

GC-MS analysis of *Holarrhena antidysentrica* *Wall Flower*

Paramanatham, M* and. Murugesan, A.

ABSTRACT

Objective: To investigate the phytoconstituent of ethyl acetate extract of holarrhena antidysentrica using GC-MS.

Methods : GC-MS analysis of flowers extract were performed using a Thermo GC-Trace Ultra VERSION :5.0 interfaced to a Thermo Mass Spectrometer DSQ II , fused Silica Capillary Column (30mmX 0.25mm 1DX1 eMdf ,composed of 100% Dimethyl polysiloxane). **Results:** The results of the GC-MS analysis confirmed the presence of thirty compounds. The most prevailing compounds are-1-Eicosanol, Hexadecane, Sigmastane 1,3-dione, Behenic alcohol etc.

* **Conclusions:** From the results, it can be concluded that the flowers extract show the presence of 30 phyto compounds. The presence of various bioactive compounds justifies the use of the plant flower for various ailments by traditional practitioners.

Index Terms:- Green Synthesis, GCMS – Analysis, Holarrhena antidysentrica, Nano particle

1. Introduction

The natural products has act as a major role in primary health care, most of the developing countries follow the medicinal plants are for medicine to be safe and effective. The value of medicinal plants as potential source of new compounds and drug development (Bobbarala V et al., 2011;). The literature survey reveal the presence of alkaloids, tannins ,xanthoproteins .carboxylic acid, coumarins, and carbohydrates(Sharma.P et al., 2010);. Hence the modern methods describing the identification and quantification of active

Paramanatham, M * Department of Chemistry, Manonmaniam Sundaranar University, Tirunelveli, Tamil Nadu, India. Tel: 8098084525

Murugesan, A. Department of Chemistry, Sriram Engineering College, Chennai.

constituents in the plant material maybe full for proper standardization of herbals and its formulations³.

Holarrhena antidysenterica Wall. (Apocyanaceae), commonly known as “kutaja” is an important plant used in indigenous systems of medicine as remedy for bronchitis, hematuria, spermatorrhoea, epilepsy, asthma, piles, leprosy, eczema, diarrhea, fever and jaundice (Bhattacharjee, 2000; Guha Basshi et al., 2001). Various parts of *H. antidysenterica* have been reported to possess antibacterial activity (Jolly and Mechery 1996; Sujan Ganapathy et al., 2008). The bark has been reported to possess a stringent and antidiarrheal properties (Chopra et al., 1982). Leaves of the plant are used to cure scabies (Prajapati et al., 2004).

H. antidysenterica (*W.*) is commonly known as "kudappalai" in vazhaithoppu tribals of Sathuragiri hills, Western Ghats of Tamil Nadu (Maruthupandian et al 2011;). The plant have been used by Ayurvedic physicians in India for external use to treat to skin problems, dermatitis, cure the snakebites⁵.

The present study was aimed to identify the chemical constituents in Ethyl acetate flower extract of *H. antidysenterica* were analyzed by the GC-MS technique through phytochemical screening

2. Materials and methods

The flowers of *H. antidysenterica* (*W.*) were collected from the sathuragiri Biosphere Reserve, Western Ghats, Tamil Nadu. The plants were shaded dried, weighed and transferred to stoppered flask, and treated with ethyl acetate until the powder is fully immersed. The flask was shaken every hour for the first 12 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.

2.1. 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 DSQ II, fused silica capillary column (30mmX0.25mm 1D X 1 f Mdf, composed of 100% Dimethyl 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 1ml/min and an injection volume of 2fl was employed (split ratio of 10:1); Injector temperature 260 °C; Ion-source temperature 350 °C. The oven temperature was programmed from 70 C (isothermal for 2 min.), with an increase of 10 C/min, to 150 C, then 5 C/ min to 220 C, 10C/min to 260c ending with a 10 min isothermal at 350 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 38 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 Turbomass.

Interpretation on mass spectrum GC-MS was conducted using the database of the South Indian Textile Research Association (SITRA) having more than 50,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the SITRA library. The Name, Molecular weight and structure of the components of the test materials were ascertained.

1. 3. Results

The phytochemical compounds present in the ethyl acetate extract of plant flower of *H. antidysentrica* were identified by GC-MS analysis (Fig.1). The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration % in the methanol extract of plant flower of *H. antidysentrica* are presented inTable1. The prevailing compounds in ethyl acetate extract of plant flower were 1-Eicosanol(11.40%), Hexadecane(9.815), Behenic alcohol (7.71%),1-Hexadecene (CAS) (7.23%),2-(1'-Naphthyl)-6-[2"-oxo-1",5"-dimethyl-4"-phenyl-(tetrahydro)pyrazol-1"-yl]-5-(hydroxymethyl)-tetrahydropyran (6.74%) 1-Octadecene (CAS) (4.12%),17-Pentatriacontene (3.84%), Styrene (3.81%),2-Propenoic acid, 2-ethylhexyl ester(3.24%)Figure2,3and 4 shows mass spectrum and structures of dl-Citrulline,Cyclobarbital,2-Pyrrolidineacetic acid and 2-Pyrrolidinone,1-ethenyl respectively and major phytochemicals and its biological activities obtained through GC-MS study of *H. antidysentrica*

