Available online at www.ijpsdr.com

International Journal of Pharmaceutical Sciences and Drug Research 2016; 8(3): 149-152

Research Article

ISSN: 0975-248X
CODEN (USA): IJPSPP

Identification of Chemical Compounds from the Ethanolic Extract of the Bark of *Bauhinia tomentosa* L. By GC-MS Analysis

Gopalakrishnan S[1*], Vadivel E[2]

[1]*Noorul Islam University, Kumaracoil-629180, K. K. District, Tamil Nadu, India*
[2]*Post-Graduate Chemistry Department, Dnyanprassarak Mandal`s College and Research Centre, Assagao-Bardez, Goa, India*

ABSTRACT
The bark of *Bauhinia tomentosa* L. is used wildly used for the treatment of varies ailments such as inflammation, wound, dysentery, skin diseases and for microbial infections. In the present study the ethanolic extract of the bark of *Bauhinia tomentosa* L. has been subjected to Gas Chromatography-Mass Spectrometry (GC-MS) analysis, while the mass spectra of the compounds found in the extract was matched with the National Institute of Standards and Technology (NIST) library. GC-MS analysis revealed the presence of fourteen phytochemical constituents, These compounds were identified by comparing their retention time and peak area with that of literature and by interpretation of mass spectra. The major chemical constituents are 4-(Benzyloxy)-1-methoxy-2-(3,7-dimethylocta-2,6-dienyl)-benzene (10.88%), Levodopa (6.39%) and (9E, 12E) - Methyl octadeca-9,12-dienoate (5.68%) which possess many biological activities. Hence these studies pave a platform to screen many bioactive components for wound healing.

Keywords: *Bauhinia tomentosa* L., Phytochemicals, GC-MS analysis, Levodopa and wound healing.

INTRODUCTION

Herbal medicines are safe than synthetic medicines because the phytochemicals in the plant extract target the biochemical pathway. [1] They are constantly being screened for their biological and pharmacological activities such as anti-diabetic, anti-oxidant, anti-microbial, laxative, wound healing and anti-cancer activities.[2-8] The medicinal plants are having numerous bioactive components which are identified even at less than 1ng by using GC-MS or LC-MS analysis. Gas Chromatography-Mass Spectroscopy, a hyphenated system which is a very compatible technique and the most commonly used technique for the identification and quantification purpose.

The unknown organic compounds in a complex mixture can be determined by interpretation and also by matching the spectra with reference spectra. [9] Gas Chromatography (GC/MS) is normally used for direct analysis of chemical components existing in herbal medicines. For the analysis of medicinal plants, GC-MS technique have been proved to be highly commended analysis for non-polar components and volatile essential oil, fatty acids, lipids and alkaloids. [10]

Bauhinia is a well known genus for its therapeutic efficacy. This genus includes more than 200 species of flowering plants. One of the most important species of this genus *Bauhinia tomentosa* L. (Family, *Fabaceae*) is commonly known as "Kanjana" in Tamil and "Phalgu" in Sanskrit. *Bauhinia tomentosa* L. is distributed

***Corresponding author: Dr. S. Gopalakrishnan,**
Senior Professor of Chemistry and Controller of Examinations, Noorul Islam University, Kumaracoil-629180, K. K. District, Tamil Nadu, India; **Tel.:** +91-9443172243; **E-mail:** sgkmsu@yahoo.co.in
Received: 28 March, 2016; **Accepted:** 18 April, 2016

throughout Southern India, Assam and Bihar. The dried leaves, buds and flowers are prescribed in dysentery, flowers are also used for anti-hyperglycemic and anti-lipidemic activity. [11] The bruised bark is applied externally to tumors and wounds. A decoction of the root-bark is administered for inflammation of the liver and it is also used for diabetic. [12] An infusion of the bark is also used as an astringent gargle.

The aim of the present study is to identify the phytocomponents of this plant and subjecting the methanol extract of the plant leaves to Gas chromatography-Mass Spectrum analysis. In the present study, volatile organic matter of ethanolic extract of the bark of *Bauhinia tomentosa* L. was analyzed for the first time. This work will help to identify the compounds, which may be used in therapeutic value.

MATERIALS AND METHODS

Plant materials

The plant was collected in the month of September from Courtallam Hills of Tirunelveli District, Tamil Nadu and identified by Prof. P. Jayaraman, Plant Anatomy Research Center, West Thambaram, Chennai. A voucher specimen (MSU/PHAR/HER–138) was preserved in the Herbarium of the Department of Pharmaceutical Chemistry, Manonmaniam Sundaranar University, Tirunelveli -627 012.

