

Phytochemical investigation and antioxidant activities of *Albizia saman* flowers

Sundar M, Anton Smith A*, Amutha Iswarya Devi J, Kottai Muthu A and Parimalkrishnan S

Department of Pharmacy, Annamalai University, Annamalainagar-608 002, Tamilnadu.

*Corresponding author: Email: auantonsmith@yahoo.co.in

ABSTRACT

The present research was directed to identify the chemical compound found in *Albizia saman* flower (Family: Leguminosae). Thirty eight compounds were identified by gas chromatography – mass spectrometry (GC-MS) Analysis. The 10 major compounds are Myo-Inositol, 4-C methyl (31.44%), n-Hexadecanoic acid (11.98%), Hexadecanoic acid methyl ester (7.35%), 9,12-Octadecadienoic acid (z,z)-(6.44%), Octadecanoic acid (5.55%), 9,12, Octadecadienoic acid, methyl ester (4.8%), 9-Octadecanoic acid (z)-methyl ester (3.86%) 1-Hexadecanol (3.31%), Stigmasteral (2.50%) Butanedioic acid, methoxy-, dimethyl ester (2.37%) and 27 minor compounds were identified in alcoholic extracts from *Albizia saman* flower. Antioxidant activity of *Albizia saman* flower was carried out using free radical scavenging activity. Hydrogen peroxide scavenging activity and superoxide ion radical scavenging activity, in which the maximum antioxidant activity is present in ethanolic extract. The results shows the highest peak of free-radical radical scavenging and antioxidant activity.

Key words: *Albizia saman*; Phytochemical; Antioxidant activity; GC-MS study

1. INTRODUCTION

Albizia saman formerly called *Samanea samam* (family Leguminosae), a fast budding tree normally used as pasture for ornamental purposes. The bark, leaves, root, seeds and pods of the tree are so far used as medicine from the traditional system (Thippeswamy, 2011). The alcoholic extract of selected leaves introverted *Mycobacterium tuberculosis*, the alkaloid portion of leaves is active on CNS and as laxative. Seeds are masticated for stinging throat. A decoction from the fresh leaves and inner bark are used for colds, diarrhea, and intestinal problem (Krithika, 2013).

The literature search indicated that *Albizia saman* contains alkaloids, glycosides, terpenoids etc. The mature pods of *Albizia saman* are black brown, almost oblong, clumpy, 10 to 20 cm long, 15 to 19 mm wide, 6 mm thick, slightly curved, eventually cracking irregularly, and filled with brownish pulp which is sticky sweetly and edible. The bottommost line is that the pods of *Albizia saman* tree are rarely used as a plant source for herbs. Knowledge on phytochemical constituents towards plant parts is mandatory in appreciative the basis for any therapeutic effect. For instance, the flavonoids, which are ubiquitous in higher plants and common part of human diet could significantly inhibit microbes which are resistant to conventional antibiotics. Recently, isolated flavonoids were reported to exhibit antimicrobial activity. In addition to that the flavonoids through their free-radical scavenging activity have evoked multiple biological functions, including vasodilatory, anti-carcinogenic, anti-inflammatory, anti-bactericidal, immune stimulatory, anti-allergic and anti-viral functions. Out of phytochemicals may possibly avoid diseases and inhibit pathogenic microorganisms. Indeed, proper composition of phytochemicals appears to confer plants and plants parts with peculiar medicinal properties. This makes the interest to screen higher plants for active agents with antifungal and antimicrobial/anti-bacterial activities against plant and human pathogens. (Staples, 2015)

Tannins could bind to proteins for the effective development of strong multiplexes with proteins and also other macromolecules. Tannins are associated with numerous therapeutic effects. Considering the immense medicinal property to Leguminosae, the present study was carried out the phytochemical constituents of various extracts from *Albizia saman* (Faisal Ferdous, 2010)

2. MATERIALS AND METHODS

“*Albizia saman*” flower was collected in July 2015 Annamalainagar, Chidambaram, Cuddalore district of Tamilnadu, India. The effect of alcoholic extract on DPPH and superoxide radical scavenging activity (SOD) were analyzed identified antioxidant activity (Ahmed Syed Muzammil, 2013).

