ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

GC-MS analysis of bioactive components of *Hugonia mystax L*. (Linaceae)

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ABSTRACT

Hugonia mystax L. belongs to the family Linaceae. It is commonly known as "Modirakanni". The present investigation was carried out to determine the possible bioactive components of leaves of *Hugonia mystax* L. using GC-MS analysis. Thirteen compounds were identified. 1,2-Benzenedicarboxylic acid, diisooctyl ester (48.75%) was found to be major component followed by n- Hexadecanoic acid (13.52%), Phytol (9.25%), Squalene (6.41%), Vitamin E (4.09%), Dianhydromannitol (3.56%), 9,12 – Octadecadienoic acid (Z,Z) – (3.20%) and 3,7,11,15 – tetramethyl -2- hexadecen -1-ol (2.85%).

Keywords: Modirakanni, GC-MS, bioactive compounds, Phytol

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ISSN: 0975-8585

INTRODUCTION

The genus *Hugonia* L. of family Linaceae comprise about 40 species in the world; of which *Hugonia mystax* L. was reported from India [1-2]. This plant *Hugonia mystax* is locally known as Modirakanni. Ethnobotanically, the fruits are used by the tribals of Kalakad Mundanthurai for the treatment of Rheumatism [3]. Roots were used as anthelmintic, astringent and also used for dysentery, snake bite, fever, inflammation and rheumatism. Biological activities such as analgesic, anti-inflammatory and ulcerogenic were reported [4-8]. Roots of *Hugonia mystax* [9] were evaluated for preliminary phytochemical screening and antimicrobial activity. Preliminary phytochemical screening showed the presence of various classes of secondary metabolites such as flavonoids, phenols, saponins, steroids, tannins and terpenoids. Antimicrobial activity of petroleum ether, chloroform, ethanol and aqueous extracts of root extracts showed significant activity against various human pathogens.

Fruits of *Hugonia mystax* were examined for preliminary phytochemical screening and antimicrobial activity. Preliminary phytochemical screening showed the presence of various classes of secondary metabolites such as flavonoids, phenols, saponins, steroids, tannins and terpenoids. Antimicrobial activity of petroleum ether, chloroform, ethanolic and aqueous fruits extracts showed significant activity against the human pathogens such as *Streptococcus pneumoniae* causing brain abscesses, pneumonia and septic arthritis; *Proteus vulgaris*, *Pseudomonas aeruginosa* causing urinary tract infections and septicaemia; *Salmonella typhi* causing typhoid fever, *Vibrio* species causing diarrheal infections and the fungus *Candida albicans* causes urinary tract infections [9].

Perusal of literature reveals that information on the GC-MS analysis of leaves of *Hugonia mystax* is totally lacking. Hence, the objective of the present study is to identify the phytochemical constituents with the aid of GC-MS technique.

MATERIALS AND METHODS

Collection of plant sample

Leaves of *Hugonia mystax* were collected from Kothagiri, Nilgiri Biosphere Reserve, Western Ghats, Tamil Nadu. With help of local flora, voucher specimen were identified and preserved in the Ethnopharmacology unit, Research Department of Botany, V. O. Chidambaram College, Tuticorin, and Tamil Nadu for further references.

Plant sample extraction

The leaves were cleaned, shade dried and pulverized to powder in a mechanical grinder. Required quantity of powder was weighed and transferred to Stoppard flask, and treated with ethanol until the powder is fully immersed. The flask was shaken every hour for the first 6 hours and then it was kept aside and again shaken after 24 hours. This process was repeated for 3 days and then the extract was filtered. The extract was collected and evaporated to dryness by

using a vacuum distillation unit. The final residue thus obtained was then subjected to GC-MS analysis.

GC-MS Analysis

GC-MS analysis of these extracts were performed using a Perkin-Elmer GC Clarus 500 system and Gas chromatograph interfaced to a Mass spectrometer (GC-MS) equipped with a Elite-I, fused silica capillary column (30mmX0.25mm 1D X 1 µ Mdf, composed of 100% Di methyl poly siloxane). For GC-MS detection, an electron ionization system with ionizing energy of 70 eV was used. Helium gas (99.999%) was used as the carrier gas at constant flow rate of 1ml/min and an injection volume of 2µl 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 70 eV; a scan interval of 0.5seconds and fragments from 45 to 450 Da. Total GC running time was 36 minutes. The relative % 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 Turbo mass.

Identification of Compounds

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute of 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 were ascertained.