Extraction of plant material

The fresh bark was cleaned with distilled water to remove extraneous matter, shade-dried until to get constant weight and then powdered. The dried powder of the bark of the plant (500 g) was successively extracted using petroleum ether (40-60°C), chloroform, ethanol and water by using Soxhlet apparatus. The last trace of solvent was removed under reduced pressure distillation and then vacuum dried. The dried crude ethanolic extract was used for the GC-MS analysis.

GC-MS analysis

Preparation of extract

2µl of the ethanolic extract of bark of *Bauhinia tomentosa* L. was employed for GC-MS analysis. [13]

Instruments and chromatographic conditions

GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: column Elite-1 fused silica capillary column (30 × 0.25 mm ID × 1EM df, composed of 100% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1 ml/min and an injection volume of 0.5 EI was employed (split ratio of 10:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min), with an increase of 10°C/min, to 200°C/min, then 5°C/min to 280°C/min, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5 s and fragments from 40 to 550 Da.

Identification of components

The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Interpretation of mass spectrum of GC-MS was done using the database of National Institute of Standard and Technology (NIST) having more than 62,000 patterns. The mass spectrum of the unknown component was compared with the spectra of the known components stored in the NIST library. The name, molecular weight and structure of the constituents isolated were ascertained. The compound prediction is based on Dr. Duke's Phytochemical and Ethnobotanical Databases by Dr. Jim Duke of the Agricultural Research Service/USDA.

RESULTS AND DISCUSSION

Gas Chromatography-Mass spectrometry (GC-MS) chromatogram of the ethanolic extract the bark of *Bauhinia tomentosa* L. (Fig. 1) showed 14 peaks indicating the presence of fourteen phytochemical constituents. On comparison of the mass spectra of the constituents with the NIST library the fourteen phytoconstituents were characterized and identified. The chemical name, retention time (RT), molecular formula, molecular weight (MW), peak area (%) and biological activity of various phytochemicals (Dr. Duke's Phytochemical and Ethnobotanical Databases) are presented in Table 1. The mass spectra of major chemical constituent's are presented in Fig. 2-Fig. 4. They were identified as 4-(Benzyloxy)-1-methoxy-2-(3, 7-dimethylocta-2,6-dienyl)-benzene (10.88%), Levodopa (6.39%) and (9E, 12E) - Methyl octadeca-9, 12-dienoate (5.68%) respectively.

Unsaturated fatty acids are important to every cell for normal growth, to support the lubricating quality of skin and to lower cholesterol levels of the blood. Presence of anti-inflammatory compound, (9E, 12E) - methyl octadeca-9,12-dienoate may possibly play a role in curing skin diseases. Levodopa was used in the clinical treatment of Parkinson's disease. Levodopa was reported in *Mucuna pruriens* by Manyam *et al.* [14] Presence of antimicrobial, antioxidant and anti-inflammatory constituents (2, 3, 5, 6, 9, 10, 11, 12, and 14) may be responsible for the wound healing activity of the plant. Phenol and polyphenols are considered to be major contributors to the antioxidant property of fruits, vegetables and mushrooms. Phenolic compounds are potent water soluble and free radical scavenger which prevent oxidative cell damage. [15] A number of studies have been focused on biological activities of a phenolic compound as a potential antioxidant and free radical scavengers. 1,2-Benzenediol (2), 2-Propyl phenol (3) and Levodopa (6) are phenolic compounds identified in the present study may be responsible for its antioxidant activity. Antioxidant compounds also increase the antimicrobial and anti-inflammatory activity.

Table 1: Phytochemical constituents identified from the ethanolic extracts of the bark of *Bauhinia tomentosa* L. and their biological activity.

