

'The Gc Ms Analysis Of Ethyl Acetate Extract Of One Herbal Plant, 'Ziziphus Rugosa'

Hassan Mohammad M¹, Sharmila D², Nandini M S³, Prabhu K⁴, Rao M R K^{5*}, Kalaivannan J⁶, Janaki CS⁷

¹Lecturer, Department of Anatomy, Faculty of Medicine, Northern Borders University, Arar, Saudi Arabia.

²Sharmila D, Assist. Professor, Dept. Of Industrial Biotechnology, Bharath Institute of Higher Education and Research, Chennai, India

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

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

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

⁶Associate Professor, Department Of Anatomy, Vinayaka Mission's Medical College And Hospital, Karikal, Vinayaka Mission's Research Foundation, Salem, Tamil Nadu, India.

⁷Associate Professor, Department of Anatomy, Bhaarath Medical College, Chennai, Tamil Nadu, India

ABSTRACT

The present study deals with the GC MS analysis of one medicinal plant, Ziziphusrugosa. The bark of Ziziphusrugosa is used to brew alcohol and a paste from it is used to cure gastric problems. The dry leaves are used to make traditional cheroots. Seeds and fruits are edible. This plant has not been much explored for its medicinal values. The plant was collected from nearby hills 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 n-Hexadecanoic acid, Octadecanoic acid, 17-methyl-, methyl ester, Octadecanoic acid, Methyl 9,10-octadecadienoate, Methyl 9,10-octadecadienoate, 3,7,11,15-Tetramethyl-2-hexadecen-1-ol, 2-((Octan-2-yloxy)carbonyl)benzoic acid, Campesterol, Stigmasterol, .beta.-Sitosterol, .beta.-Amyrin, .beta.-Amyrintrimethylsilyl ether, Lupeol, Benzoic acid, octadecyl ester were shown in the GC MS profile of the ethyl acetate leaf extracts of this plant. These molecules could throw some light on the possible medicinal roles of this plant. Further work In this regard is required.

KeyWords GC MS, Ziziphusrugosa, n-Hexadecanoic acid, Campesterol, Stigmasterol, .beta.-Sitosterol, .beta.-Amyrin, .beta.-Amyrintrimethylsilyl ether, Lupeol, Benzoic acid,

INTRODUCTION

The bark of *Ziziphus rugosa* is used to brew alcohol and a paste from it is used to cure gastric problems. The dry leaves are used to make traditional cheroots. Seeds and fruits are edible. There are not many scientific reports about this plant. Premaet al, 2018 have studied the antimicrobial activity of the pericarp of *Z. rugosa*. Prashithet al, 2011 have reported the antioxidant role of the methanolic extract of this plant. Krishnamurthy and Sarala have shown the nutritive value of the fruit of *Z. rugosa*. The present work reports the GC MS pattern of the ethyl acetate extracts of *Ziziphus rugosa* leaves. This is in continuation of our endeavour to establish the medicinal efficacy of the herbal and traditional systems of Ayurveda, Sidhha and Unani systems of medicine (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 *Ziziphus rugosa* was collected from the nearby hills 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 *Ziziphus rugosa* extract are tabulated in Table 1. Figure 1 represents the GC-MS profile of ethyl acetate extract of the whole plant of *Ziziphus rugosa*. The identification of metabolites was 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 ethnobotanical data base (National Agriculture Library, USA) and others as shown in Table 1. From the details as shown in Table 1, this plant leaf contain some highly medicinal value compounds such as n-Hexadecanoic acid, Octadecanoic acid, 17-methyl-, methyl ester, Octadecanoic acid, Methyl 9,10-octadecadienoate, Methyl 9,10-octadecadienoate, 3,7,11,15-Tetramethyl-2-hexadecen-1-ol, 2-((Octan-2-yloxy)carbonyl)benzoic acid, Campesterol, Stigmasterol, .beta.-Sitosterol, .beta.-Amyrin, .beta.-Amyrintrimethylsilyl ether, Lupeol, Benzoic acid, octadecyl ester. Further work to establish the relationship of these molecules and finding out the possible medicinal uses of this plant is going on. It is important since this plant could be used as an effective medicine once its medicinal roles as established.

CONCLUSION

It is concluded that the types of medicinal values the molecules have, as is shown in the Table, *Ziziphus rugosa* can prove itself as an important medicinal plant.

