

## **‘The Gc Ms Analysis Of Ethyl Acetate Extract Of One Herbal Plant, ‘Crotalaria Pallida’**

**Hassan Mohammad M<sup>1</sup>, Kanagasabai V<sup>2</sup>, Nandini M S<sup>3</sup>, Prabhu K<sup>4</sup>, Rao M R K<sup>5\*</sup>, Kalaivannan J<sup>6</sup>, Janaki CS<sup>7</sup>**

<sup>1</sup>Lecturer, Department of Anatomy, Faculty of Medicine, Northern Borders University, Arar, Saudi Arabia.

<sup>2</sup>Vice Chancellor, Bharath Institute of Higher Education And Research Bharath University, Chennai, India

<sup>3</sup>Assistant Professor, Department of Microbiology, SreeBalaji Medical College and Hospital, Chennai, Tamil Nadu, India

<sup>4</sup>Associate Professor, Department of Anatomy, SreeBalaji Medical College and Hospital, Chennai, Tamil Nadu, India

<sup>5</sup> Consultant Scientist, M/s. Noahs Laboratories, No, 8/1, Old Mahabalipuram Road, Thirupurur, Tamil Nadu 603110, India.

<sup>6</sup>Associate Professor, Department Of Anatomy, Vinayaka Mission’s Medical College And Hospital, Karikal, Vinayaka Mission’s Research Foundation, Salem, Tamil Nadu, India.

<sup>7</sup>Associate Professor, Department of Anatomy, Bhaarith Medical College, Chennai, Tamil Nadu, India

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### **ABSTRACT**

The present study deals with the GC MS analysis of one medicinal plant, ‘Crotalaria pallida’. Crotalaria pallida Aiton, (Family: Fabaceae) is a plant found on the road side and in the wild, is traditional medicine, used to treat urinary problems, swelling of joints and the juice of the leaves are used to eliminate intestinal worms. Known as “rattle or rattlesnake” is known for its anti-inflammatory, antimicrobial and antifungal roles. This plant was collected from nearby fields of Chengalpattu, Tamilnadu. The ethyl acetate extract of the aerial parts of the plant was subjected to GC MS study following standard protocols. It was observed that some very important molecules such as trans-2-methyl-4-n-pentylthiane, S,S-dioxide, 7-Octadecyne, 2-methyl-, n-Hexadecanoic acid, 4-(2,4-Dimethylcyclohex-3-enyl)but-3-en-2-one, 2-((Octan-2-yloxy)carbonyl)benzoic acid, Squalene, Sulfurous acid, butyl heptadecyl ester, 5-Nonadecen-1-ol, dl-.alpha.-Tocopherol, Campesterol, Stigmasterol, Lupeol etc. These molecules do represent the medicinal roles of this plant as anti-inflammatory, antimicrobial and other ethno-medicinal uses.

**KeyWord** GC MS, Crotalaria pallida, Squalene, Sulfurous acid, butyl heptadecyl ester, dl-.alpha.-Tocopherol, Campesterol, Stigmasterol, Lupeol

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## INTRODUCTION

*Crotalaria pallida* Aiton. (Fabaceae). It is a plant found on the road side and in the wild is traditional medicine, used to treat urinary problems, swelling of joints and the juice of the leaves are used to eliminate intestinal worms. (Kiruthiga et al, 2014). Known as “rattle or rattlesnake” is known for its anti-inflammatory, antimicrobial and antifungal roles. Lasker and Roy, 2016, have reported the phytochemical characterization of antimicrobial role of this plant. Paul, 2019 gave a comprehensive review on the various aspects of this plant in which he has indicated the this plant's role as antioxidant, anti-inflammatory, anthelmintic, anti-diabetic, anti-proliferative, cytotoxic, apoptotic and analgesic. Physicochemical characterization and antibacterial activity of the leaf oil of *Crotalaria pallida* Aiton. Roy, 2016 has reported this plant's antimicrobial activity. Boldrin et al, 2013 have reported the estrogenic and mutagenic activities of this plant measured by recombinant yeast assay and Ames test. The present work deals with the GC MS analysis of the ethyl acetate extract of the aerial parts of *Crotalaria pallida*. This work is in continuation of our work to establish the efficacy of the herbal plants, Ayurvedic and Siddha medicines. (Priyadarshini et al, 2017; Jayakumari et al, 2017; Rao et al, 2018; Vijayalakshmi and Rao, 2019; Yuvaraj et al, 2019; Muttevi et al, 2019, Rao et al, 2019; Muttevi et al, 2020; Vijayalakshmi and Rao, 2020; Janaki et al, 2021, Peruma et al, 2021).

## MATERIALS AND METHODS

The plant *Crotalaria pallida* was collected from the nearby fields at Chengalpattu, Tamil Nadu. The plant was identified by a qualified botanist at Chennai. The ethyl acetate extract of the shade dried leaves were collected after 48 h of soaking. The extract was evaporated and the dried powder was used for GC-MS analysis by standard procedures.