S. No.	RT	Name of the compound	Molecular Formula	MW	Peak Area %	Compound Nature	* Biological Activity
1.	3.82	Tetrahydrofurazone-3,4-diol	$C_4H_8O_3$	104	4.00	Alcohol	Antimicrobial
2.	4.62	1,2-Benzenediol	$C_6H_6O_2$	110	3.86	Phenol	Antimicrobial, antioxidant and Anti-inflammatory
3.	6.65	2-Propyl phenol,	$C_9H_{12}O$	136	3.72	Phenol	Antimicrobial, Antioxidant and Anti inflammatory
4.	7.66	Sucrose	$C_{12}H_{22}O_{11}$	342	12.35	Disaccharide	Preservative
5.	9.03	(R)-3-(4-(hydroxy methyl)phenyl)propane-1-ol	$C_{10}H_{14}O_2$	166	1.40	Alcohol	Antimicrobial
6.	9.46	Levodopa	$C_9H_{11}NO_4$	197	6.39	Phenolic	Antimicrobial, Anti-inflammatory
7.	11.30	(2R,3S,4R,5R)-2,4,5,6-tetrahydroxy-3-methoxyhexanal	$C_7H_{14}O_6$	194	4.42	Disaccharide	Preservative
8.	11.67	1-Methyl cyclohexane-1, 2, 3, 4, 5, 6-hexol	$C_7H_{14}O_6$	194	4.77	Alcohol	Antidiabetic
9.	13.08	[R-(Z)]-Methyl -12-acetoxyoctadec-9-enoate	$C_{21}H_{38}O_4$	354	3.58	Ricinolic acid ester	Antimicrobial and Anti-inflammatory
10.	13.51	Butyl isobutyl phthalate	$C_{16}H_{22}O_4$	278	1.40	Plasticizer	Antimicrobial
11.	15.27	(9E,12E)- methyl octadeca-9,12-dienoate	$C_{19}H_{34}O_2$	294	5.68	Linoleic acid ester	Anti-inflammatory and Cancer preventive
12.	16.08	Dibenzyl sulfane	$C_{14}H_{14}S$	214	2.67	Sulfur	Antimicrobial
13.	21.23	4-(benzyloxy)-1-methoxy-2-((E)-3,7-dimethylocta-2,6-dienyl)-benzene,	$C_{24}H_{30}O_2$	350	10.88	Aromatic	No activity reported
14.	33.85	(1S,3E,4S)-3-[(2E)-2-[(1R,3aS,7aR)-1-[(E,2R,5R)-5,6- Dimethylhept-3-en-2-yl]-7a-methyl-2,3,3a,5,6,7- hexahydro-1H-inden-4-ylidene]ethylidene]-4-methylcyclohexan-1-ol	$C_{28}H_{46}O$	398	3.86	Steroid	Antimicrobial, Antiasthmatic, Anti-inflammatory, Anticancer and Diuretic

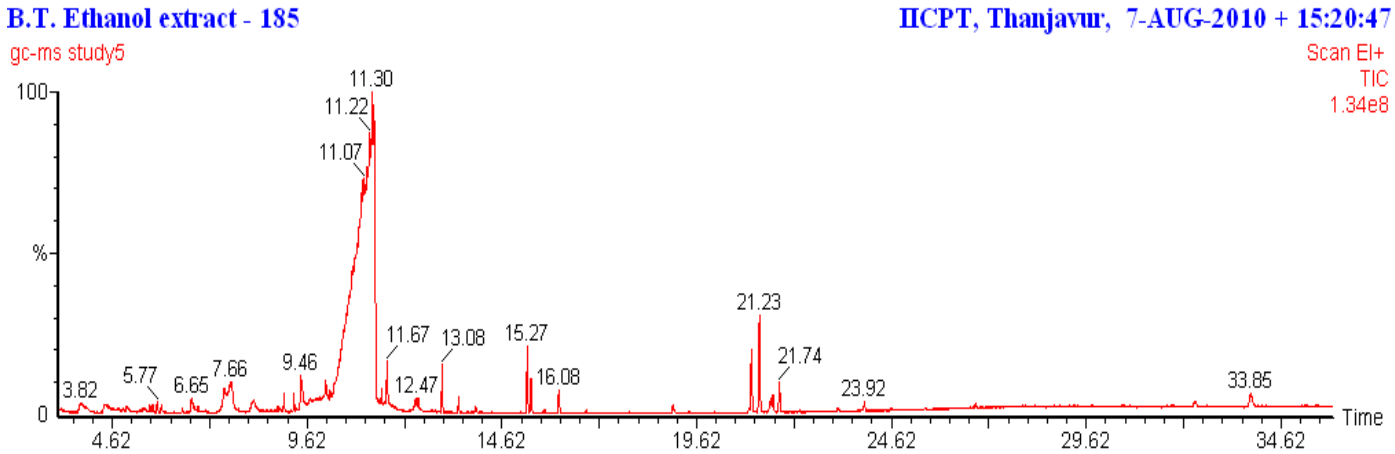

Fig. 1. GC-MS chromatogram of the ethanolic extract of bark of *Bauhinia tomentosa* L.

