophenone	No Activity reported
16.	3-Hydroxy-4-methoxybenzoic acid	No Activity reported
17.	Phenol, 2,6-dimethoxy-4-(2-propenyl)-	No Activity reported
18.	Tetradecanoic acid	Antioxidant, Cancer preventive, Nematicide, Lubricant Hypocholesterolemic <sup>12</sup>
19.	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	No Activity reported
20.	2,6,8-Trimethylbicyclo[4.2.0]oct-2-ene-1,8-diol	No Activity reported
21.	2H-Pyran, 6-heptyltetrahydro-2,2-dimethoxy-	No Activity reported
22.	Pentadecanoic acid	No Activity reported
23.	2,6-Di-O-methyl-d-galactopyranose	No Activity reported
24.	Hexadecanoic acid, ethyl ester	Lubricant, antiandrogenic, antioxidant, 5-alpha-reductase inhibitor. <sup>12</sup>
25.	n-Hexadecanoic acid	Antifungal, Antioxidant, hypocholesterolemic, nematicide, anti-androgenic flavour, haemolytic, 5-Alpha reductase inhibitor, potent antimicrobial agent, antimalarial and antifungal. <sup>16,17</sup>
26.	4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester	No Activity reported
27.	Phytol	Anti-microbial, anti-cancer, anti-Inflammatory. <sup>12</sup>
28.	9,12-Octadecadienoic acid, ethyl ester	No Activity reported
29.	9-Octadecenoic acid, ethyl ester	Nematicide, hepatoprotective, antihistaminic, anticoronary. <sup>12</sup>
30.	6-Octadecenoic acid, (Z)-	No Activity reported
31.	Z-2-Octadecen-1-ol	No Activity reported







(Fig A-V: The individual fragmentation pattern of the important compounds)

## Discussion

GC-MS chromatogram of the ethanol extract of whole plant of *Leptadenia reticulata* (Fig1) clearly showed thirty two peaks indicating the presence of thirty two phyto chemical compounds. The identification of the phyto chemical compounds was confirmed based on the peak area, retention time and molecular formula. The active principles with their retention time (RT), molecular formula (MF), molecular weight (MW) and peak area in percentage (%) were tabulated in Table 1. The components corresponding to the peaks were determined as follows

2-Propanone, 1-(ethylthio)- (0.98%), Glycerin (2.42%), 1-Penten-4-one, 2-acetyl-1-dimethylamino- ((Z)- or (E)-) (0.80%) , Ethyl hydrogen succinate (0.46%),  $\alpha$ -d-Allopyranoside, methyl 6-deoxy-2-O-methyl- (0.97%), Benzenecarboxylic acid (1.1%), 2-Furancarboxaldehyde, 5-(hydroxymethyl)- (3.67%), 2-Methoxy-4-vinylphenol (0.80%), Phenol, 2,6-dimethoxy- (0.53%), Benzaldehyde, 3-hydroxy-4-methoxy- (0.85%), Phenol, 2,4-bis(1,1-dimethylethyl)- (0.41%), 2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl- (0.69%), Dodecanoic acid (1.26%), 3',5'-Dimethoxyacetophenone (0.98%), 3-Hydroxy-4-methoxybenzoic acid (1.37%), Phenol, 2,6-dimethoxy-4-(2-propenyl)- (0.42%), Tetradecanoic acid (0.94%), 3,7,11,15-Tetramethyl-2-hexadecen-1-ol (2.84%), 2,6,8-Trimethylbicyclo[4.2.0]oct-2-ene-1,8-diol (0.11%), 2H-Pyran, 6-heptyltetrahydro-2,2-dimethoxy- (1.14%), Pentadecanoic acid (0.91%), 2,6-Di-O-methyl-d-galactopyranose (0.86%), Hexadecanoic acid, ethyl ester (8.99%), n-Hexadecanoic acid (36.5%), 4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester (2.07%), Phytol(2.96%), 9,12-Octadecadienoic acid, ethyl ester (5.34%), 9-Octadecenoic acid, ethyl ester (6.46%), 6-Octadecenoic acid, (Z)- (9.60%), Z-2-Octadecen-1-ol (1.09%). The spectrum sketch out of GC-MS confirmed the presence of thirty one components with the retention time 4.28, 8.70, 10.11, 11.17, 11.84, 12.02, 12.11, 12.75, 13.04, 14.05, 14.66, 15.88, 17.23, 17.82, 18.12, 18.22, 19.15, 20.31, 21.27, 21.27, 21.81, 22.02, 22.24, 22.80, 24.01, 24.67, 25.31, 26.47, 25.31, 26.47, 27.57, 28.70, 28.85, 29.50, 34.63 min respectively which is shown in Figure. 1. The individual fragmentation patterns of necessary components were illustrated in Figures A-V. The phytochemical compounds recognized through GC-MS analysis showed many biological activities are listed in Table 2.