### GC-MS Procedure

Instrument: GC (Agilent: GC: (G3440A) 7890A. MS/MS: 7000 Triple Quad GCMS) was equipped with MS detector.

### Sample Preparation

About 100 ml sample was dissolved in 1 ml of suitable solvents. The solution was stirred vigorously using vortex stirrer for 10 s. The clear extract was determined using GC for analysis.

### GC-MS Protocol

Column DB5 MS (30 mm × 0.25 mm ID × 0.25 µm, composed of 5% phenyl 95% methylpolysiloxane), electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1 ml/min injector temperature 280°C; auxiliary temperature: 290°C ion-source temperature 280°C.

The oven temperature was programmed from 50°C (isothermal for 1.0 min), with an increase of 40°C/min, to 170°C C (isothermal for 4.0 min), then 10°C/min to 310°C (isothermal for 10 min) fragments from 45 to 450 Da. Total GC running time is 32.02 min. The compounds are identified by GC-MS Library (NIST and WILEY).

## RESULTS AND DISCUSSION

The results of the GC-MS analysis of the whole plant ethyl acetate extract, along with the possible medicinal role of each molecule of *Crotalaria pallida* extract are tabulated in Table 1. Figure 1 represents the GC-MS profile of ethyl acetate extract of the whole plant of *Crotalaria pallida*. The identification of metabolites as accomplished by comparison of retention time and fragmentation pattern with mass spectra in the NIST spectral library stored in the computer software (version 1.10 beta, Shimadzu) of the GC-MS along with the possible pharmaceutical roles of each bio molecule as per Dr. Duke's Phytochemical and ethno-botanical data base (National Agriculture Library, USA) and others as shown in Table 1. From the results it was observed that this plant contained some very important biomolecules such as trans-2-methyl-4-n-pentylthiane, S,S-dioxide, 7-Octadecyne, 2-methyl-, n-Hexadecanoic acid, 4-(2,4-Dimethylcyclohex-3-enyl)but-3-en-2-one, 2-((Octan-2-yloxy)carbonyl)benzoic acid, Squalene, Sulfurous acid, butyl heptadecyl ester, 5-Nonadecen-1-ol, dl-.alpha.-Tocopherol, Campesterol, Stigmasterol, Lupeol etc. These molecules do represent the medicinal roles of this plant as antiinflamantory, antimicrobial and other ethno-medicinal uses.

## CONCLUSION

Thus it can be concluded that due to the presence of these molecules, *Crotalaria pallida* has the medicinal roles for which it is used. Further work to isolate and understand the molecular mechanism is warranted.

## ACKNOWLEDGMENT

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Figure 1. Shows the GC MS profile graph of ethyl acetate extract of *Crotalaria pallida*

## Qualitative Compound Report

Data File 280121042.D Sample Name Crotalaria pallida  
 Sample Type Position 129  
 Acq Method GC Screening New Method.M Acquired Time 31-01-2021 AM05:05:24  
 Comment

User Chromatogram

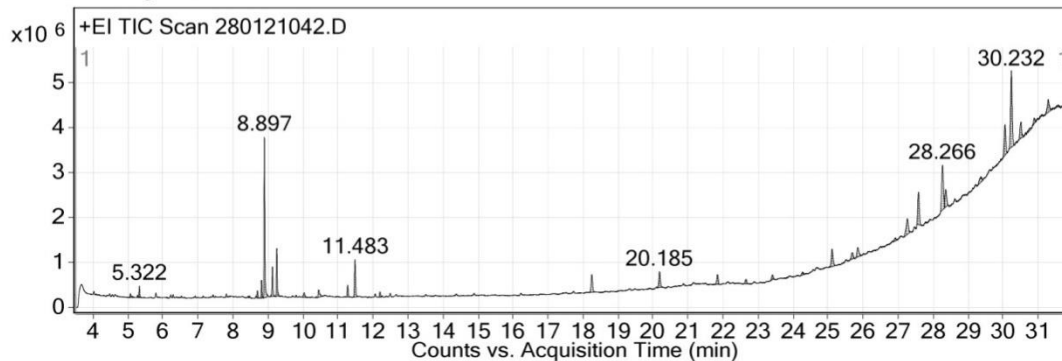


Table1. Indicates the retention time, types of possible compound, molecular formula, molecular mass, percentage peak area and the possible medicinal roles of each compound as shown in the GC MS profile of *Crotalaria pallida*