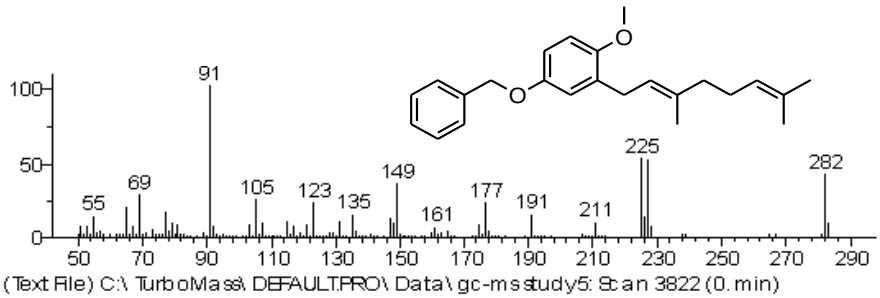

Fig. 2: Mass spectrum of 4-(benzyloxy)-1-methoxy- 2- ((E)-3, 7-dimethylocta-2, 6-dienyl)-benzene, (RT: 21.23)

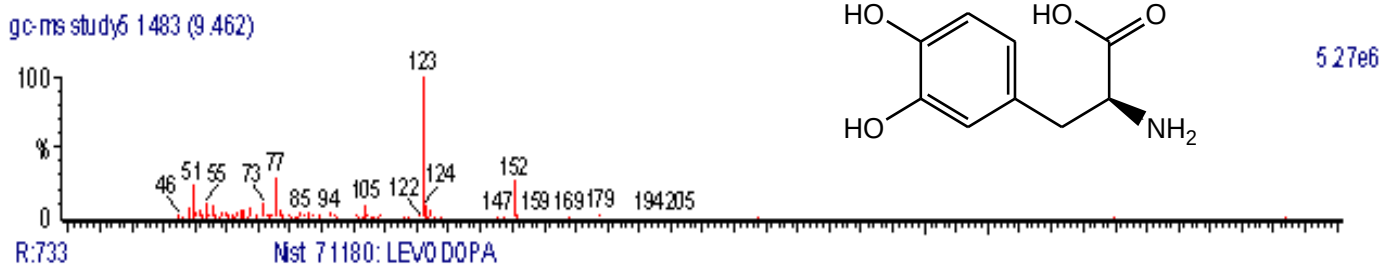

Fig. 3: Mass spectrum of Levodopa (RT: 9.46)

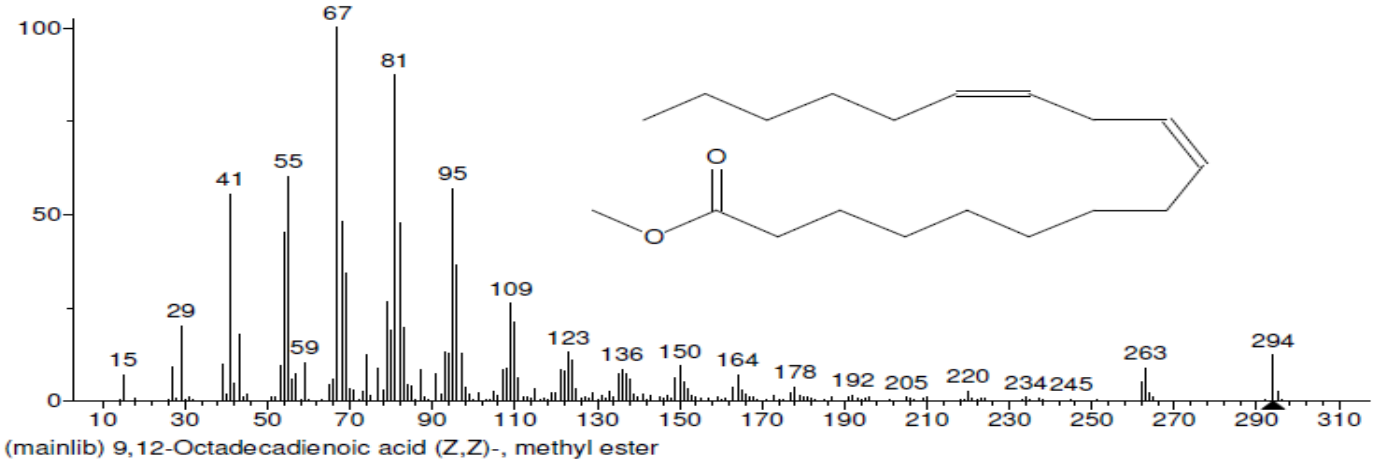

Fig. 4: Mass spectrum of (9E, 12E)- methyl octadeca-9, 12-dienoate (RT: 15.27)

Thus, GC-MS analysis of the ethanolic extract of the bark of *Bauhinia tomentosa* L. is the first step towards understanding the nature of active principles in this medicinal plant and this type of study will be helpful for further detailed phytochemical study. The investigation concluded that the stronger extraction capacity of ethanol could have produced number of active constituents responsible for many biological activities.