RESULTS AND DISCUSSION

The components present in the ethanol extract of leaves of *Hugonia mystax* were identified by GC-MS analyzed (Figure 1). The active principals with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) in the ethanol extract of leaves of *Hugonia mystax* are presented in Table 1. Thirteen components were detected in ethanol extract of *Hugonia mystax* leaves. The results revealed that 1,2-Benzenedicarboxylic acid and Diisooctyl ester (48.75%) were found to be major components followed by n-Hexadecanoic acid (13.52%), Phytol (9.25%), Squalene (6.41%), Vitamin E (4.09%), Dianhydromannitol (3.56%), 9,12 – Octadecadienoic acid (Z,Z) – (3.20%) and 3,7,11,15 – Tetramethyl -2- hexadecen -1.ol (2.85%). Figure 2,3,4,5 and 6 shows the mass spectrum and structure of Hexadecanoic acid, Ethyl ester, 11, 14, 17-Eicosatrienoic acid, Phytol, Squalene, Vitamin E. Table 2 listed the major phytocomponents and its biological activities obtained through GC-MS study of *Hugonia mystax.*

Among the identified phytochemicals, n-Hexadecanoic acid and vitamin E may have the role in antioxidant and antiinflammatory effects [10], Squalene have the property of antioxidant

ISSN: 0975-8585

[11]. Recently Squalene possesses chemopreventive activity against colon carcinogenesis [12]. Phytol is detected in *Hugonia mystax* leaves which were also found to be effective at different stages of the arthritis. It was found to give food as well as preventive and therapeutic results against arthritis. The results show that, reactive oxygen species –promoting substances such as phytol constitute a promising novel class of pharmaceuticals for the treatment of rheumatic arthritis and possibly other chromic inflammatory diseases [13].

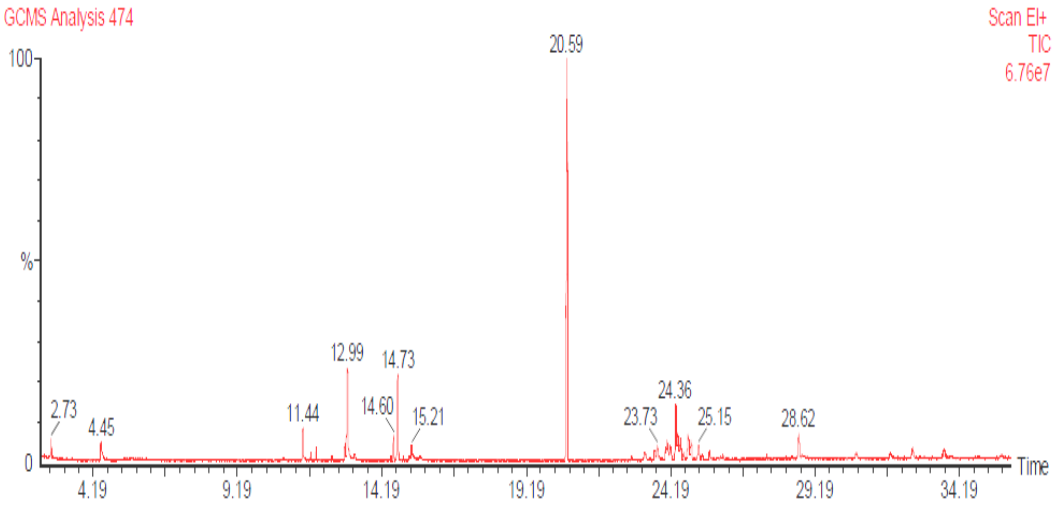

Figure 1: GC-MS chromatogram of the ethanol extract of the leaf of *Hugonia mystax*

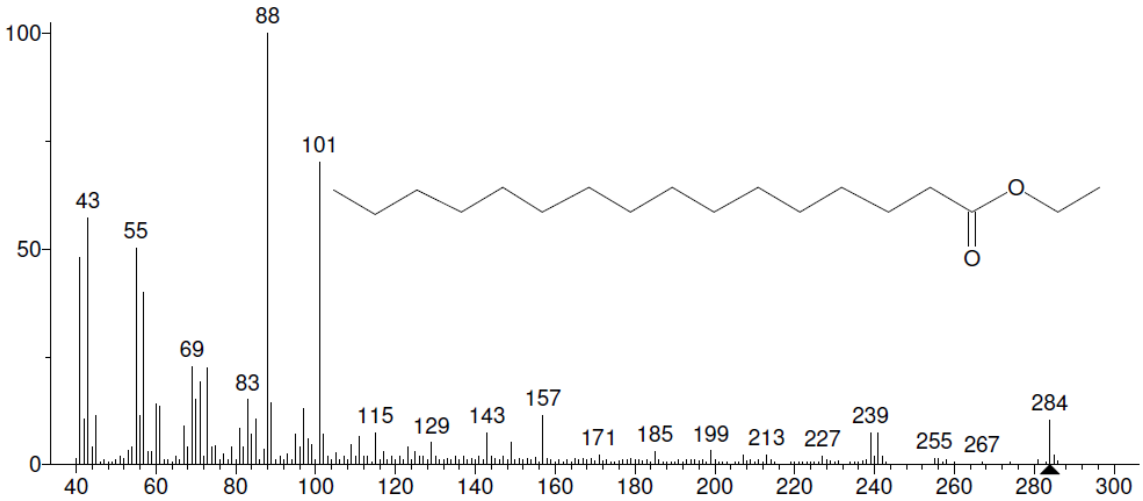

Figure 2: Mass spectrum of hexadecanoic acid, ethyl ester.

