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Chemical Investigation of Aerial Parts of *Gmelina asiatica* Linn by GC-MS

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ABSTRACT

This study was carried out to analyze the active constituents present in aerial parts of *Gmelina asiatica* Linn. (*Verbenaceae*). Twenty-two compounds in chloroform extract and 12 compounds in ethanolic extract were identified by Gas Chromatography - Mass Spectrometry (GC-MS) analysis. 1, 2-Benzenedicarboxylic acid, diisooctyl ester (31.22 %) was the prevailing compound in chloroform extract and Monolinoleoylglycerol trimethylsilyl ether (38.51%) was the major constituent of ethanolic extract. This is the first report of identification of active constituents from aerial parts of *Gmelina asiatica* by GC-MS.

Keywords: *Gmelina asiatica* (*Verbenaceae*), GC-MS, 1,2-Benzenedicarboxylic acid diisooctyl ester, Monolinoleoylglycerol trimethylsilyl ether.

INTRODUCTION

Most traditional medicines are developed from nature. They have not yet fulfilled the scientific requirements so as to be classified as modern medicines (1-2). For purposes of scientific back up, a study is needed to examine their bioactive components, their efficacy and safety (3-4). Usually, most components that are useful for medicinal purposes are secondary metabolites (5-6).

In the development of medicinal plant industry, plant medicines are classified into three groups: herbs (Jammu), standardized extracts and phytopharmaceuticals (7). There are strict requirements for standardizing the extracts. Some of them include correctness and proven restorative power, uniformity of active constituents, their efficacy, safety and assurance, both in quality and quantity (8-9).

Gmelina asiatica L (*Verbenaceae*) popularly known as *Nilakkumil* in Tamil and *Gopabhandra* in Sanskrit, is a large straggling shrub found in South India. Its roots are used for the treatment of gonorrhoea, catarrh of the bladder, rheumatism and for purification of the blood (10). Since this plant is claimed to be useful in the treatment of rheumatism, it is said to possess anti-inflammatory action (11). The root of the plant also has potent hypoglycaemic activity (12). The aerial

parts of *Gmelina asiatica* have antimicrobial activity (13). Extensive phytochemical studies have been carried out in roots but thorough phytochemical work has not been carried out in the aerial parts of this plant. Since there are no reports on the phytochemical aspects of *Gmelina asiatica* aerial parts, it was chosen as the subject for this study. The aim of this paper is to validate a rapid method for the quantitative determination of organic compounds in the aerial parts of *Gmelina asiatica* using rapid fingerprint procedure.

MATERIALS AND METHODS

Plant material

Aerial parts of the *Gmelina asiatica* were collected from Therkkumalai Estate, Courtallam Hills, Western Ghats, Tamil Nadu, India and identified by Dr.V.Chelladurai, Research Officer, Central Council for Research in Ayurveda and Siddha, Palayamkottai. Voucher specimens were prepared and preserved in the Department of Pharmacognosy, KMCP, Madurai for further reference (Voucher specimen no. KMCP/GA/23).

Preparation of extract

Aerial parts of *Gmelina asiatica* were shade dried and defatted with petroleum ether. The defatted material was extracted using soxhlet apparatus with chloroform

and 95% ethanol. The extracts were then filtered through Whatmann filter paper No.41 along with 2gm sodium sulfate to remove the sediments and traces of water in the filtrate. Before filtering, the filter paper along with sodium sulphate was wetted with chloroform and 95% ethanol respectively. The filtrate was then concentrated by bubbling nitrogen gas into the solution. The extract contained both polar and non-polar phytochemicals of the plant material used. 2 µl sample of these solutions was employed for GC/MS analysis.

GC 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 (GC-MS) instrument employing the following conditions: column Elite-1 fused silica capillary column (30mm×0.25mm ID ×1µM 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 1ml/min and an injection volume of 0.5 µl 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 9 min isothermal at 280 °C. Mass spectra were taken at 70 eV; a scan interval of 0.5 seconds and fragments from 40 to 550 Da.

Identification of Components

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns.

The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight and structure of the components of the test materials was ascertained.

RESULTS AND DISCUSSION

The compounds present in the chloroform and ethanolic extracts of *Gmelina asiatica* were identified by GC-MS analysis (Figures 1 and 2). The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) in the chloroform and ethanolic extracts of *Gmelina asiatica* are presented in Tables 1 and 2. Twenty-two compounds were identified in chloroform extract. The prevailing compound in chloroform extract was 1,2-Benzenedicarboxylic acid, diisooctyl ester (31.22 %). Figures 3 and 4 show mass spectrum and structures of this compound which is suggested to be a plasticizer compound and is used as an antimicrobial and antifouling agent.

