

GAS CHROMATOGRAPHY AND MASS SPECTROSCOPIC ANALYSIS OF *Caesalpinia pulcherrima* FLOWER EXTRACT

Chemistry

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ABSTRACT

The aim of this study was to carry out for identification of bioactive compounds from the methanolic extract of *Caesalpinia pulcherrima* flower by Gas chromatography and Mass spectroscopy (GC-MS). GCMS analysis of methanolic extract was done by standard protocol using the equipment Perkin-Elmer Gas Chromatography–Mass Spectrometry, while the mass spectra of the compounds found in the extract was matched with the National Institute of Standards and Technology (NIST) library. The GC-MS analysis revealed the presence of various compounds like Tetradecane, 1,2-Benzenedicarboxylic acid mono (phenylmethyl) ester and Hexadecanoic acid, 2-hydroxy methyl ester in the methanolic extract of *Caesalpinia pulcherrima*. These findings support the traditional use of *Caesalpinia pulcherrima* in various disorders.

KEYWORDS

Gas chromatography and Mass spectroscopy, *Caesalpinia pulcherrima*, Phytochemistry.

INTRODUCTION

Plant products have been part of phytomedicines since time immemorial. These can be derived from any part of the plant like bark, leaves, flowers, roots, fruits, seeds etc., i.e. any part of the plant may contain active components. Herbal medicines have become more popular in the treatment of many diseases due to popular belief that green medicine is safe, easily available and with less side effects. Many plants are cheaper and more accessible to most people especially in the developing countries than orthodox medicine, and there is lower incidence of adverse effects after use. These reasons might account for their worldwide attention and use. The medicinal properties of some plants have been documented by some researchers (Nishaa *et al.*, 2013). Medicinal plants constitute the main source of new pharmaceuticals and healthcare products. Extraction and characterization of several active phytochemicals from these green factories have given birth to some high activity profile drugs. Indeed, the market and public demand has been so great that there is a great risk that many medicinal plants today, face either extinction or loss of genetic diversity. Knowledge of the chemical constituents of plants is desirable because such information will be value for the synthesis of complex chemical substances. Such phytochemical screening of various plants is reported by many researchers. A growing body of evidence indicates that secondary plant metabolites play critical roles in human health and may be nutritionally important (Harborne, 1973).

Caesalpinia pulcherrima (Caesalpinaceae) (English name: Barbados pride; Tamil: Mayurkonrai) (Caesalpinaceae) is a small thorny tree, 6-9m in height and 15-25 cm in diameter with a few prickly branches. It is commonly known as Patag. The tree grows wild, in mountains and is cultivated in the gardens for its large panicles of yellow flowers. The tree was formerly cultivated in South-East Asia for the red dye, obtained from its heartwood. *C. pulcherrima* is distributed in Tamilnadu, Kerala, Karnataka, Andhra Pradesh and West Bengal. The aim of this paper is to determine the organic compounds present in the *Caesalpinia pulcherrima* flower extract with the aid of GC-MS Technique, which may provide an insight in its use in traditional medicine (Varier, 1994).

MATERIAL AND METHODS

Plant materials:

The flowers of *Caesalpinia pulcherrima* were collected from Thanjavur, Thanjavur District, Tamil Nadu, India from a single tree.

Preparation of extracts:

The *Caesalpinia pulcherrima* flowers were first washed well and dust was removed from the flower. Then the flowers were dried at room temperature and coarsely powdered. The powder was extracted with methanol for 24 hours. A semi solid extract was obtained after complete elimination of alcohol under reduced pressure. The extract was stored in desiccator until used. The percentage yield was 4.50%

(4g gives 300mg extract).

GC-MS analysis

GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer instrument employing the following conditions: column Elite-1 fused silica capillary column (30 x 0.25mm ID x 1µMdf, composed of 100% Dimethyl polydioxane), operating in electron impact mode at 70eV; Helium gas (99.999%) was used as carrier gas at a constant flow of 1 ml/min and an injection volume of 0.5µl 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, then 5°C/min to 280°C, ending with a 9min isothermal at 280°C. Mass spectra were taken at 70eV; a scan interval of 0.5 seconds and fragments from 40 to 450 Da. Total GC running time is 36min. min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adopted to handle mass spectra and chromatograms was a TurboMass Ver 5.2.0

RESULTS AND DISCUSSION

Twenty compounds were identified in *Caesalpinia pulcherrima* flower by GC-MS analysis. The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) are presented in (Table 1 and Fig 1). The prevailing compounds were Tetradecane, 1,2-Benzenedicarboxylic acid mono (phenylmethyl) ester and Hexadecanoic acid, 2-hydroxy methyl ester.

Among the identified phytochemicals hexadecanoic acid is suggested to be a fatty acid ester and it may employed as antioxidant, antimicrobial, flavor, hypocholesterolemic agent and larvicidal activities (Bodoprost and Rosemeyer, 2007; Falodun *et al.*, 2009).

Compounds like n-hexadecanoic acid, 12-octadecanoic acid, dodecanoic acid, tetradecanoic acid, 1, 2-benzene dicarboxylic acid, butyl octyl ester, hexadecanoic acid, ethyl ester and 9,12-octadecadienoic acid (Z,Z) were identified in the ethanolic leaf extract of *Vitex altissima*, a Verbenaceae member (Sathish *et al.*, 2012). Likewise, hexadecane, dodecanoic acid, nonadecane, eicosane, tetradecanoic acid, oleic acid, heptacosane, 9,12- octadecenoic acid, ethyl ester; n-hexadecanoic acid; 1,2-benzenedicarboxylic acid and 9-octadecenoic acid (Z)-ethyl ester were reported in *Clerodendrum inermis* and *C. phlomidis* leaves (Anandhi and Ushadevi, 2013; Balaji and Kilimozhi, 2014).

