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'The Gc Ms Analysis Of Ethyl Acetate Extract Of One Herbal Plant, 'Crotalaria Verrucosa'

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ABSTRACT

The present study deals with the GC MS analysis of one medicinal plant, *Crotalaria verrucosa*is common road side plant which is usedtreat scabies, jaundice, cough, biliousness, fever, cardiac problems and oral diseases.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 3,7,11,15-Tetramethyl-2-hexadecen-1-ol, n-Hexadecanoic acid, 3-Chloropropionic acid, tridec-2-ynyl ester, 2-((Octan-2-yloxy)carbonyl)benzoic acid, Squalene, (25R)-5.alpha.-Spirostan-2.alpha.,3.beta.-diol, Campesterol, Stigmasterol, .beta.-Sitosterol, .beta.-Amyrin, Lupeol, 7,8-Epoxylanostan-11-ol, 3-acetoxy-, i-Propyl 9,12,15-octadecatrienoate were observed in the GC MS profile. These molecules have many important medicinal which augur well with the medicinal role of *Crotalaria verrucosa* as such. Further study, however is requiredto isolate each compound and understand their molecular roles.

Key Words GC MS, *Crotalaria verrucosa*, Campesterol, Stigmasterol, .beta.-Sitosterol, .beta.-Amyrin, Lupeol, 7,8-Epoxylanostan-11-ol, 3-acetoxy-, i-Propyl 9,12,15-octadecatrienoate

INTRODUCTION

Crotalaria verrucosa is common road side plant which is used treat scabies, jaundice, cough, biliousness, fever, cardiac problems and oral diseases, ethno-medicinally. Only very few reports on its medicinal roles are available. Crotalaria verrocosa leaf extracts were found to suppress inflammatory mediators by Billahet al, 2020 and Narwinet al, 2016. The hepato-protective, antifertility and efficacy against estrogenic implantation is also reported. (Narwin et al, 2016). Ahmed et al, 2018 have reported the antioxidant and cytotoxic roles of leaf extract of Crotalaria verrucosa. Manokari and Shekhawat, 2016 have synthesized Zinc nanoparticles from the leaf extracts of this plant. The present works deals with the GC MS analysis of the ethyl acetate extract of the aerial parts of Crotalaria verrucosa. This work is in continuation of our work to establish the efficacy of the herbal plants, Ayurvedic and Sidhha medicines. (Priyadarshiniet al, 2017; Jayakumariet al, 2017; Raoet al, 2018; Vijayalakshmi and Rao, 2019; Yuvarajet al, 2019; Mutteviet al, 2019, Raoet al, 2019; Mutteviet al, 2020; Vijayalakshmi and Rao, 2020; Janakiet al, 2021, Perumalet al, 2021).

MATERIALS AND METHODS

The plant Crotalaria verrucosawas 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; auxilary 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 ofCrotalaria verrucosaextract are tabulated in Table 1. Figure 1 represents the GC-MS profile of ethyl acetate extract of whole plant of Crotalaria verrucosa. 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 as3,7,11,15-Tetramethyl-2-hexadecen-1-ol, n-Hexadecanoic acid, 3-Chloropropionic acid, tridec-2-ynyl ester, 2-((Octan-2-yloxy)carbonyl)benzoic acid, Squalene, (25R)-5.alpha.-Spirostan-2.alpha.,3.beta.-diol, Campesterol, Stigmasterol, .beta.-Sitosterol, .beta.-Amyrin, Lupeol, 7,8-Epoxylanostan-11-ol, 3-acetoxy-, i-Propyl 9,12,15-octadecatrienoate were observed in the GC MS profile. These molecules have many important medicinal which augur well with the medicinal role of Crotalaria verrucosa as such. Further study, however is required to isolate each compound and understand their molecular roles. Further research is warranted on this unexplored plant.