Nine compounds were identified in ethanolic extract of the plant. Monolinoleoylglycerol trimethylsilyl ether, a steroid compound was identified by GC-MS analysis as the main constituent (38.51%) of ethanolic extract. Figures 5 and 6 show the mass spectrum and structure of this compound, which is suggested to be a steroid compound and is used as an antimicrobial, anti-inflammatory, anti-arthritic, anti-diuretic and an anti-asthmatic agent. Based on the chemical nature and percentages of compounds present in the chloroform and ethanolic extracts, they may contribute to varying pharmacological activities.

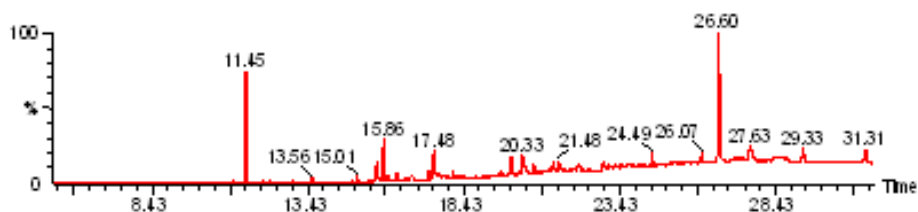


Fig 1. GC-MS chromatogram of chloroform extract of *Gmelina asiatica*

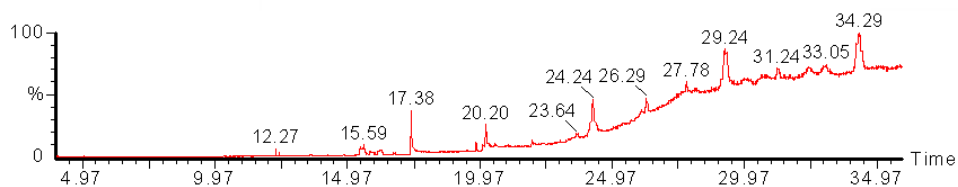


Fig 2. GC-MS chromatogram of ethanolic extract of *Gmelina asiatica*

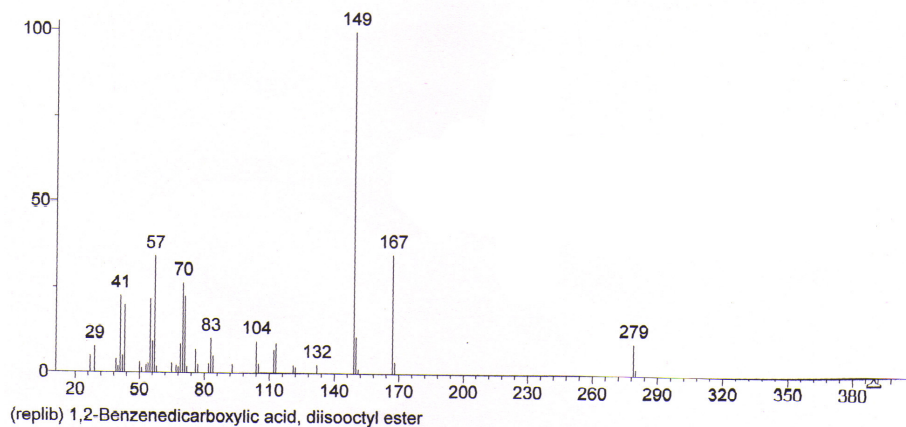


Fig. 3. Mass spectrum of 1,2-Benzenedicarboxylic acid, diisooctyl ester

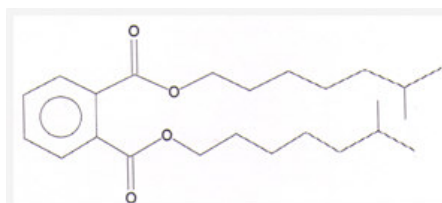


Fig. 4. Structure of 1,2-Benzenedicarboxylic acid, diisooctyl ester

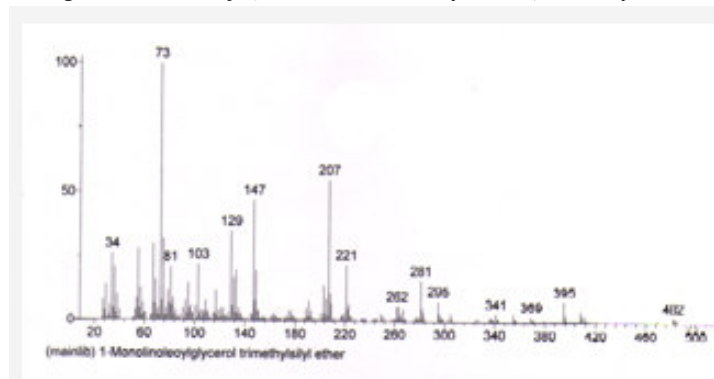


Fig. 5. Mass spectrum of 1-Monolinoleoylglycerol trimethylsilyl ether

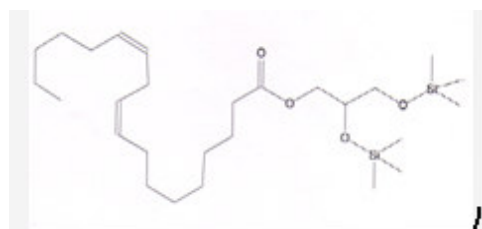


Fig. 6. Structure of 1-Monolinoleoylglycerol trimethylsilyl ether

Table 1. Phytocomponents in the chloroform extract of *Gmelina asiatica* [GC MS study]

No	RT	Name of the compound	Molecular Formula	MW	Composition %
1	6.91	Decane, 4-methyl-	C ₁₁ H ₂₄	156	0.01
2	9.64	Hydroxylamine, O-decyl-	C ₁₀ H ₂₃ NO	173	0.04
3	10.92	Isooctane, (ethenyloxy)-	C ₁₀ H ₂₀ O	156	0.05
4	11.09	Phenol, 2,4-bis(1,1-dimethylethyl)-	C ₁₄ H ₂₂ O	206	0.20
5	11.45	Benzoic acid, 2-hydroxy-, pentyl ester	C ₁₂ H ₁₆ O ₃	208	11.75
6	11.99	Diethyl Phthalate	C ₁₂ H ₁₄ O ₄	222	0.23
7	12.20	Hexadecane	C ₁₆ H ₃₄	226	0.45
8	13.56	Nonadecane	C ₁₉ H ₄₀	268	6.06
9	15.64	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	C ₂₀ H ₄₀ O	296	4.26
