VOLUME - 12, ISSUE - 05, MAY - 2023 • PRINT ISSN No. 2277 - 8160 • DOI : 10.36106/gjra

Original Research Paper

Botany

GC-MS ANALYSIS OF ETHANOLIC SEED EXTRACT OF HYGROPHILA AURICULATA (SCHUM) HEINE

M.Revathi* Assistant professor of Botany*Corresponding Author

R.Manonmani Assistant professor of Botany

ABSTRACT This study is aimed at determining the phytochemical constituents by using spectroscopic technique like GC-MS analysis from Hygrophila auriculata (Schum) Heine. The studies on the bioactive components of ethanolic extract of seed of Hygrophila auriculata (Schum) Heine. by GC-MS analysis clearly showed the presence of several bioactive compounds. The various compounds present in ethanolic extract of leaves were 1,3-Dioxane, 4,4-Dimethyl, 1-Tetradecene, 1-Hexadecene, 1,3-Benzodioxol,4,5-Dimethoxy-6-(-Propnyl), Tetradecanoic Acid, 1-Nonadecene, 1,2-Benzenedicarbxylicacid,Bis(2-Methoxyethyl)Ester, N-Hexadecanoi Acid, Hexadecanoic Acid, Ethyl Ester, 9,12-Octadecadienoic Acid(Z,Z)-,Methyl Ester, 9,12-Octadecadienoic Acid (Z,Z), Octadec-9-Enoic Acid, Linoleic Acid Ethyl Ester, Octadecanoic Acid Octadecanoic Acid, Ethyl Ester, Carbamic Acid, 2 (Dimethylamino)Ethylester, Oxiraneoctanoic Acid, 3-Octyl-, Cis, 13-Tetradecenal Butyl 9,12-Octadecadienoate, Squalene, z,z -8,10-Hexadecadien-1-ol, 9,12-Octadecadienoic acid(z,z), 2-Aminoethanethiol hydrogen sulfate(Ester), 9,12-Octadecadienoic acid(z,z)-2,3-Dihydroxypropyl Ester. The presence of various bioactive compounds justifies the use of the seeds of Hygrophila auriculata (Schum) Heine. for various ailments by traditional practitioners.

KEYWORDS : Hygrophila auriculata, ethanolic extract, GC-MS analysis, bioactive compounds

INTRODUCTION

Medicinal plants are the richest bioresource of drugs for traditional systems of medicine; therefore man has been using plant extracts to protect himself against several diseases and also to improve his health and life-style. The different phytoconstituents present in medicinal plants are flavonoids, alkaloids, phenols, tannins, carboxylic acids, terpenes and amino acids and inorganic acids. These phytoconstituents give specific distinctiveness and properties to plants [1].

India has a rich, vibrant and diverse cultural history. An important component of this culture and tradition is that of health and healing. India is the largest producer of medicinal herbs and is rightly called the botanical garden of the world. There are very few medicinal herbs of commercial importance, which are not found in this country. India officially recognizes over 3000 plants for their medicinal value. It is generally estimated that over 6000 plants in India are in use in traditional, folk, and herbal medicine, representing about 75% of the medicinal needs of the third world countries [2].

Phytoconstituents are the natural bioactive compounds found in plants. These phytoconstituents work with nutrients and fibers to form an integrated part of defense system against various disease and stress conditions [3].

GC is used to separate the volatile and thermally stable substitutes in a sample whereas GC-MS fragments the analyte to be identified on the basis of its mass. The further addition of mass spectrometer in it leads to GC-MS/MS. Superior performance is achieved by single and triple quadrupole modes [4-6].

Hygrophila auriculata (Schum) Heine. belonging to the family Acanthaceae. The plant contains various groups of phytoconstituents, namely, phytosterols, fatty acids, minerals, polyphenols, proanthocyanins, mucilage, alkaloids, enzymes, amino acids, carbohydrates, hydrocarbons, flavonoids, terpenoids, vitamins, and glycosides. This study was aimed at determining the phytochemical constituents using spectroscopic techniques like GC-MS analysis from *Hygrophila auriculata* (Schum) Heine.

MATERIALS AND METHODS

COLLECTION AND SOURCE OF THE PLANT MATERIAL

Seeds of the experimental plant *Hygrophila auriculata* (Schum) Heine. were collected from the herbal shop.

PREPARATION OF SEED EXTRACTS

The seeds of the plant *Hygrophila auriculata* (Schum) Heine. washed thoroughly with water, away the insects and dust particles. Then, the seeds were ground with electric grinder. A known quantity of fine powder of seeds was mixed with 500ml of ethanol for three days. The extract was filtered through Whatman No.1 filter paper. Filtrate was preserved in the air tight bottles until further use.