Ret. Time	Molecule	Mol. Formula	Mol. Mass	% Peak Value	Possible Medicinal Role
5.32	Naphthalene	C <sub>10</sub> H <sub>8</sub>	128.1	0.99	Not Known
8.81	trans-2-methyl-4-n-pentylthiane, S,S-dioxide	C <sub>11</sub> H <sub>22</sub> O <sub>2</sub> S	218.1	1.67	Glutathione-S-Transferase-Inhibitor, Catechol-O-Methyl-Transfearse inhibitor, Myo-neuro-stimulator, NitricOxideSynthase inhibitor, NO savaenger, Stimulates Morepinephrine production, Stimulates Sympathetic nervous system, decrease

					glutamate oxaloacetate transaminase, decrease glutame pyruvate transaminase, Glycosyltrabsferase inhibitor, inceresesglyoxalate transamination, reverse transcriptase inhibitor, smart drug, adrenal supporter
8.90	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl-	C10H18	138.1	14.96	Not known
9.25	7-Octadecyne, 2-methyl-	C19H36	264.3	4.45	Catechol o methyl Transferase inhibitor, methyl donar, methyl guanidine inhibitor
10.45	n-Hexadecanoic acid	C16H32O2	256.2	1.38	Acidifier, Arachidonic acid Inhibitor, Increases Aromatic Amino acid decarboxylase activity, Inhibits production of uric acid, Urine acidifier, Anaphylactic, Arylamine N acetyltransferase inhibitor, decreases norepinephrine production, Down regulates nuclear and

					cytosol androgen reuptake, GABA-nergic, Increase NK cell activity, inhibits production of tumor necrosis factor, Myo-neuro-stimulator
11.27	4-(2,4-Dimethylcyclohex-3-enyl)but-3-en-2-one	C <sub>12</sub> H <sub>18</sub> O	178.1	1.30	Decrease Endothelial Leukocyte Adhesion, Decrease Endothelial Platelet Adhesion, Encephalopathic, Endoanesthetic, Endocrinactive, Endorphinogenic, Endothelium-Dependent, Endothelium-Derived Relaxing Factor Promoter, Endrocrin-Tonic, Energizer
11.48	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1.alpha.,2.beta.,5.alpha.)-(./-.)-	C <sub>10</sub> H <sub>20</sub> O	156.2	4.42	Not Known
18.25	2-((Octan-2-ylloxy)carbonyl)benzoic acid	C <sub>16</sub> H <sub>22</sub> O <sub>4</sub>	278.2	3.07	Acidifier, Arachidonic acid Inhibitor, Increases Aromatic Amino acid



					decarboxylase activity, Inhibits production of uric acid, Urine acidifier
20.19	Squalene	C30H50	410.4	2.87	Plant steroid use as food additive and has cholesterol lowering role
21.84	Sulfurous acid, butyl heptadecyl ester	C21H44O3S	376.3	1.83	Acidifier, Arachidonic acid inhibitor, Increases Aromatic Amino acid Decarboxylase activity
25.12	5-Nonadecen-1-ol	C19H38O	282.3	3.74	Oligosaccharide provider
25.69	dl-.alpha.-Tocopherol	C29H50O2	430.4	1.27	Tocopherol synergist, 5 alpha reductase inhibitor, Alpha agonist, Alpha amylase inhibitor, Alpha glucosidase inhibitor, HIF-1 alpha inhibitor, Ikappa B- alpha phosphorylation inhibitor, Increase alpha mannosidase activity, Interleukin 1- alpha inhibitor, Testosterone-5- Alpha-Reductase-Inhibitor, TNF- alpha inhibitor

25.85	cis-1-Chloro-9-octadecene	C18H35Cl	286.2	2.12	Not known
27.26	Campesterol	C28H48O	400.4	4.25	Plant steroid use as food additive and has cholesterol lowering role
27.58	Stigmasterol	C29H48O	412.4	6.32	Precursor of progesterone , acts as intermediate in the biosynthesis of androgens and estrogens, anti-osteoarthritic, antihypercholesterolemic, cytotoxic, antitumor, hypoglycemic, antimutagenic, antioxidant, anti-inflammatory, analgesic
28.27	.beta.-Sitosterol	C29H50O	414.4	9.22	17 beta dehydrogenase inhibitor, androgen blocker, anti-amyloid beta, anticancer, Anti TGF beta, Beta 2-receptor, beta blocker, beta-galactosidase inhibitor, beta-glucuronidase inhibitor
28.36	7-Heptadecyne, 17-chloro-	C17H31Cl	270.2	3.63	Not known
29.36	Gitoxigenin	C23H34O <sub>5</sub>	390.2	1.54	Not known

30.05	1,1,6-trimethyl-3-methylene-2-(3,6,9,13-tetramethyl-6-ethenyl-10,14-dimethylene-pentadec-4-enyl)cyclohexane	C <sub>33</sub> H <sub>56</sub>	452.4	5.96	Not known
30.23	Lupeol	C <sub>30</sub> H <sub>50</sub> O	426.4	15.42	anti-inflammatory, antioxidant, anti-diabetic, and anti-mutagenic effects