Similar results were also observed in the leaves of *Gmelina asiatica* which showed Pregnane – 3,11, 12,14,20 – pentol, 3,12, 20, triacetate 11 (hydroxyacetate), (3a, 11a, 12a, 14a), Tridecanoic acid, methyl ester, 10-Octadecanoic acid, methyl ester, 16-Octadecanoic acid, methyl ester, 2,7- Diphenyl-1,6-dioxopyridazino (4,5:2,3) pyrrolo (4,5,-d) pyridazine, spiro (androstane-3,2- thiazolidine) were

anthelmintic, Anti- Inflammatory and Anti-microbial activities and anti-cancerous activity of the leaf extract (Azhagumurugan and Rajan, 2014).

Similarly Merlin *et al.* (2009) identified twenty-two chemical compounds from the chloroform extract of *G. asiatica* aerial parts, of which six compounds were similar to that of the results obtained in the present study. The compounds were 1, 2 benzene dicarboxylic acid, diisooctyl ester, benzoic acid, 2-hydroxy, phenyl ester; n-hexadecanoic acid; octadecanol, 2-bromo; octadecanoic acid and nonadecane.

Conclusion

In the present study twenty chemical constituents have been identified from Methanolic extract of the whole plant of *Caesalpinia pulcherrima* flower by Gas Chromatogram Mass spectrometry (GC-MS) analysis. The presence of various bioactive compounds justifies the use of flower various ailments by traditional practitioners.

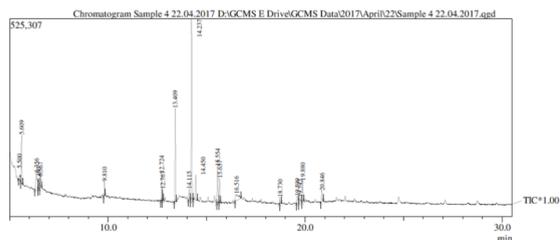


Figure 1: Chromatogram obtained from the GC/MS with the extract of *Caesalpinia pulcherrima*

Table 1 Shows the components identified in methanolic extract of *Caesalpinia pulcherrima*. (GC MS study)

Peak#	R.Time	Area%	Height %	Molecular formula	Name of the compounds
1	5.500	1.51	0.65	C ₆ H ₇ D ₅ O ₂	1-Isopropoxyacetone
2	5.609	7.23	8.41	C ₁₀ H ₁₆	Bicyclo[3.1.1]Hept-2-Ene, 2,6,6-Trimethyl-
3	6.356	5.94	2.84	C ₆ H ₇ [15N]	[15N]-Aniline
4	6.467	1.58	1.31	C ₉ H ₁₈ O ₃	Carbonic Acid, Dibutyl Ester
5	6.567	02.18	2.01	C ₁₁ H ₂₀ O ₄	Oxalic acid, isobutyl pentyl
6	9.810	1.90	2.55	C ₁₄ H ₃₀	Tetradecane
7	12.724	4.02	4.80	C ₁₅ H ₂₄	Gamma. 1-Cadinene
8	12.767	1.31	2.04	C ₁₁ H ₂₄	Nonane, 3,7-Dimethyl-
9	13.409	13.56	16.41	C ₁₅ H ₂₄	Bicyclo[7.2.0]Undec-4-Ene, 4,11,11-Trimethyl-8-Methylene-, [1r-(1r*,4e,9s*)]-
10	14.115	1.57	1.89	C ₁₅ H ₂₄	2-Norpinene, 2,6-Dimethyl-6-(4-Methyl-3-Pentenyl)-, Trans(-)-
11	14.237	27.35	31.83	C ₁₅ H ₂₄	1,6-Cyclodecadiene, 1-Methyl-5-Methylene-8-(1-Methylethyl)-, [S-(E,E)]-
12	14.450	4.86	4.47	C ₁₀ H ₁₆	Santolina triene
13	15.554	7.21	6.40	C ₂₀ H ₃₆ O ₄	Phthalic acid, di-(1-hexen-5-yl) ester
14	15.657	4.14	4.32	C ₁₆ H ₂₈	4-Hexadecen-6-yne, (E)-
15	16.516	4.63	1.21	C ₇ H ₁₀ D ₆	1,7-D6-Heptane
16	18.730	1.04	1.09	C ₁₈ H ₃₈ O	Bis-(3,5,5-Trimethylhexyl) Ether
17	19.599	2.26	1.21	C ₈ H ₁₈ O	4-Octanol

18	19.742	1.58	0.93	C ₁₅ H ₁₂ O ₄	1,2-Benzenedicarboxylic acid, mono(phenylmethyl) ester
19	19.880	3.73	3.70	C ₁₈ H ₃₆ O ₂	Hexadecanoic Acid, Ethyl Ester
20	20.846	2.41	1.93	C ₃₆ H ₇₅ O ₃ P	Phosphonic Acid, Dioctadecyl Ester
		100.00	100.00		

Table 2: Activity of phyto-components identified in the methanolic extract of the *Caesalpinia pulcherrima* by GC-MS.

PEAK NO.	R.TIME	HEIG HT%	NAME OF THE COMPOUNDS	BIOLOGICAL ACTIVITY**
1	9.810	1.90	Tetradecane	Antimicrobial activity
2	19.742	1.58	1,2-Benzenedicarboxylic acid, mono(phenylmethyl) ester	Antimicrobial, Antifouling, Plasticizers
3	19.880	3.73	Hexadecanoic acid, 2-hydroxy-, methyl ester	Antioxidant, Flavor, Hypocholesterolemic Nematicide, Pesticide, Lubricant, Antiandrogenic, Hemolytic, 5-Alpha reductase inhibitor

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