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GC-MS Analysis of Chemical Substances from Ethanolic Extract of *Prosopis juliflora* Leaves

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ABSTRACT

The nutraceuticals in leaves are among the major contributors of their therapeutic relevance to human. This study evaluates the relevance of nutraceuticals obtained from the GC-MS analyses of ethanolic extracts of leaves of *Prosopis juliflora*. Leaves of *Prosopis juliflora* harvested from their natural territory and identified at the Plant Science Department of Mohamed Sathak A.J. College of Pharmacy, Sholinganallur, Chennai. Ethanol was used for extracting the crushed plant leaves (5 g) in order. Hot continuous soxhlet extraction was used to complete the extraction. The resulting extracts were kept at -4 °C until their next usage. The current investigation made use of ethanol extract. In the present investigation, the phytochemicals in the extracted substances were identified using the GC-MS method. Results shows 72 phytoconstituents in leaves of *Prosopis juliflora*, with gamma-Sitosterol, Epilupeol, Benzene methanol, 2-hydroxy-5-methyl-, as highest nutraceutical components with percentage concentrations of 12.94, 11.92 and 10.07 respectively. In order to improve human health, broad-spectrum pharmaceutical formulations are being developed that take advantage of the multiple therapeutic potentials of the main nutraceuticals found in *Prosopis juliflora*.

Keywords: Nutraceuticals, Ethanolic extract, Pharmaceuticals, Phytoconstituents, *Prosopis juliflora*.

INTRODUCTION

Prosopis juliflora, also referred to as Velayati babul, is a highly common shrub in the Fabaceae family. Its light green, geminate-pinnate, deciduous leaves are with 12 to 20 leaflets per leaf. According to various sources, *Prosopis juliflora* has a number of conventional pharmaceutical uses, notably the treatment of wounds and soothing the symptoms of measles, diarrhoea, inflammation, hoarseness, colds, dysentery, and sore throats. Some of the easily obtainable phytoconstituents in the plant include flavonoids, alkaloids, and saponins [1,2].

Since herbal remedies and associated products are frequently made from raw plant-based extracts, which include a complicated mixture of various phytochemical ingredients, plants with medicinal properties are of significant interest to the pharmaceutical industry. It has been claimed that natural substances taken from plants, especially higher plants, could serve as an alternate supply of antibiotics, antipustul, antitumor, antipyretic, anti-emetic [3]. The chemical characteristics of these components vary greatly between species. This strategy is appealing in part because they represent a possible source of bioactive substances that have been deemed relatively safe by the general public and frequently act at many and novel target locations, minimizing the likelihood of resistance [4].

Prior analysing components, the primary step in recovering and isolating phytochemicals that are bioactive from plant sources is extraction [5]. Therefore, it is necessary to identify the active components in medicinal herbs in order to search for potential compounds for usage as therapeutic medications [6]. The GC-MS method is a useful tool for determining the concentration of certain of the active ingredients in plant parts.

It integrates two methods of analysis into one approach for analysing chemical compound combinations. The components of the mixture are separated using gas chromatography, alongside every one of them is then individually analysed using mass spectroscopy.

Multiple investigations into *Prosopis juliflora* revealed a range of fascinating biological activities [7-9]. Therefore, the objective of the current investigation was to conduct out the gas chromatography and mass spectrometry (GC-MS) detection of bioactive substances from the ethanol extract of *Prosopis juliflora* leaves [10].

MATERIAL & METHOD

Collection of Plant materials:

From India's southern countryside, leaf samples of *Prosopis juliflora* were gathered between December, 2022 and January, 2023. Department of Botany, Mohamed Sathak A.J. College of Pharmacy, Sholinganallur, Chennai, further identified and verified the botanical specimens. The leaves were properly cleaned with distilled water & tap water, dried out in the shade for seven days, and then crushed into a smooth powder. These were placed in sealed air-tight zipper bags, labelled and preserved until further usage following sieving (80 mesh).

Preparation of Plant extracts

Ethanol was used as solvent to extract the powdered plant leaves (5 g). Hot continuous soxhlet extraction was employed to complete the extraction. The extracts were kept at -4 °C until their next usage. The present investigation used ethanol as solvent for extraction.

