Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

J P P Journal of Pharmacognosy and Phytochemistry

ISSN 2278-4136
ISSN 2349-8234
JPP 2014; 2 (6): 79-82
Received: 08-02-2014
Accepted: 27-02-2014

Sheema Bai
Medicinal Plant Lab.,
Dept. of Biotechnology,
Kurukshetra University,
Kurukshetra-136119, Haryana,
India.
Tel: +91-8950213221;
E-mail: sheema.yadav8@gmail.com

Pooja Bharti
Medicinal Plant Lab.,
Dept. of Biotechnology,
Kurukshetra University,
Kurukshetra-136119, Haryana,
India.
Tel: +91-9416105012;
E-mail: poojalangyan@gmail.com

Leena Seasotiya
Medicinal Plant Lab.,
Dept. of Biotechnology,
Kurukshetra University,
Kurukshetra-136119, Haryana,
India.
Tel: +91-8901579113;
E-mail: leenaseasotiya22@gmail.com

Anupma Malik
Address: Medicinal Plant Lab.,
Dept. of Biotechnology,
Kurukshetra University,
Kurukshetra-136119, Haryana,
India.
Tel: +91-9468045455;
E-mail: anamika.malik86@gmail.com

Dr. Sunita Dalal
Assistant Professor,
Dept. of Biotechnology,
Kurukshetra University,
Kurukshetra-136119, Haryana,
India.
Tel: +91-9812001469;
E-mail: sdalal@kuk.ac.in

Correspondence:
Dr. Sunita Dalal
Assistant Professor,
Dept. of Biotechnology,
Kurukshetra University,
Kurukshetra-136119, Haryana,
India.

GC-MS analysis of chloroform extract of *Acacia nilotica* L. leaves

Sheema Bai, Leena Seasotiya, Anupma Malik, Pooja Bharti and Sunita Dalal

ABSTRACT

The aim of this study was to carry out for identification of bioactive compounds from the chloroform extract of *Acacia nilotica* L. leaves by Gas chromatography and Mass spectroscopy (GC-MS). The GCMS analysis revealed the presence of various compounds like 2,4 dimethyl-butylphenol, palmitic acid, linolenic acid, stearic acid, 2-methylresorcinol acetate, 1,3,4 eugenol, megastigmatrienone, neophytadiene, myristic acid, lariciresinol, 3,4,7-trimethylquercetin, δ-5-avenasterol, and arachidonic acid in the chloroform extract of *A. nilotica* leaves. Further studies are needed to isolate active compounds of the extract as well as to explicate their exact mechanism of action in various disorders.

Keywords: *Acacia nilotica*, GC-MS analysis, Chloroform extract.

1. Introduction

Medicinal plants are at great interest to drug industries, as herbal medicines and their derivative products are often prepared from crude plant extracts, which comprise a complex mixture of different phytochemical constituents. Natural compounds extracted from plants, particularly higher plants, have been suggested as alternative sources for antibiotics. The chemical features of these constituents differ considerably among different species. This approach is alluring, in part, because they constitute a potential source of bioactive compounds that have been professed by the general public as comparatively safe and often act at multiple and novel target sites, thereby reducing the potential for resistance [1].

Acacia nilotica (L.) commonly known as Babool belonging to family Fabaceae is very commonly growing medium sized tree. The leaves are bipinnate, with 3-6 pairs, of pinnulae and 10-30 pairs of leaflets each, tomentose, rachis with a gland at the bottom of the last pair of pinnulae. *A. nilotica* is reported to have characteristic medicinal uses such as appetite enhancer, strength and nutrient supplement, for sore joints, stomach ache and clear out wounds. Natural antioxidants such as flavonoids, phenolics, tannins, curcumin and terpenoids are found in this plant [2].

Extraction is the main step for the recovery and isolation of bioactive phytochemicals from plant materials, before component analysis [3]. Hence, for the discovery of lead compounds for use as therapeutic drugs, the active principals in medicinal plants needs to be identified [4]. GC-MS method can serve as an interesting tool for testing the amount of some active principles of herbs. It combines two analytical techniques to a single method of analyzing mixtures of chemical compounds. Gas chromatography separates the components of the mixture and mass spectroscopy analyzes each of the components separately.