GC-MS Analysis: GC-MS analysis (Abou-Donia, 2008.) of ethanolic extract was performed by means of a Perkin-Elmer GC clauses 500 system with GC that was interfaced to a mass spectrometer (GC-MS) employed a fused silica capillary column packed with Elite-1. To detect an EI system (70eV) was used. An inert helium gas (99.999%) used here as carrier gas at constant stream rate 1ml/min and an injection volume of 2 µl was employed (Split ratio of 10:1) injector temperature setting at 250°C; ion-source temperature was fixed as 280°C. The temperature of the oven was set from 110°C (2min) with an rise of 10°C/min to 200°C, then 5°C/min to 280°C, finish with a 9 min isothermal at 280°C. Mass spectra was taken at 70 eV; as canister val of 0.5 seconds and fragments from 45 to 450 Da. (Pius OU, 2011).

DPPH photometric assay: The influence of alcoholic extract on DPPH radical was analysed using Mensor method. The estimation was performed as described as Elizabeth K, 1990.

Hydroxyl radical scavenging activity: This was assayed as described by Elizabeth and Rao method. The analysis is established on quantitation of degradation product of 2-deoxy ribose by condensation with TBA. Hydroxyl radical was generated by the Fe^{3+} -Ascorbate-EDTA- H_2O_2 system (Fenton reaction) (Bennick, 2002).

Superoxide radical scavenging activity: Superoxide radical (O_2^-) was generated after the photo reduction of riboflavin was deducted by nitro blue tetrazolium dye (NBT) reduction method (Kandaswami, 1998). Plant have good antioxidant ability and are safer than the synthetic antioxidants. Secondary metabolites from therapeutic plants as minor molecular weight antioxidants but their particular mechanism of action are variable and depend both on the construction and environment (Abou-Donia, 2008).

The present research has shown the total antioxidant potential of different extracts of *Albizia saman* ethanolic extract of *Albizia saman* exhibited strongest antioxidant activity among all the extracts.

The free radical scavenging activity of the 1,1-diphenyl-2-picryl-hydrazyl radical (DPPH) assay, hydrogen peroxide scavenging activity and Superoxide ion radical scavenging activity were performed with various extracts such as, ethyl acetate, petroleum ether, ethanol of selected samples were analyzed to estimate their antioxidant properties (Limuma, et al., 1994).

3. RESULTS AND DISCUSSION

The results relating to GC-MS analysis helps in identification of number of compounds that were identified from the GC fractions of the alcoholic extract of *Albizia saman*. Those compounds were identified through mass spectrometry attached with GC. The outcome of present study were tabulated in Table 1. The compound prediction based on Dr. Duke's Phytochemical and Ethnobotanical databases (Evans, 2002). The ethanolic extract showed the highest DPPH scavenging activity which was followed by petroleum ether and ethyl acetate extracts which was present of in Table 2, hydrogen peroxide scavenging activity was presented in Table 3. The SOD activity present in Table 4 (Ferreira, 2008).