Gas chromatography and Mass spectrometry of Extracts:

The current investigation utilised the GC-MS technique for identifying the phytochemicals in the extracts. The Shimadzu Mass Spectrometer-2010 series was used to conduct a GC-MS analysis of the tested extracts. A sample volume of 1 microlitre was injected into a gas chromatograph–mass spectrometer (GC–MS) with an integrated injector, a PE Auto system XL gas chromatograph, and a turbo–mass spectrometric mass selective detection system.

In the EI mode (70 eV), the MS was used. As the carrier gas, helium was used, and its flow rate had been adjusted to 1.2 ml/min. The system's analytical column was a Rtx-5 capillary column with a length of 60 m with an inner diameter of 0.25 mm and a film with a thickness of 0.25 μm. To 196.6 kPa, the column head pressure was changed.

The temperature of the column was programmed to rise from 100°C (2 minutes) to 200°C at 10°C/min and from 200°C to 300°C at 15°C/min with hold times of 5 and 22 minutes, respectively. A six-minute solvent delay was chosen. The temperature of the injector was fixed at 260 °C. At 280 °C, the GC-MS interface was kept constant. The MS was run in ACQ mode, scanning between m/z 40 and 600.0. At an electron energy of 70 eV, full scan mode electron ionisation (EI) mass spectra in the 40–600 (m/z) range were captured.

RESULT & DISCUSSION

GC-MS Analysis

Advancement, modernisation, and quality assurance of herbal medicines depend heavily on the evaluation and separation of plant material. Therefore, the goal of the current work was to identify the bioactive substances in the ethanol extract of *Prosopis juliflora* using gas chromatography and mass spectrometry. There are 72 bioactive phytochemicals found in the ethanol extract of *Prosopis juliflora* leaves, are illustrated in Table 1 and Fig. 1 together with their peak height, concentration (peak area %), and retention time (RT).

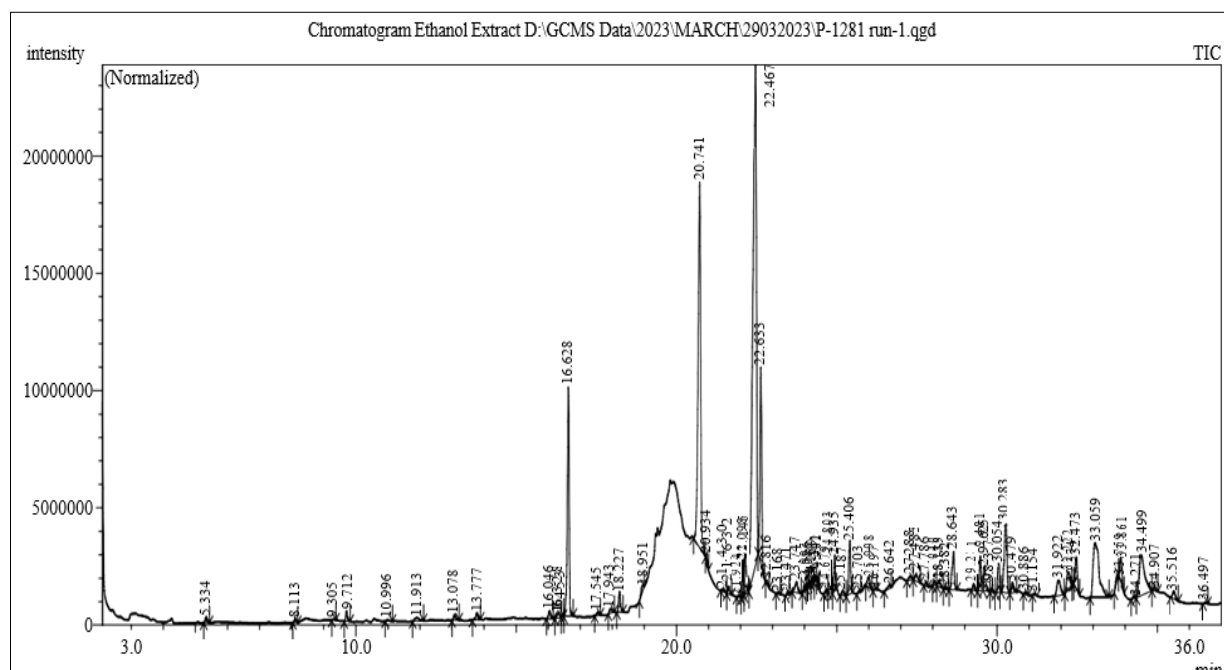


Figure 1: GC-MS Chromatogram of Ethanol extract of *Prosopis juliflora* leaves

Table 1: GC-MS properties of compound identified from Ethanol extract of *Prosopis juliflora* leaves:

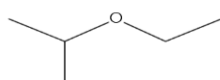
Peak No.	Retention Time	Area	Area%	Height	Height %	A/H	Name
1	5.334	911598	0.19	240164	0.27	3.80	Propane, 2-ethoxy-
2	8.113	639168	0.13	183163	0.20	3.49	4-Ethyl-5-hydroxy-3,5-dimethylfuran-2-one
3	9.305	153986	0.03	45106	0.05	3.41	Propane, 1,1,3-triethoxy-

4	9.712	1595875	0.33	461085	0.51	3.46	Propane, 1,1-diethoxy-2-methyl-
5	10.996	297109	0.06	59932	0.07	4.96	Octanoic acid
6	11.913	1419947	0.29	140941	0.16	10.07	Benzene methanol, 2-hydroxy-5-methyl-
7	13.078	1175591	0.24	246866	0.27	4.76	2-Methoxy-4-vinylphenol
8	13.777	1372135	0.28	296663	0.33	4.63	n-Decanoic acid
9	16.046	1494339	0.31	348773	0.39	4.28	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4
10	16.328	1229392	0.25	212548	0.24	5.78	Phenol, 4-ethenyl-2,6-dimethoxy-
11	16.453	78697	0.02	28209	0.03	2.79	Fumaric acid, ethyl 2-methylallyl ester
12	16.628	37095738	7.58	9775618	10.83	3.79	Diethyl Phthalate
13	17.545	595673	0.12	133023	0.15	4.48	4-(6,6-Dimethyl-2-methylenecyclohex-3-enyli
14	17.943	933846	0.19	177596	0.20	5.26	2(3H)-Benzothiazolone
15	18.227	3385028	0.69	873733	0.97	3.87	Naphthalene, 1,6-dimethyl-4-(1-methylethyl)-
16	18.951	1415042	0.29	233315	0.26	6.06	Loliolide
17	20.741	90489627	18.49	15622503	17.30	5.79	l-(+)-Ascorbic acid 2,6-dihexadecanoate
18	20.934	740756	0.15	281140	0.31	2.63	Ethyl 13-methyl-tetradecanoate
19	21.430	532471	0.11	96322	0.11	5.53	7-Tetradecenal, (Z)-
20	21.632	1200673	0.25	337353	0.37	3.56	Eicosanoic acid
21	21.924	330961	0.07	59271	0.07	5.58	cis-1-Chloro-9-octadecene
22	22.098	4723952	0.97	1543601	1.71	3.06	1-Dodecanol, 3,7,11-trimethyl-
23	22.145	6585217	1.35	1627467	1.80	4.05	Phytol
24	22.467	143646237	29.35	21357931	23.66	6.73	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
25	22.633	31630508	6.46	8368972	9.27	3.78	Octadecanoic acid
26	22.816	419878	0.09	142054	0.16	2.96	Heptadecanoic acid, 15-methyl-, ethyl ester
27	23.168	209998	0.04	59452	0.07	3.53	2-Furanmethanamine, tetrahydro-N-[(tetrahyd
28	23.471	478804	0.10	142807	0.16	3.35	l-(+)-Ascorbic acid 2,6-dihexadecanoate
29	23.747	2883491	0.59	467853	0.52	6.16	2-(Dimethylamino)ethyl (9Z,12Z)-octadeca-9,
30	23.997	735286	0.15	215328	0.24	3.41	Dichloroacetic acid, tridec-2-ynyl ester
31	24.086	111519	0.02	43530	0.05	2.56	Arachidonic acid
32	24.167	122953	0.03	43775	0.05	2.81	Cyclohexanebutanal, 2-methyl-3-oxo-, cis-
33	24.239	275500	0.06	98013	0.11	2.81	4,8,12,16-Tetramethylheptadecan-4-olide
34	24.341	376902	0.08	136626	0.15	2.76	Eicosanoic acid
35	24.402	631262	0.13	221897	0.25	2.84	1,3-Dioctanoin
36	24.679	627323	0.13	136064	0.15	4.61	10,12-Tricosadiynoic acid
37	24.803	5397912	1.10	1619298	1.79	3.33	Phenol, 2,4-bis(1-phenylethyl)-
38	24.935	4885933	1.00	1540550	1.71	3.17	Phenol, 2,4-bis(1-phenylethyl)-
39	25.187	964031	0.20	208100	0.23	4.63	{2-[(3-Bromophenyl)amino]ethyl}dimethylam
40	25.406	8698715	1.78	2288371	2.53	3.80	Phenol, 2,4-bis(1-phenylethyl)-
41	25.703	339813	0.07	103373	0.11	3.29	Bis(2-ethylhexyl) phthalate
42	25.998	2318374	0.47	450915	0.50	5.14	Decanoic acid, 2-hydroxy-3-[(1-oxooctyl)oxy]
43	26.197	231778	0.05	69941	0.08	3.31	Dotriacontane, 1-iodo-
44	26.642	959298	0.20	152105	0.17	6.31	Cholest-22-ene-21-ol, 3,5-dehydro-6-methoxy
45	27.288	1032885	0.21	163204	0.18	6.33	Hexadecanoic acid, 2-[(1-oxododecyl)oxy]-1,
46	27.483	1857820	0.38	293257	0.32	6.34	1-Decanoyl-3-dodecanoylglycerol
47	27.786	633596	0.13	194552	0.22	3.26	Squalene
48	28.040	778023	0.16	208944	0.23	3.72	.alpha.-Tocospiro B
49	28.189	1243152	0.25	266391	0.30	4.67	.alpha.-Tocospiro B