ACKNOWLEDGMENTS

The authors express their sincere thanks to all who have helped in this project.

REFERENCES

1. Prema, G., Chitra, M., Kanagasabai. R. (2018) Studies on antimicrobial activity of *Ziziphus rugosa* lam. pericarp. *IJPSR*, 10(11), 4942-4948
2. Prashith, K. T. R., Vinayaka, K. S., Mallikarjun, N., et al. (2011) Antibacterial, Insecticidal and Free radical scavenging activity of methanol extract of *Ziziphus rugosa* Lam. (Rhamnaceae) fruit pericarp. *Pharmacognosy J*, 2(18), 65-68
3. Krishnamurthy, S. R., Sarala, P. (2012) Determination of nutritive value of *Ziziphus rugosa* L., A famine edible fruit and medicinal plant of Western ghats. *Ind J of Nat Prod and Reso*, 3(1), 20-27.
4. Vinayaka, K. S., Ravindra, P., Kumaraswamy, H. G. Nutritive composition of fruit of *Ziziphus rugosa* Lam. *Research and Reviews in Biomedicine and Biotechnology*, 2(1&2), 20-24

5. MutteviHyagreva Kumar, Prabhu K, Mudiganti Ram Krishna Rao, Lakshmisundram R, SampadShil, Sathish Kumar M, Vijayalakshmi N. The GC MS study of one medicinal plant, Aristolochia indica .DIT, 2020; 12(12): 2919-2923.
6. GomathiPriyadarshini, Arul Amutha Elizabeth, Jacintha Anthony, Mudiganti Ram Krishna Rao, Prabhu. K., Aiswarya Ramesh, Vani Krishna. (2017) The GC MS analysis of one medicinal plant, Premnatomentosa. Journal of Pharmaceutical Sciences and Research, 9(9), 1595-1597
7. Jayakumari, S., Prabhu, K., Mudiganti Ram Krishna Rao, Bhupesh, G., Kumaran, D., Aishwariya Ramesh. (2017) The GC MS Analysis of a Rare Medicinal Plant Aloe barbadensis. J. Pharm. Sci. & Res.9(7), 1035-1037
8. Rao, M. R. K., Vijayalakshmi, N. (2018) Preliminary phytochemical and GC MS analysis of different extracts of Sphaeranthus indicus leaves. Indo American J of Pharmaceuical Sciences, 5(3), 1511-1520
9. Vijayalakshmi, N., Mudiganti Ram Krishna Rao. (2019) The antioxidant studies of two medicinal plants, Sphaeranthus indicus and Psophocarpus tetragonolobus. Asian J of pharmaceutical and Clinical Res, 12(1), 321-327.
10. Yuvaraj, R., Mudiganti Ram Krishna Rao, Prabhu, K., Lakshmisundram, R., SampadShil, Sathish Kumar, M., Vijayalakshmi, N. (2019) The GC MS study of one medicinal plant, Stachytarpheta indica. Drug Invention Today, 12(9), 1665-1669
11. MutteviHyagreva Kumar, Prabhu, K., Mudiganti Ram Krishna Rao, Lakshmisundram, R., SampadShil, Sathish Kumar, M., Vijayalakshmi, N. (2019) The GC MS study of one medicinal plant, Dodonea angustifolia. Drug Invention Today, 12(9), 1661-1664
12. Mudiganti Ram Krishna Rao, Vijayalakshmi, N., Prabhu, K., Sathish Kumar, M. (2019) The gas chromatography–mass spectrometry study of Moringa oleifera seeds. DIT, 12(10), 2172-2175
13. MutteviHyagreva Kumar, Prabhu, K., Mudiganti Ram Krishna Rao, Lakshmisundram, R., SampadShil, Sathish Kumar, M., Vijayalakshmi, N. (2020) The GC MS study of one medicinal plant, Aristolochia indica .DIT, 12(12), 2919-2923.