Table.1. GC-MS analysis of the ethanolic extract of *Albizia saman*

No.	RT	Name of the compound	Molecular Formulae	Molecular Weight	Peak Area %
1.	3.75	E-2-Octenyl tiglate	$\text{C}_{13}\text{H}_{22}\text{O}_2$	210	0.66
2.	4.31	2H-Pyran-2-one, 6-heptyltetrahydro-	$\text{C}_{12}\text{H}_{22}\text{O}_2$	198	0.27
3.	5.18	9-Methyltricyclo[4.2.1.1(2,5)]deca-3,7-diene-9,10-diol	$\text{C}_{11}\text{H}_{14}\text{O}_2$	178	0.32
4.	5.77	Benzofuran, 2,3-dihydro-	$\text{C}_8\text{H}_8\text{O}$	120	0.16
5.	6.05	Butanedioic acid, methoxy-, dimethyl ester	$\text{C}_7\text{H}_{12}\text{O}_5$	176	2.37
6.	6.18	4-Hydroxylysine	$\text{C}_6\text{H}_{14}\text{N}_2\text{O}_3$	162	1.52
7.	7.24	3,5-Heptadienal, 2-ethylidene-6-methyl-	$\text{C}_{10}\text{H}_{14}\text{O}$	150	0.21
8.	8.18	2-(2-Hydroxyethyl)piperidine	$\text{C}_7\text{H}_{15}\text{NO}$	129	0.03
9.	9.18	1,2-15,16-Diepoxyhexadecane	$\text{C}_{16}\text{H}_{30}\text{O}_2$	254	0.01
10.	11.15	2-Cyclohexylpiperidine	$\text{C}_{11}\text{H}_{21}\text{N}$	167	0.50
11.	13.09	Myo-Inositol, 4-C-methyl-	$\text{C}_7\text{H}_{14}\text{O}_6$	194	31.44
12.	13.93	Adenine	$\text{C}_5\text{H}_5\text{N}_5$	135	2.35
13.	14.53	1-Hexadecanol	$\text{C}_{16}\text{H}_{34}\text{O}$	242	0.67
14.	15.10	Hexadecanoic acid, methyl ester	$\text{C}_{17}\text{H}_{34}\text{O}_2$	270	7.35
15.	15.63	n-Hexadecanoic acid	$\text{C}_{16}\text{H}_{32}\text{O}_2$	256	11.98
16.	16.00	Hexadecanoic acid, ethyl ester	$\text{C}_{18}\text{H}_{36}\text{O}_2$	284	0.35
17.	16.45	Dodecanoic acid, 2,3-bis(acetyloxy)propyl ester	$\text{C}_{19}\text{H}_{34}\text{O}_6$	358	0.15
18.	17.31	1-Hexadecanol	$\text{C}_{16}\text{H}_{34}\text{O}$	242	3.31
19.	17.40	9,12-Octadecadienoic acid, methyl ester	$\text{C}_{19}\text{H}_{34}\text{O}_2$	294	4.93
20.	17.50	9-Octadecenoic acid (Z)-, methyl ester	$\text{C}_{19}\text{H}_{36}\text{O}_2$	296	3.86
21.	17.65	Phytol	$\text{C}_{20}\text{H}_{40}\text{O}$	296	0.44
22.	17.88	Stearic acid, methyl ester	$\text{C}_{19}\text{H}_{38}\text{O}_2$	298	4.84
23.	18.01	9,12-Octadecadienoic acid (Z,Z)-	$\text{C}_{18}\text{H}_{32}\text{O}_2$	280	6.44
24.	18.45	Octadecanoic acid	$\text{C}_{18}\text{H}_{36}\text{O}_2$	284	5.55
25.	18.85	Octadecanoic acid, 17-methyl-, methyl ester	$\text{C}_{20}\text{H}_{40}\text{O}_2$	312	0.57
26.	20.27	Erucic acid	$\text{C}_{22}\text{H}_{42}\text{O}_2$	338	0.45
27.	20.83	cis-13-Eicosenoic acid	$\text{C}_{20}\text{H}_{38}\text{O}_2$	310	0.28
28.	21.35	L-Tryptophan, N-acetyl-, methyl ester	$\text{C}_{14}\text{H}_{16}\text{N}_2\text{O}_3$	260	0.91
29.	22.65	12-Methyl-E,E-2,13-octadecadien-1-ol	$\text{C}_{19}\text{H}_{36}\text{O}$	280	0.27
30.	23.27	Behenic alcohol	$\text{C}_{22}\text{H}_{46}\text{O}$	326	0.82
31.	23.41	13-Docosenoic acid, methyl ester, (Z)-	$\text{C}_{23}\text{H}_{44}\text{O}_2$	352	0.11
32.	24.28	Eicosanoic acid, methyl ester	$\text{C}_{21}\text{H}_{42}\text{O}_2$	326	0.70
33.	24.70	Cholestan-3-ol, 2-methylene-, (3 β ,5 α)-	$\text{C}_{28}\text{H}_{48}\text{O}$	400	0.38
34.	26.08	9,12,15-Octadecatrienoic acid, 2,3-dihydroxypropyl ester, (Z,Z,Z)-	$\text{C}_{21}\text{H}_{36}\text{O}_4$	352	1.16
35.	27.46	8,11,14-Eicosatrienoic acid, (Z,Z,Z)-	$\text{C}_{20}\text{H}_{34}\text{O}_2$	306	1.00
36.	27.79	trans-Farnesol	$\text{C}_{15}\text{H}_{26}\text{O}$	222	0.20
37.	28.64	Hexacosyl acetate	$\text{C}_{28}\text{H}_{56}\text{O}_2$	424	0.96
38.	37.15	Stigmasterol	$\text{C}_{29}\text{H}_{48}\text{O}$	412	2.50