Numerous studies on *A. nilotica* showed various interesting biological activities [5, 6, 7]. Hence, the present study was aimed to carry out the identification of bioactive compounds from the chloroform extract of *Acacia nilotica* leaves by Gas chromatography and Mass spectroscopy (GC-MS).

2. Material and methods

2.1 Collection of plant material

A. nilotica leaves were collected from Northern rural areas (around Rewari region) of Haryana, India, during the period 2011-2012. Further identification and authentication of the specimens was done from Department of Botany, Kurukshetra University, Kurukshetra. The leaves were thoroughly washed with tap water followed by distilled water, were dried under shade for 7 days and ground into fine powder. After sieving (80 mesh) they were transferred to air-tight polyethylene zipper bags, labeled and stored till further use. Voucher plant specimen was deposited at the Wild Life Institute of India, Dehradun, under specimen number GS440 for future reference.

2.2 Preparation of Plant Extracts

The powdered plant leaves (5 g) were successively extracted with hexane, chloroform, and ethyl acetate. The extraction was done by hot continuous soxhlet extraction method. The extracts were stored at -4 °C till further uses. Chloroform extract was used for the present study.

2.3 GC-MS analysis

GC-MS technique was used in this study to identify the phytocomponents present in the extracts. The tested extracts were analyzed by GC-MS using Shimadzu Mass Spectrometer-2010 series. 1 µl of sample was injected in GC-MS equipped with a split injector and a PE Auto system XL gas chromatograph interfaced with a Turbo-mass spectrometric mass selective detector system. The MS was operated in the EI mode (70 eV). Helium was employed as the carrier gas and its flow rate was adjusted to 1.2 ml/min. The analytical column connected to the system was an Rtx-5 capillary column (length-60 m × 0.25 mm i.d., 0.25 µm film thickness). The column head pressure was adjusted to 196.6 kPa. Column temperature programmed from 100 ˚C (2 min) to 200 ˚C at 10 ˚C/min and from 200-300 ˚C at 15 ˚C/min withhold time 5 and 22 min respectively. A solvent delay of 6 min was selected. The injector temperature was set at 260 °C. The GC-MS interface was maintained at 280 °C. The MS was operated in the ACQ mode scanning from m/z 40 to 600.0. In the full scan mode, electron ionization (EI) mass spectra in the range of 40–600 (m/z) were recorded at electron energy of 70 eV. Compounds were identified by comparing mass spectra with library of the National Institute of Standard and Technology (NIST), USA/Wiley.

3. Results and discussion

3.1 GC-MS analysis

The analysis and extraction of plant material play an important role in the development, modernization and quality control of herbal formulations. Hence the present study was aimed to find out the bioactive compounds present in the chloroform extract of *A. nilotica* by using Gas chromatography and Mass spectroscopy. The active compounds with their peak number, concentration (peak area %), and retention time (RT) are presented in Table 1 and Fig. 1 which shows the presence of 63 bioactive phytochemical compounds in the chloroform extract of *A. nilotica*.

Table 1: GC-MS spectral analysis of chloroform extract of *A. nilotica* leaves

Peak no.	Area %	Compound	Retention time
1	0.28	Decane, 3,7-dimethyl-	7.273
2	0.36	Dihydrocitronellol	7.790
3	0.60	Pelargonaldehyde	8.459
4	1.02	Undecane	10.616
5	0.34	6-dimethylamine-saccharin	12.178
6	0.39	Nonane, 5-(2-methylpropyl)-	12.442
7	0.48	1-Chlorohexadecane	12.904
8	0.47	Hexadecane	13.477
9	3.59	1,3,4-Eugenol	14.375
10	0.58	Pentadecane	15.085
11	0.51	Phosphoric acid, bis(trimethylsilyl)monomethyl ester	15.500
12	22.36	2,4 Dimethyl-butylphenol	17.608
13	0.51	Heptadecane	17.967
14	0.90	2,6,6-Trimethyl-2-hydroxycyclohexylidene acetolactone	18.034
15	-21.58	3',5'-Dimethoxyacetophenone	17.608
16	2.40	Fumaric acid, ethyl 2-methylallyl ester	18.972
17	0.32	Phthalic acid	19.206
18	0.61	Megastigmatrienone	19.845
19	0.76	4-(1,5-Dihydroxy-2,6,6-trimethylcyclohex-2-enyl)but-3-en-2-one	20.083
20	0.34	3-Oxo-.alpha.-ionol	20.300
21	0.73	Cinnamic acid, 3-hydroxy-4-methoxy	21.318
22	0.58	(3-Oxo-2-pent-2-enylcyclopentyl)acetic acid	21.783
23	0.89	Myristic acid	22.325
24	6.30	2-Methylresorcinol, acetate	22.967
25	1.29	Neophytadiene	23.460
26	0.69	Phthalic acid,butyl octyl ester	24.064
27	0.76	Tetracosane	24.846
28	0.23	1,54-Dibromotetrapentacontane	25.342
29	1.21	Eicosane	25.534