GC-MS ANALYSIS

GC-MS analysis was carried out on a GC Clarus 500 Perkin Elmer system and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: Column Elite-5MS fused silica capillary column (30mm x 0.25mm ID x 1 mdf, composed of 5% Diphenyl / 95% 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 2 l 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 45 to 450 Da. Total GC running time was 36 min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. The mass-detector used in this analysis was Turbo-Mass Gold-Perkin-Elmer, and the software adopted to handle mass spectra and chromatograms was a Turbo-Mass ver-5.2.

IDENTIFICATION OF COMPONENTS [7]

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 were ascertained.

RESULTS AND DISCUSSION

The studies on the bioactive components of ethanolic extract of seed of *Hygrophila auriculata* (Schum) Heine. by GC-MS

analysis clearly showed the presence of 24 bioactive compounds (Table-1).

Table-1
List of Identified phytochemicals from seed extract of *Hygrophilla auriculata* (Schum) Heine. by GC-MS analysis

RETENTION TIME	NAME OF THE COMPOUND	MOLECULAR FORMULA	MOLECULAR
4.717	1,3-DIOXANE, 4,4DIMETHYL-	C6H12O2	116.16
15.861	1-TETRADECENE	C14H28	196.37
19.996	1-HEXADECENE	C16H32	224.42
20.555	1,3-BENZODIOXOLE, 4,5-DIMETHOXY-6-(2PROPENYL)-	C12H14O4	222.24
23.209	TETRADECANOIC ACID	C14H28O2	228.37
23.72	1-NONADECENE	C19H38	266.5
26.46	1,2-BENZENEDICARBOXYLIC ACID, BIS(2-METHOXYETHYL) ESTER	C14H18O6	282.2891
26.71	N-HEXADECANOIC ACID	C16H32O2	256.4
27.08	HEXADECANOIC ACID, ETHYL ESTER	C18H36O2	284.5
28.642	9,12-OCTADECADIENOIC ACID (Z,Z)-, METHYL ESTER	C19H34O2	294.5
29.468	9,12-OCTADECADIENOIC ACID (Z,Z)-	C18H32O2	280.4
29.518	OCTADEC-9-ENOIC ACID	C18H34O2	282.5
29.665	LINOLEIC ACID ETHYL ESTER	C20H36O2	308.5
29.806	OCTADECANOIC ACID	C18H36O	284.5
30.165	OCTADECANOIC ACID, ETHYL ESTER	C20H40O2	312.5
31.472	CARBAMIC ACID, 2 (DIMETHYLAMINO)ETHYLESTER	C5H13CIN2O 2	168.6
32.203	OXIRANEOCTANOIC ACID, 3-OCTYL-, CIS-	C18H34O3	298
32.3	13-TETRADECENAL	C14H26O	210.3
35.025	BUTYL 9,12-OCTADECADIENOATE	C22H40O2	336.6
38.248	SQUALENE	C30H50	410.7
31.986	Z,z -8,10-Hexadecadien-1-ol,		
32.203	9,12-Octadecadienoic acid(z,z),	C18H32O2	280
32.618	2-Aminoethanethiol hydrogen sulfate(Ester),	C2H7NO3S2	157
36.824	9,12-Octadecadienoic acid(z,z)-2,3-Dihydroxypropyl Ester.	C21H38O4	354

The GC-MS chromatogram of the bioactive compounds detected were shown in figure-I. The various compounds present in ethanolic extract of leaves were 1,3-Dioxane, 4,4-Dimethyl, 1-Tetradecene, 1-Hexadecene, 1,3-Benzodioxol,4,5-Dimethoxy-6-(-Propnyl), Tetradecanoic Acid, 1-Nonadecene, 1,2-Benzenedicarbxylicacid,Bis(2-Methoxyethyl)Ester, N-Hexadecanoi Acid, Hexadecanoic Acid, Ethyl Ester, 9,12-Octadecadienoic Acid(Z,Z)-,Methyl Ester, 9,12-Octadecadienoic Acid (Z,Z), Octadec-9-Enoic Acid, Linoleic Acid Ethyl Ester, Octadecanoic Acid Octadecanoic Acid, Ethyl Ester, Carbamic Acid, 2 (Dimethylamino)Ethylester, Oxiraneoctanoic Acid, 3-Octyl-, Cis, 13-Tetradecenal Butyl 9,12-Octadecadienoate, Squalene, z,z -8,10-Hexadecadien-1-ol, 9,12-Octadecadienoic acid(z,z), 2-Aminoethanethiol hydrogen sulfate(Ester), 9,12-Octadecadienoic acid(z,z)-2,3-Dihydroxypropyl Ester.