ISSN: 0975-8585

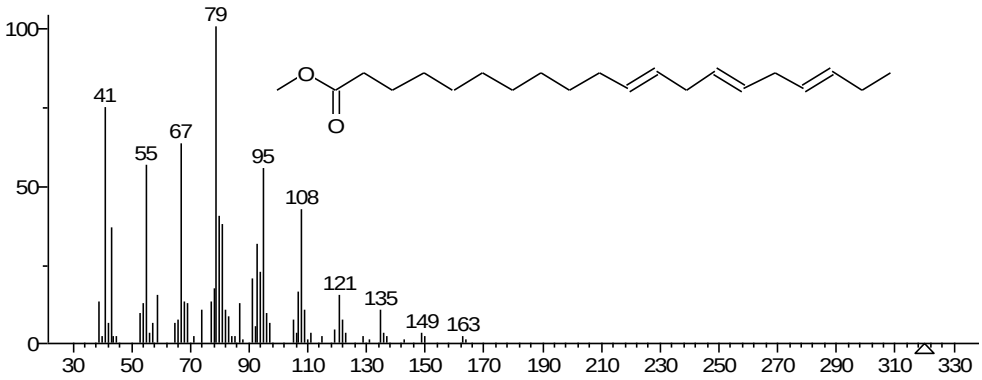

Figure 3: Mass spectrum of 11, 14, 17-Eicosatrienoic acid, methyl ester.

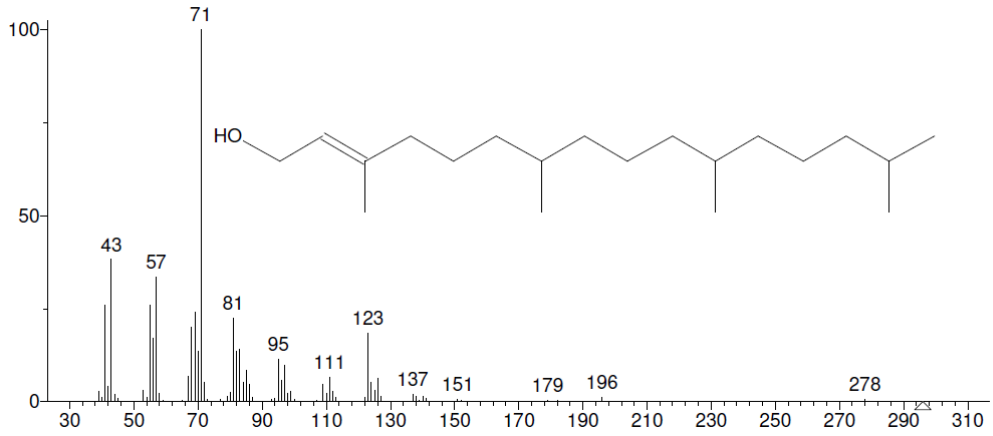

Figure 4: Mass spectrum of phytol

In the present study, thirteen compounds have been identified from ethanol extract of the leaves of *Hugonia mystax* by GC-MS analysis. The presence of various bioactive compounds justifies the use of the leaf for various ailments by traditional practitioners. So it is recommended as a plant of phytopharmaceutical importance. However further studies will need to be undertaken to ascertain fully its bioactivity.

Table 1. Components detected in the leaf of ethanol extract of *Hugonia mystax*

No.	RT	Name of the compound	Molecular formula	MW	Peak Area %
1	2.73	Propane, 1,1,3-triethoxy-	$C_9H_{20}O_3$	176	2.49
2	4.45	Dianhydromannitol	$C_6H_{10}O_4$	146	3.56
3	11.44	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$	296	2.85
4	11.91	1-Octadecyne	$C_{18}H_{34}$	250	1.07
5	12.89	Dibutyl phthalate	$C_{16}H_{22}O_4$	278	1.60
6	12.99	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	13.52
7	13.25	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	1.25
8	14.60	11,14,17-Eicosatrienoic acid, methyl ester	$C_{21}H_{36}O_2$	320	1.96
9	14.73	Phytol	$C_{20}H_{40}O$	296	9.25
10	15.21	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280	3.20
11	20.59	1,2-Benzenedicarboxylic acid, diisooctyl ester	$C_{24}H_{38}O_4$	390	48.75
12	24.36	Squalene	$C_{30}H_{50}$	410	6.41
13	28.62	Vitamin E	$C_{29}H_{50}O_2$	430	4.09