30	8.70	Palmitic acid	25.680
31	1.34	Palmitic acid, ethylester	26.031
32	0.81	Isopropyl Palmitate	26.503
33	1.06	1,11-Hexadecadiyne	27.389
34	0.34	Linolenic acid, methyl ester	27.713
35	1.05	Cedrane-8,13-diol	27.808
36	0.67	Tetracosane	28.177
37	12.40	Linolenic acid	28.435
38	9.04	Stearic acid	28.741
39	1.61	Stearic acid ethyl ester	29.067
40	0.57	Dotriacontane	29.318
41	0.59	Arachidonic acid	30.408
42	0.77	Dipalmitin	30.560
43	0.80	γ-Linolenic acid	31.036
44	1.92	Octadecane, 3-ethyl-5-(2-ethylbutyl)-	31.635
45	0.97	Tetracosane	31.772
46	0.52	1,3,5-Trisilacyclohexane	32.999
47	0.84	Palmitoyl chloride	33.110
48	0.32	Phenol, 4,4'-methylenebis[2,6-bis(1,1-dimethylethyl)-	34.471
49	0.36	Tetrapentacontane	34.573
50	0.79	Decyl sulfide	37.567
51	0.60	Hexatriacontane	39.883
52	0.30	Oxirane, hexadecyl-	40.592
53	0.43	Hexatriacontane	40.904
54	0.68	Tetrapentacontane	42.308
55	1.17	(+)-Lariciresinol	42.654
56	1.78	(+)-Lariciresinol	42.794
57	0.37	δ-5-Avenasterol	43.342
58	0.31	3,4,7-trimethylquercetin	44.333
59	1.03	1,3,4-Eugenol	44.709
60	0.24	Lariciresinol	44.875
61	0.73	Phthalic acid	44.960
62	0.67	Terephthalic acid ester of neopentyl glycol cyclic dimer	45.194
63	18.28	3,5,7-Tris (trimethylsiloxy)-2-[3,4-di(trimethylsiloxy) phenyl]-4H-1-Benzopyran-4-one	46.061
	100		