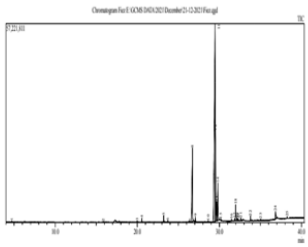

FIG-I GC-MS ANALYSIS OF ETHANOLIC SEED EXTRACT OF HYGROPHILLA AURICULATA (SCHUM) HEINE.

Our results were in accordance with the results of GC-MS analysis of leaves, stems and seeds of *Ocimum tenuiflorun*. They contain useful chemical compounds like eugenol, phytol, chlorophyll, Terpineol, -Copaene, Limiflavine, 2H-1-Benzopyran-4,7-diol, 3,4-dihydro-2-phenyl, flavone and caryophylline etc. The chemical constituents identified were responsible for various medicinal activities [8].

Similarly, the GC-MS analysis of *Calanthe masuca* revealed the presence of methanol leaf extract 20 compounds identified in the methanol extract [9].

The chromatogram of methanol plant leaf extract of *Nervilia aragoana* shows 5 prominent peaks as L-Proline, 1-Acety-($C_7H_{11}NO_3$) with retention time of 12.038 and peak area of (8.91%), Neophytadiene ($C_{20}H_{38}$) with retention time of 24.515 and peak area of (6.04 %), n-Hexadecanoic acid ($C_{16}H_{32}O_2$) with retention time of 26.666 and peak area of (15.85%), 9,12-Octadecadienoic acid, methyl ester ($C_{18}H_{32}O_2$) with retention time of 29.369 and peak area of (24.92%) and 3-Tetradecanynoic acid ($C_{14}H_{24}O_2$) with retention time of 29.455 and peak area of (26.16) [10-11].

CONCLUSION

The present study reveals that the seed extract of *Hygrophila auriculata* (Schum) Heine. consists of various bioactive compounds. The GC-MS identified phytochemicals are found reasonably responsible for the multi-therapeutic uses and effects of *Hygrophila auriculata* (Schum) Heine. in various health disorders including liver cirrhosis. The findings and results of this paper could help to evaluate and assess the therapeutic multipurpose use of *Hygrophila auriculata* (Schum) Heine. more rationally and further opened the scope for development of novel phytochemo-therapeutic drugs from the plant, which may serve as improved therapeutic agents and can create an awareness of the need of *in situ* conservation of this most wanted medicinal plant.

REFERENCES

1. Parekh, J., Chanda, S. 2008. Phytochemicals screening of some plants from western region of India. Plant Arch. 8:657-62.
2. Rajshekharan, P.E. 2002. Herbal medicine. In: World of Science, Employment News, 21-27; p. Krishnaiah, D., Devi, T., Bono, A., Sarbatly, R. 2009. Studies on

phytochemical constituents of six Malaysian medicinal plants. J Med Plants Res. 3(2):67-72. 19.
3. Krishnaiah, D., Devi, T., Bono, A. and Sarbatly, R. 2009. Studies on phytochemical constituents of six Malaysian medicinal plants. J Med Plants Res. 3(2):67-72.
4. Jenke, D.R. 1996. Chromatographic Method Validation: A review of Current Practices and Procedures. I. General Concepts and Guidelines. J Liq Chrom & Rel Technol . 19: 737-757.
5. Sahil, K., Prashant, B., Akanksha, M., Premjeet, S., Devashish, R .2011. GC-MS: Applications. International Journal Pharma & Biological Archives 2: 1544-1560.
6. Rowley, A.G. 2001 .Evaluating Uncertainty for Laboratories. A Practical Hand book. Ver 1:35-46.
7. Adams R. 1995. Identification of essential oil components by Gas Chromatography / Mass Spectroscopy. Allured Publishing Co., Carol Stream, Illinios. USA. 5: 55-60.
8. Selvaraju, R., Sakuntala, P, Kaleem Ahmed ,Jaleeli. 2020. Study on Different Parts of Ocimum tenuiflorum Plant using GC-MS, XRD and UV-Visible Methods. Int. J. Pharm. Sci. Rev. Res., 63(1), No. 14, 90-94.
9. Perraudin, F., Popovici, J., & Bertrand, C. 2006. Analysis of headspace-solid micro extracts from flower of Maxillaria tenuifolia Lindl. By GC-MS. Electronic Journal of Natural Substances. 1: 1-5.
10. Thomas, E., Aneesh, TP, Thomas, D.G., & Anandan. 2013. GC-MS Analysis of Phytochemical compounds present in the rhizomes of Nervilia aragoana gaud. Asian J Pharma and clinic Res. 6(3): 68-74.
11. Katta, J., Rampilla, V., & Mohamad, K.S. 2020. Evaluation of Phytochemical and Pharmacological aspects of epiphytic orchid Luisia zeylanica Lindl. Intern J Pharmaceu Sci and Res. 11(3): 1333-1349.