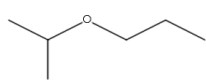
50	28.382	332567	0.07	66664	0.07	4.99	benzothiazole, 2,2'-thiobis-
51	28.643	8070302	1.65	1631123	1.81	4.95	Glycerol tricaprylate
52	29.271	1061523	0.22	279119	0.31	3.80	Cholesta-4,6-dien-3-ol, (3.beta.)-
53	29.481	5669785	1.16	1481547	1.64	3.83	Phenol, 2,4,6-tris(1-phenylethyl)-
54	29.625	3996082	0.82	1078288	1.19	3.71	Phenol, 2,4,6-tris(1-phenylethyl)-
55	29.837	619342	0.13	165881	0.18	3.73	3H-pyrazol-3-one, 4-[[4-(diethylamino)-2,6-di
56	30.054	4603592	0.94	1245145	1.38	3.70	Cholesta-4,6-dien-3-ol, (3.beta.)-
57	30.283	15899540	3.25	2932780	3.25	5.42	2-(Decanoyloxy)propane-1,3-diyl dioctanoate
58	30.479	2019901	0.41	451722	0.50	4.47	Vitamin E
59	30.886	410871	0.08	103083	0.11	3.99	Lumisterol 3
60	31.154	301812	0.06	70682	0.08	4.27	Phenol, 2,4-bis(1-phenylethyl)-
61	31.922	5371711	1.10	660864	0.73	8.13	Campesterol
62	32.222	5092128	1.04	760161	0.84	6.70	Stigmasterol
63	32.374	61817	0.01	27007	0.03	2.29	2-(Octanoyloxy)propane-1,3-diyl bis(decanoat
64	32.473	5862100	1.20	1366545	1.51	4.29	2-(Octanoyloxy)propane-1,3-diyl bis(decanoat
65	33.059	30276002	6.19	2339229	2.59	12.94	gamma-Sitosterol
66	33.759	1196682	0.24	170507	0.19	7.02	beta-Amyrin
67	33.861	4885816	1.00	1074597	1.19	4.55	Lup-20(29)-en-3-one
68	34.271	503228	0.10	113981	0.13	4.42	Stigmasta-3,5-dien-7-one
69	34.499	20537784	4.20	1722738	1.91	11.92	Epilupeol
70	34.907	629482	0.13	132205	0.15	4.76	gamma-Sitostenone
71	35.516	1816732	0.37	337598	0.37	5.38	Decanoic acid, 1,2,3-propanetriyl ester
72	36.497	237032	0.05	53849	0.06	4.40	Phytyl tetradecanoate
		489343643	100.00	90283010	100.06		