ISSN: 0975-8585

Table.2 Activity of phytochemical identified in the ethanol extracts of leaf of *Hugonia mystax*

No	Name of the compound	Compound Nature	Activity
1	Dianhydromannitol	Sugar alcohol	Antimicrobial
2	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	Terpene alcohol	Antimicrobial Antiinflammatory
3	Dibutyl phthalate	Plasticizer compound	Antimicrobial Antifouling
4	n-Hexadecanoic acid	Palmitic acid	Antioxidant, Hypocholesterolemic Nematicide, Pesticide,Lubricant, Antiandrogenic, Flavor, Hemolytic, 5-Alpha reductase inhibitor
5	Hexadecanoic acid, ethyl ester	Palmitic acid ester	Antioxidant, Hypocholesterolemic Nematicide, Pesticide,Lubricant, Antiandrogenic, Flavor, Hemolytic 5-Alpha reductase inhibitor
6	11,14,17-Eicosatrienoic acid, methyl ester	Unsaturated fatty acid ester	Antiarthritic Anticoronary Antiinflammatory
7	Phytol	Diterpene	Antimicrobial Anti-inflammatory Anti cancer Diuretic
8	9,12-Octadecadienoic acid (Z,Z)-	Linoleic acid	Anti-inflammatory Hypocholesterolemic Cancer preventive Hepatoprotective Nematicide Insectifuge, Antihistaminic Antieczemic Antiacne, 5-Alpha reductase inhibitor Antiandrogenic Antiarthritic Anticoronary Insectifuge
9	1,2-Benzenedicarboxylic acid, diisooctyl ester	Plasticizer compound	Antimicrobial Antifouling
10	Squalene	Triterpene	Antibacterial,Antioxidant,Antitumor, Cancer preventive, Immunostimulant, Chemo preventive, Lipoxygenase-inhibitor,Pesticide Diuretic
12	Vitamin E	Vitamin compound	Antiageing, Analgesic,Antidiabatic, Antiinflammatory, Antioxidant, Antidermatitic, Antileukemic, Antitumor, Anticancer, Hepatoprotective, Hypocholesterolemic, Antiulcerogenic, Vasodilator, Antispasmodic, Antibronchitic, Anticoronary

****Source: Dr.Duke's: Phytochemical and Ethnobotanical Databases**

ISSN: 0975-8585

Figure 5: Mass spectrum of squalence.

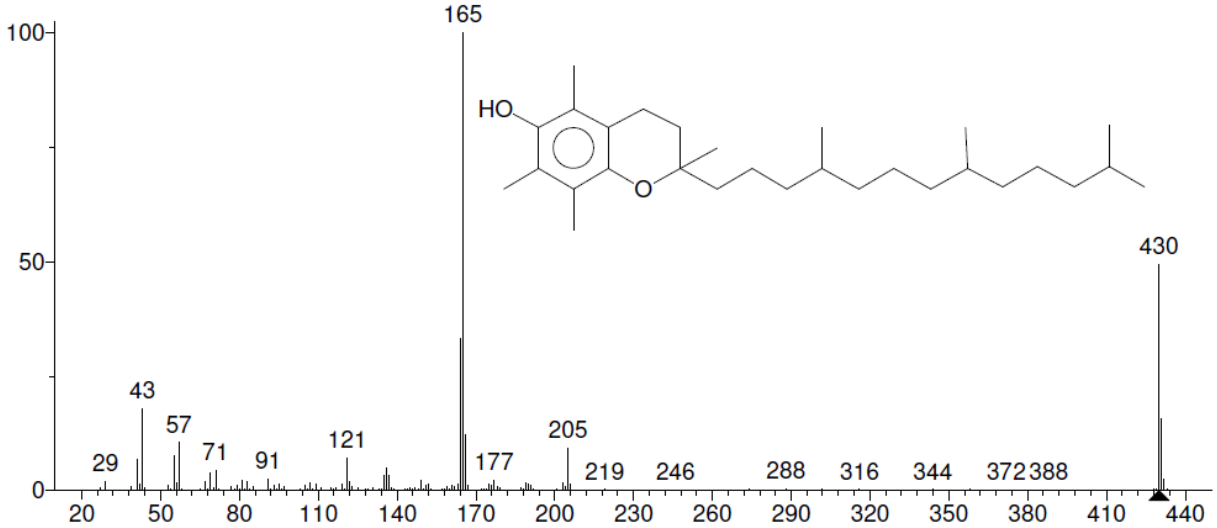

Figure 6: Mass spectrum of vitamin E.

ACKNOWLEDGEMENT

The authors are thankful to Dr.R. Sampathraj, Honorary Director, Dr. Samsun, Clinical Research Laboratory, Thirupur for providing necessary facilities to carry out this work.

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