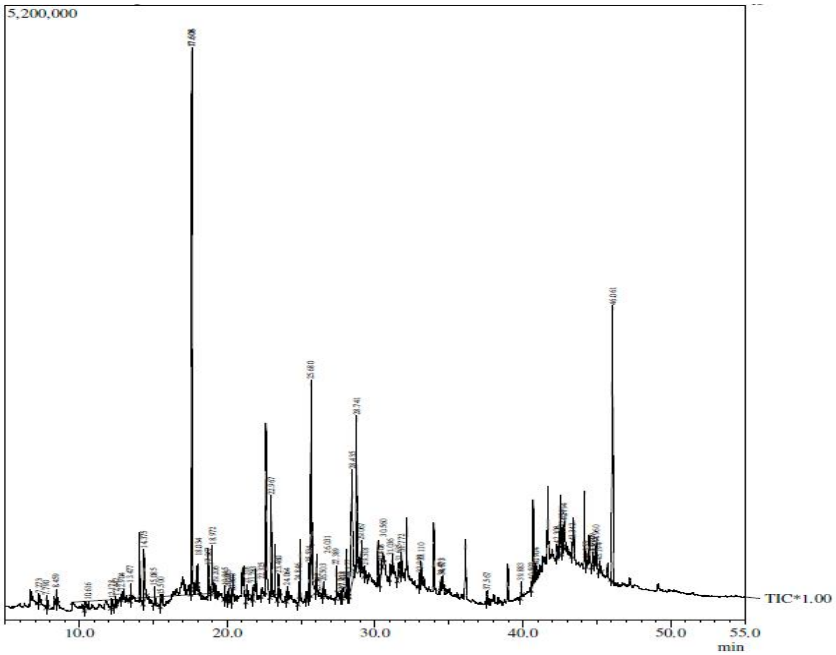

Fig 1: GC-MS chromatogram for chloroform extract of *A. nilotica* leaves

The percentage content of compounds are 2,4 dimethyl-butylphenol (R_t 17.608); 3,5,7-tris (trimethylsiloxy)-2-[3,4-di(trimethylsiloxy) phenyl]-4H-1-Benzopyran-4-one (R_t 46.061), linolenic acid (R_t 28.435); stearic acid (R_t 28.741), palmitic acid (R_t 25.680), 2-methylresorcinol (R_t 22.967), 1,3,4-eugenol (R_t 14.375) observed found to be 22.36, 18.28, 12.40, 9.04, 8.70, 6.30 and 3.59% respectively. Some other constituents of significance were megastigmatrienone, dihydrocitronellol, neophytadiene, arachidonic acid, (+)-Lariciresinol, δ-5-Avenasterol, and 3,4,7-trimethylquercetin. Due to the presence of above mentioned compounds in the chloroform extract of *A. nilotica* leaves, it can be used in various pharmaceutical and industrial applications.

4. Conclusion

The demand in study of plants, which is one of the richest sources of promising versatile chemical compounds, is growing persistently throughout the world during the last few decades. Plant could play a great role in exploring new resources against the threats of new and recent diseases. From this study it can be concluded that the *A. nilotica* may serve as a new potential source of medicines due to the presence of these phytochemicals and bioactive compounds.

5. Acknowledgement

This work was financially supported by the Haryana State Council for Science & Technology, Panchkula under Grant HSCST Fellowship 2011/1, letter no. 1788-97.

6. References

1. Raskin I, Ribnicky DM, Komarnytsky S, Ilic N, Poulev A, Borisjuk N, Brinker A, Moreno DA, Ripoll C, Yakoby N, O'Neal JM, Cornwell T, Pastor I, and Fridlender B. Plants and human health in the twenty-first century. Trends in Biotechnology 2002; 20: 522-531.
2. Kahkonen MP, Hopia AI, Vuorela HJ, Rauha JP, Pihlaja K, Kujala TS and Heinonen M. Antioxidant activity of plant extracts containing phenolic compounds. Journal of Agricultural and Food Chemistry 1999; 47: 3954-3962.
3. Karimi E and Jaafar HZE. HPLC and GC-MS determination of bioactive compounds in microwave obtained extracts of three varieties of *Labisia pumila* benth. Molecules 2011; 16: 6791–6805.
4. Vuorela P, Leinonen M, Saikku P, Tammela P, Rouha JP, Wennberg T, and Vuorela H. Curr Med Chem 2004; 11:1375–1389.
5. Meena PD, Kaushik P, Shukla S et al. Anticancer andantimutagenic properties of *Acacia nilotica* (Linn.) on 7,12- dimethylbenz(a) anthracene-induced skin papillomagenesis in Swiss albino mice. Asian Pac J Cancer Prev 2006; 7: 627-32.
6. Bachaya HA, Iqbal Z, Khan MN, Sindhu ZD, and Jabbar A. Anthelmintic activity of *Ziziphus nummularia* (bark) and *Acacia nilotica* (fruit) against Trichostrongylid nematodes of sheep. J Ethnopharmacol 2009; 123: 325-9.
7. Singh R, Singh B, Singh S, et al (2010). Umbelliferone – An antioxidant isolated from *Acacia nilotica* (L.) Willd. Ex Del Food Chem 2010; 120: 825-30.