10	16.30	3-Eicosyne	C ₂₀ H ₃₈	278	1.88
11	16.77	7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione	C ₁₇ H ₂₄ O ₃	276	1.05
12	17.48	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256	5.53
13	17.64	Phthalic acid, butyl undecyl ester	C ₂₃ H ₃₆ O ₄	376	0.82
14	18.10	Methoxyacetic acid, 2-tridecyl ester	C ₁₆ H ₃₂ O ₃	272	1.04
15	19.94	Phytol	C ₂₀ H ₄₀ O	296	4.49
16	20.33	9,12-Octadecadienoic acid (Z,Z)-	C ₁₈ H ₃₂ O ₂	280	7.31
17	20.67	Octadecanoic acid	C ₁₈ H ₃₆ O ₂	284	2.55
18	21.48	Octadecanal, 2-bromo-	C ₁₈ H ₃₅ BrO	346	1.87
19	24.49	tert-Hexadecanethiol	C ₁₆ H ₃₄ S	258	2.87
20	26.60	1,2-Benzenedicarboxylic acid, diisooctyl ester	C ₂₄ H ₃₈ O ₄	390	31.22
21	27.63	Heptacosane	C ₂₇ H ₅₆	380	5.46
22	29.33	1-Monolinoleoylglycerol trimethylsilyl ether	C ₂₇ H ₅₄ O ₄ Si ₂	498	4.69

Table 2: Phytochemicals in the Ethanolic extract of *Gmelina asiatica* [GC MS study]

No	RT	Name of the compound	Molecular Formula	MW	Composiiton %
1	3.60	Butane, 1,1-diethoxy-2-methyl-	C ₉ H ₂₀ O ₂	160	0.20
2	12.27	1H-Cycloprop[e]azulen-7-ol, decahydro-1,1,7-trimethyl-4-methylene-, [1ar- (1aà, 4aà, 7á, 7aá, 7bà)]- (Synonym: Spathulenol)	C ₁₅ H ₂₄ O	220	1.08
3	12.38	1H-3a,7-Methanoazulene, octahydro-1,4,9,9-tetramethyl-(Synonyms: Patchoulane)	C ₁₅ H ₂₆	206	0.61
4	15.59	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	C ₂₀ H ₄₀ O	296	3.87
5	17.38	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256	14.65
6	19.82	Phytol	C ₂₀ H ₄₀ O	296	2.42
7	20.20	9,12-Octadecadienoyl chloride, (Z,Z)-	C ₁₈ H ₃₁ ClO	298	11.08
8	24.24	Heptacosane 1-Monolinoleoylglycerol	C ₂₇ H ₅₆	380	27.58
9	29.24	trimethylsilyl ether	C ₂₇ H ₅₄ O ₄ Si ₂	498	38.51

CONCLUSION

This investigation has helped to identify the compounds present in the aerial parts of *Gmelina asiatica*, a hitherto uninvestigated species. Evaluation of pharmacological activity in the chloroform and ethanolic extracts is in progress.

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