Table.2.Effect of various extracts in free radical scavenging activity (DPPH)

Concentration (µg/ml)	Percentage inhibition of free radical generation				
	L-Ascorbic acid	BHA	EtOH	EA	PE
25	51.710±0.406*	43.632±1.551*	29.142±2.291	9.342±1.231	6.769±0.997
50	55.831±0.482*	57.573±0.330*	28.174±0.402	18.574±3.402	16.796±2.987
75	57.740±1.572*	60.643±1.242*	31.343±0.934	29.143±0.734	26.654±0.543
100	59.878±1.749*	66.254±1.870*	49.752±2.735*	35.452±0.735	33.564±0.654
250	66.458±1.797*	69.732±0.271*	65.658±0.331*	46.658±3.331	45.765±2.256
500	68.235±1.289*	71.243±1.282*	67.739±1.432*	49.399±1.432	47.871±1.562

n=3. Statistical analysis was carried out using one way ANOVA followed by Dunnett's multiple comparison tests.

*P < 0.05

Table.3.Effect of various extracts in hydrogen peroxide scavenging activity

Concentration (µg/ml)	Percentage inhibition of hydrogen peroxide generation				
	L-Ascorbic acid	BHA	EtOH	EA	PE
25	13.720±0.046*	15.232±2.251*	12.391±2.653*	8.039±2.253	6.762±1.287
50	24.431±0.682*	18.573±0.480*	26.592±0.872*	16.594±3.892	14.656±2.453
75	35.940±0.172*	37.643±0.342*	36.256±1.254*	32.154±1.454	29.652±1.435
100	49.178±0.189*	48.654±0.270*	47.291±0.525*	46.242±0.825*	36.135±0.463
250	56.458±0.797*	58.742±0.771*	57.295±0.532*	52.238±0.521*	45.157±1.435
500	58.835±0.389*	67.643±0.282*	61.403±0.087*	57.320±0.277*	47.546±0.495

n=3. Statistical analysis was carried out using one way ANOVA followed by Dunnett's multiple comparison tests.

*P < 0.05

Table.4.Effect of various extracts in superoxide anion radical scavenging activity

Concentration (µg/ml)	Percentage inhibition of superoxide generation				
	L-Ascorbic acid	BHA	EtOH	EA	PE
25	10.231±1.214*	15.632±1.251*	0.932±0.302	0.349±0.010	0.234±0.073
50	15.831±0.682*	19.873±0.630*	3.774±0.052	2.174±0.020	1.783±0.102
75	18.240±0.002*	26.843±1.242*	17.543±0.854*	5.343±0.154	4.432±1.230
100	28.478±0.249*	52.584±1.870*	27.642±0.315*	10.542±0.015	5.563±0.251
250	70.158±2.297*	82.232±0.271*	61.458±0.431*	11.850±0.131	10.985±1.325
500	78.835±0.289*	88.693±1.412*	76.799±0.456*	15.989±1.056	11.213±0.329

n=3. Statistical analysis was carried out using one way ANOVA followed by Dunnett's multiple comparison tests.

*P < 0.05

4. CONCLUSION

Numerous phytochemical screening studies have been carried out in different parts of the world using GC-MS. In the present study, we characterized the chemical profile of *Albizia saman* using GC-MS. The gas chromatogram shows 38 compounds eluted. This mass spectrum were the fingerprint of that compound which can be identified from the data library. This report is the first of its kind to analyze the chemical constituents of *Albizia saman* using GC-MS. In addition to this, the results of the GC-MS profile can be used as pharmacognostical tool for the identification of the plant. The result of the present study supported and supplemented the previous observations (Staples GW, 2015). The presence of various bioactive compounds confirms the application of *Albizia saman* for various ailments by traditional practitioners. The ethanolic extract showed the maximum DPPH and total antioxidant activities, among the antioxidant activity of study (Elizabeth, 1990).

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