## Conclusion

In the present study, thirty two phytochemical constituents have been identified from the ethanol extract of whole plant of *Leptadenia reticulata* by Gas Chromatogram - Mass Spectrometry (GC - MS) analysis. The presence of these phytochemical constituents proves the use of this plant for various diseases. Isolation of individual photochemical constituents and subjecting it to biological activities are being undertaken.

## Acknowledgement

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## References

1. D.,Takhi, M.Quinten and M.,Yousfi (2011).Study of Antimicrobial Activity of Secondary Metabolites extracted from spontaneous plants from the area of Laghouat, Algeria. *Advances in environmental Biology* ,5(2): 469-476.
2. Kirtikar KR, Basu BD (1995). *Indian Medicinal plants*. Vol 1, International book distributors, Deharadun, India,pp.830 -832.
3. Kadir MF, Bin Sayeed MS, Mia MM. Ethnopharmacological survey of medicinal plants used by traditional healers in Bangladesh for gastrointestinal disorders. *Journal of Ethnopharmacology* 2013; 147: 148–156.
4. Vinoth S, Rajesh Kanna P, Gurusaravanan P and Jayabalan N, Evaluation of phytochemical, antimicrobial and GC-MS analysis of extracts of *Indigofera trita* L.F. spp. *Subulata* (Vahl ex poir). *Int J Agric Res*. (2011); 6(4): 358-367.
5. Savithamma N, Venkateswarlu P, Suhurulatha D, Basha SKM and Venkataramanadevi CH, Studies of *Boswellia ovalifoliolata* Bal. and Herny – An endemic and endangered medicinal plant. *The Biosc.*, (2010); 5: 359-362.
6. Mukherjee PK, Kumar V, Houghton PJ. Screening of Indian medicinal plants for acetyl cholinesterase inhibitory activity. *Phytother Res* 2007; 21: 1142-5.
7. Chauhan , N S, Saraf DK, Dixit VK, (2010) : Effect of vajikaranrasayana herbs on pituitary gonadal axis , *Eur. J. Integr. Med* , 2010 ,2, 89 - 91 .
8. Dandiya pc, chopra ym. 1970. cns-active drugs from plants indigenous to india. *indian j.pharm.*, 2:67–90.
9. Bhatt T, Jain V, Jayathirtha MG, Banerjee G, Mishra SH. 2006. *In vitro* regeneration of roots of *Phylla nodiflora* and *Leptadenia reticulata*, and comparison of roots from cultured and natural plants for secondary metabolites. *Indian J. Exp. Biol.*, 40:1382–1386.
10. Vaidya BG, Nighantu A. 1965. Shri Swami Atmanand Saraswati Ayurved Sahakari Pharmacy Ltd, Surat, 2: p.1106.
11. Kalimuthu K. Prabakaran R. Preliminary phytochemical screening and GC - MS analysis of Methanol extract of *ceropogia pusilla*; *International Journal of Research in Applied, Natural and Social Sciences* 2013; 1 (3): 49-58
12. Dr. Duke's Phytochemical and Ethnobotanical Databases
13. [.http://pubchem.ncbi.nlm.nih.gov/rest/chemical/benzoic+acid](http://pubchem.ncbi.nlm.nih.gov/rest/chemical/benzoic+acid)
14. G.Rajeswari,M.Murugan & V.R.Mohan. GC-MS analysis of bioactive components of *Hugonia mystax* L. bark (Linaceae). *Journal of pharmaceutical and biomedical sciences (J Pha Biomed Sci.)* 2013, April; 29(29): 818-824. (Article no 17)
15. <http://www.chemicalland21.com/specialtychem/finechem/2,4-DI-ter BUTYLPHENOL.htm>
16. Hema, R., Kumaravel, S., and Alagusundaram. (2011). GC/MS Determination of Bioactive components of *Murraya koenigii*. *Journal of American Science*, 7(1).
17. Pietro, Z., Maurizio, S., Maurizio, B., Antonella, M., Sergio, R., Carmen, F. and Felice, S., (2010). Essential oil composition of stems and fruits of *Caralluma europaea* N.E.Br. (Apocynaceae). *Molecules*,15, 627-638.

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