The percentage content of compounds are Epilupeol (Rt 32.222), gamma-Sitosterol (Rt 33.059), Vitamin E (Rt 30.479), beta-Amyrin (Rt 33.759); Stigmasterol (Rt 28.741), Campesterol (Rt 31.922), Eicosanoic acid (Rt 24.341), Benzene methanol, 2-hydroxy-5-methyl- (Rt 11.913) observed found to be 4.20, 6.19, 0.41, 0.24, 1.04, 1.10,

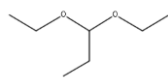
0.08, 0.29% respectively. Some other constituents of significance were Decanoic acid, 1,2,3-propanetriyl ester, Glycerol tricaprylate, Squalene, 1,3-Dioctanoin. Due to the presence of above mentioned compounds in the ethanol extract of *Prosopis juliflora* leaves, it can be used in various pharmaceutical and industrial applications.



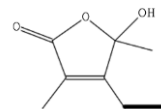
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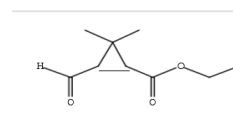
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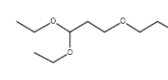
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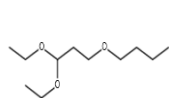
C₈H₁₂O₃



C₉H₁₄O₃



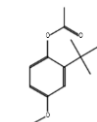
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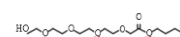
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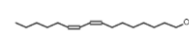
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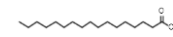
C₁₃H₁₈O₃



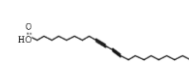
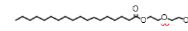
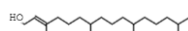
C₁₄H₂₈O₇



C₁₆H₃₀O



C₁₇H₃₄O₂



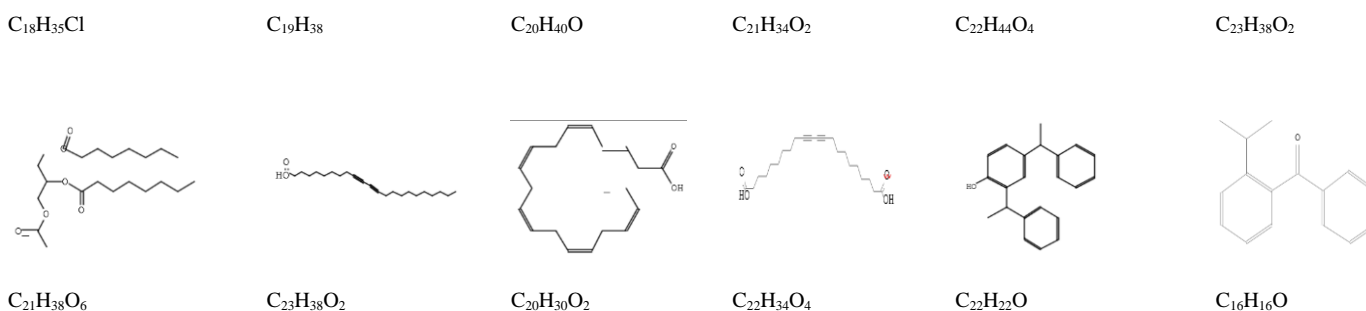


Figure 2: Structures of the GC–MS identified compounds from the Ethanol extract of *Prosopis juliflora* leaves

The 2-D structures of the identified compounds are shown in Fig.2

CONCLUSION

Over the past several years, there has been a steady increase in fascination with studying plants, one of the richest sources of potential, diverse chemical compounds. Plants could be very helpful in discovering new resources to combat the challenges posed by contemporary and novel diseases. *Prosopis juliflora* may be an entirely novel source of pharmaceuticals because of the existence of these phytochemicals and bioactive substances, according to the findings of this investigation.

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Conflict of Interest

None declared.

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