CONCLUSION

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

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

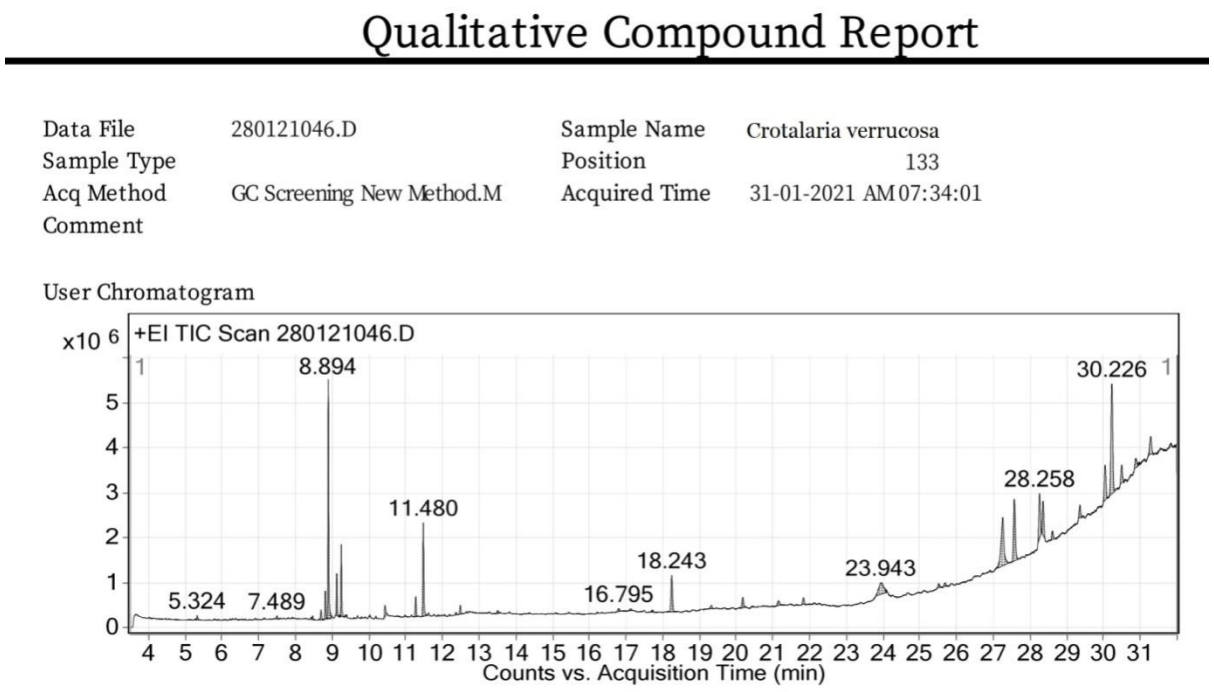

Table1. Indicates the retention time, types of possible compound, molecular formula, molecular mass, percentage peak area and the possible medicinal roles of each compound as shown in the GC MS profile of Crotalaria verrucosa.

Ret. Time	Molecules	Mol. formula	Mol. Mass	% Peak Area	Possible Medicinal Role
8.81	1,13-Tetradecadien-3-one	C14H24O	208.2	1.68	Not known
8.89	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl-	C10H18	138.1	14.73	Not known
9.12	9-Octadecyne	C18H34	250.3	2.38	Not known
9.25	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	C20H40O	296.3	4.12	Oligosaccharide provider
10.44	n-Hexadecanoic acid	C16H32O2	256.2	0.91	Acidifier, Acidulant, Arachidonic acid-Inhibitor, Increase Aromatic Amino Acid Decarboxylase Activity, Inhibit Production of Uric Acid, Anaphylactic, Antitumor, Arylamine-N-Acetyltransferase-Inhibitor, Decrease Norepinephrine Production, Down regulation of nuclear and cytosol androgen reuptake, GABA-nergic, Increase Natural Killer (NK) Cell Activity, Inhibit Production of Tumor Necrosis Factor, Myo-neuro-stimulant
11.27	3-Chloropropionic acid, tridec-2-ynyl ester	C16H27ClO2	286.2	1.50	Acidifier, Acidulant, Arachidonic acid-Inhibitor, Increase Aromatic Amino Acid Decarboxylase Activity, Inhibit Production of Uric Acid

11.48	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1.alpha.,2.beta.,5.alpha.)-(.+/-.)-	C10H20O	156.2	7.12	Not known
18.24	2-((Octan-2-yloxy)carbonyl)benzoic acid	C16H22O4	278.2	4.02	Acidifier, Acidulant, Arachidonic acid-Inhibitor, Increase Aromatic Amino Acid Decarboxylase Activity, Inhibit Production of Uric Acid
20.18	Squalene	C30H50	410.4	1.27	Plant steroid use as food additive and has cholesterol lowering role
23.94	(25R)-5.alpha.-Spirostan-2.alpha.,3.beta.-diol	C27H44O4	432.3	4.30	Aldose-reductase inhibitor, Anfiotensin receptor blocker Anti X radiation, Arginine rich, benzodiazepine receptor, agonist, Boron rich, carotenoid rich, decreases lactate/pyruvate ratio, dopamine receptor inhibitor, down regulates nuclear and cytosol androgen reuptake, Endothelium derived relaxing factor promoter, fertility regulator, fibre rich, free radical scavenger,
27.26	Campesterol	C28H48O	400.4	10.90	Plant steroid use as food additive and has cholesterol lowering role

27.57	Stigmasterol	C29H48O	412.4	8.93	Precursor of progesterone , acts as intermediate in the biosynthesis of androgens and estrogens, anti-osteoarthritic, antihypercholesterolemic, cytotoxic, antitumor, hypoglycemic, antimutagenic, antioxidant, anti-inflammatory, analgesic
28.26	.beta.-Sitosterol	C29H50O	414.4	5.01	17 beta dehydrogenase inhibitor, androgen blocker, anti-amyloid beta, anticancer, Anti TGF beta, Beta 2- receptor, beta blocker, beta-galactosidase inhibitor, beta-glucuronidase inhibitor
28.35	3-Eicosyne	C20H38	278.3	5.95	Not known
28.62	Phytonadione	C31H46O2	450.4	0.97	Not known
29.36	.beta.-Amyrin	C30H50O	426.4	1.39	17 beta hydroxysteroid dehydrogenase inhibitor, Antiamyloid beta, Anti TGF beta, Beta receptor agonist, Beta adrenergic receptor blocker, beta blocker, beta galactosidase inhibitor, beta glucuronidase inhibitor, ER beta binder
30.23	Lupeol	C30H50O	426.4	15.12	anti-inflammatory, antioxidant, anti-diabetic, and anti-mutagenic effects
30.88	7,8-Epoxylanostan-11-ol, 3-acetoxy-	C32H54O4	502.4	1.33	Oligosaccharide provider
31.28	i-Propyl 9,12,15-octadecatrienoate	C21H36O2	320.3	2.49	11Beta HSD inhibitor, 17-beta-hydroxysteroid dehydrogenase

					inhibitor, 5 HETE inhibitor, 5 HT inhibitor, 8 HETE inhibitor, Anti 5-HT, Anti HIV integrase, Aryl hydrocarbon hydroxylase inhibitor, HDL genic, Hematopoietic, 6 lipoxygenase inhibitor, aldehyde oxidase inhibitor, alcohol dehydrogenase inhibitor, AHH inhibitor, Adenylatecyclase inhibitor, Ache inhibitor, Acetyl Co ACarboxylase inhibitor, A beta inhibitor