In the present study fourteen chemical constituents have been identified from ethanolic extract of the bark of *Bauhinia tomentosa* L. by GC-MS analysis. The presence of various bioactive compounds justifies the use of the bark for various ailments by traditional practitioners. However isolation of individual phytochemical constituents and subjecting it to biological activity will definitely give fruitful results. It could be concluded that the ethanolic extract of the bark of *Bauhinia tomentosa* L. contains various bioactive compounds. So it is recommended as a plant of phytopharmaceutical importance. However, further studies will need to be undertaken to ascertain fully its bioactivity, toxicity profile, effect on the ecosystem and agricultural products.

ACKNOWLEDGEMENT

Authors acknowledge the valuable help rendered by S.Kumaravel, Scientist, IICPT, Thanjavur for this analysis and validation of the results.

REFERENCES

1. Zaidan M, Rain N, Badrul A. *In vitro* screening of five local medicinal plants for antibacterial activity using disc diffusion method. Trop. Biomed. 2005; 22: 165-170.
2. Evans WC, Trease, Evan's pharmacognosy. 14th edition. WB Saunders Company Limited, UK, 1997, pp. 3.
3. Anbuselvi S, Jeyanthi Rebecca L, Sathish Kumar M, Senthilvelan T. GC-MS study of phytochemicals in black gram using two different organic manures. J Chem Pharma Res. 2012; 4(2): 1246-1250.
4. Makky EA, Mashitah M, Yusoff MM, Ibrahim. Impact of Medicinal Plants Phytocomponents against Antibiotic Resistant Bacteria. J Chem Pharma Res. 2012; 4(1): 881-893.
5. Kavitha R, Kamalakannan P, Deepa T, Elamathi R, Sridhar S, Suresh Kumar J. *In vitro* Antimicrobial Activity and Phytochemical Analysis of Indian Medicinal Plant *Couroupita guianensis* Aubl. J Chem Pharma Res. 2011; 3(6): 115-121.
6. Meurer-Grimes B, Mcbeth DL, Hallihan B, Delph S. Antimicrobial activity in medicinal plants of the *Scrophulariaceae* and *Acanthaceae*. Int J Pharmacog. 1996; 34: 243-248.
7. Koduru S, Grierson DS, Afolayan AJ. Antimicrobial activity of *Solanum aculeastrum*. Pharmaceul Biol. 2006; 44: 283-286.
8. Dhanapal V, Maheswari S, Premjanu N. Pharmocognostical and Phytochemical evaluation of Stem ff *Abutilon indicum*. Int J Pharma Sci Res. 2014; 5(6): 286-290.
9. Merlin NJ, Parthasarathy V, Manavalan R, Kumaravel S. Chemical Investigation of Aerial Parts of *Gmelina asiatica* Linn by GC-MS. Pharmacog Res. 2009; 1(3): 152-156.
10. Sahaya SS, Janakiraman N, Johnson M. Phytochemical analysis of *Vitex altissima* L. using UV-Vis, FTIR and GC-MS. Int J Pharm Sci Drug Res. 2012; 4(1):56-62.
11. Gopalakrishnan S, Vadivel E. Antibacterial and Antifungal activity of the bark of *Bauhinia tomentosa* L. Pharma Science Monitor. 2011; 2(3): 103-109.
12. Gopalakrishnan S, Vadivel E. Pharmacognostical and Physico-chemical analysis of the bark of *Bauhinia tomentosa* L. E-J Chem. 2012; 9(2): 1022-1028.
13. Gopalakrishnan S, Vadivel E. GC-MS Analysis of some bioactive constituents of *Mussaenda frondosa* Linn. Int J Pharma and Bio Sci. 2011; 2(1): 313-320.
14. Manyam BV, Dhanasekaran M, Hare TA. Neuroprotective effects of the antiparkinson drug *Mucuna pruriens*. Phytother Res. 2004; 18: 706-712.
15. Senthilkumar V, Sathishkumar G, Sivaramakrishnan S, Sujatha K, Razia M. Evaluation of phytoconstituents, *In vitro* antioxidant and antimicrobial activities of *Agaricus bisporus*. Int J Pharmacy Pharmaceu Sci. 2016; 8(3), 67-71.

Source of Support: Nil, Conflict of Interest: None declared.