14. Vijayalakshmi, N., Mudiganti Ram Krishna Rao. (2020) 'Preliminary phytochemical and antioxidant studies of leaf extracts of one medicinal plant, Vitexnegundo".RJPT, 13(5), 2167-2173

15. Janaki C. S., Prabhu K., Mudiganti Ram Krishna Rao, VenkatRamaiah, ShrutiDinkar, Vijayalakshmi, N., Kalaivannan. J. (2021) The GC MS analysis of Ethyl acetate extract of Merremiaemerginata'.Ind J of Natural Sciences, 12(67), 33638-33646

16. Perumal, G. M., Prabhu, K., Rao, M. R. K., Janaki, C. S., Kalaivannan, J., Kavimani, M. (2021) The GC MS analysis of Ethyl acetate extract of 'Flueggealeucopyrus. Nat. Volatiles & Essential Oils, 8(5), 4035-4040

17. Dr.Duke's Phytochemical and Ethnobotanical Databases.U.S. Department of Agriculture, Agricultural Research Service.1992-2016. Dr. Duke's Phytochemical and Ethnobotanical Databases. Home Page, <http://phytochem.nal.usda.gov/> <http://dx.doi.org/10.15482/USDA.ADC/1239279>

Qualitative Compound Report

Data File	280121028.D	Sample Name	Ziziphus rugosa
Sample Type		Position	115
Acq Method	GC Screening New Method.M	Acquired Time	30-01-2021 PM08:24:06
Comment			

User Chromatogram



Figure 1. Represents the GC MS graph of ethyl acetate extract Ziziphusrugosa'.

Table 1. Indicates the retentions 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 Ziziphusrugosa'.

Ret. Time	Molecule	Mol. Formula	Mol. Mass	% Peak Area	Possible Medicinal Role
10.49	n-Hexadecanoic acid	C16H32O2	256.2	8.68	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
11.51	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1.alpha.,2.beta.,5.alpha.)-(+/-)-	C10H20O	156.2	4.78	Not known
12.23	Octadecanoic acid, 17-methyl-, methyl ester	C20H40O2	312.3	0.69	Catechol-O-methyl-Transferase Inhibitor, methyl Donar, Methyl Guanidine Inhibitor, Acidifier, Arachidonic acid inhibitor, Increases Aromatic Amino acid Decarboxylase

					activity
12.29	Octadecanoic acid	C18H36O2	284.3	5.17	Acidifier, Arachidonic acid Inhibitor, Increases Aromatic Amino acid decarboxylase activity, Inhibits production of uric acid, Urine acidifier
12.39	Methyl 9,10-octadecadienoate	C19H34O2	294.3	0.87	Catechol-O-methyl-Transferase Inhibitor, methyl Donar, Methyl Guanidine Inhibitor
12.52	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	C20H40O	296.3	3.81	Oligosaccharide provider
18.28	2-((Octan-2-yloxy)carbonyl)benzoic acid	C16H22O4	278.2	4.18	Acidifier, Arachidonic acid Inhibitor, Increases Aromatic Amino acid decarboxylase activity, Inhibits production of uric acid, Urine acidifier
27.30	Campesterol	C28H48O	400.4	0.47	Plant steroid use as food additive and has cholesterol lowering role
27.63	Stigmasterol	C29H48O	412.4	4.26	Precursor of progesterone , acts as intermediate in the biosynthesis of androgens and estrogens, anti-osteoarthritic, antihypercholesterolemic, cytotoxic, antitumor, hypoglycemic, antimutagenic, antioxidant, anti-inflammatory, analgesic
28.31	.beta.-Sitosterol	C29H50O	414.4	3.88	17 beta dehydrogenase inhibitor, androgen blocker, anti-amyloid beta, anticancer, Anti TGF beta, Beta 2-receptor, beta blocker, beta-galactosidase inhibitor, beta-glucuronidase inhibitor
29.41	.beta.-Amyrin	C30H50O	426.4	8.88	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
30.31	Lupeol	C30H50O	426.4	46.17	anti-inflammatory, antioxidant, anti-diabetic, and anti-mutagenic effects
30.79	Lup-20(29)-en-3-ol, acetate, (3.beta.)-	C32H52O2	468.4	1.00	Not Known
31.02	Benzoic acid, octadecyl ester	C25H42O2	374.3	0.85	Acidifier, Arachidonic acid Inhibitor, Increases Aromatic Amino acid decarboxylase activity, Inhibits production of uric acid, Urine acidifier

17-beta-hydroxysteroid dehydrogenase inhibitor, Antiamyloid-Beta, Anti TGF-Beta, Beta-2-Receptor-Agonist, Beta-Adrenergic receptor blocker, Beta Galactosidase inhibitor, Beta-Glucuronidase inhibitor, Aldehyde oxidase inhibitor