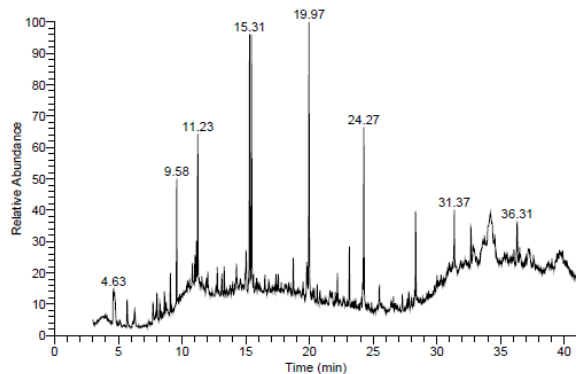


Fig.GC-MSSpectrumofHolarrhenaantidysentrica

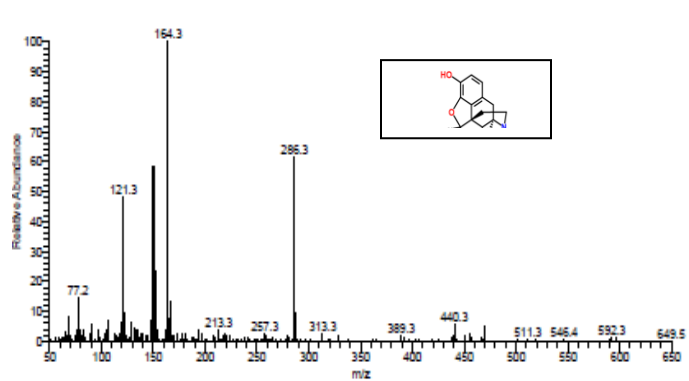


Fig2 .Mass Spectrum of Burpernorphine

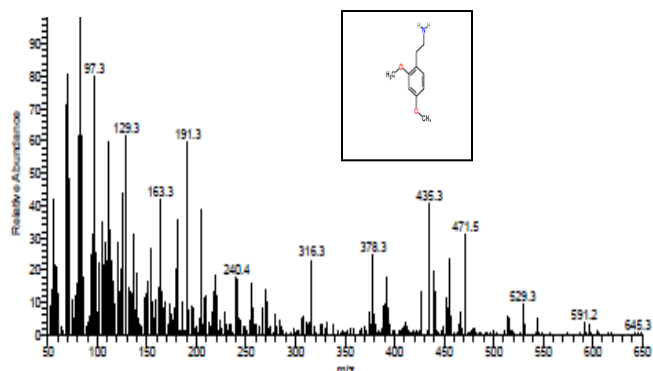


FIG2. 2,4-Dimethoxybenzene ethanamine.

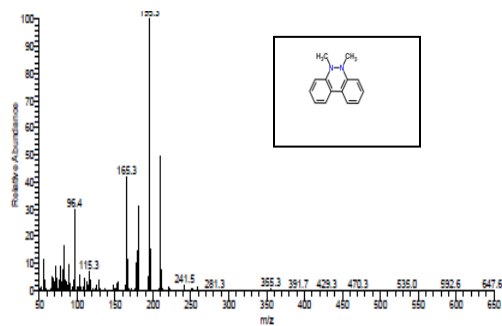


Fig3. 5,6-dihydro-5,6-dimethylbenzo[c]cinnolin

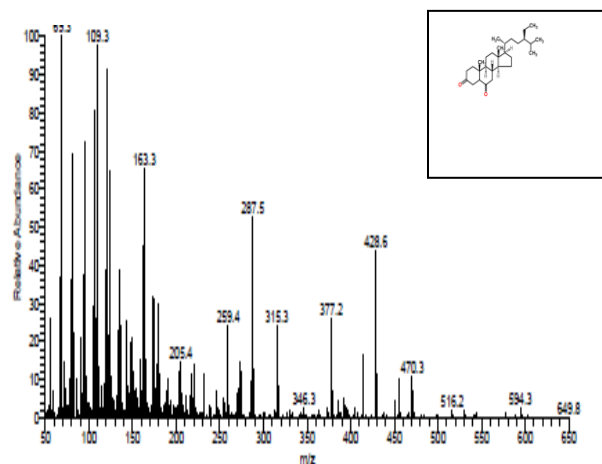


Fig4. Stigmastane-3,6-dione

Discussion

The Authentic and chemical level is a critical step in the use of these botanical ethyl acetate extract for both scientific research purposes and synthetic commercial preparations. For any living organism, identity is very important in order to distinguish itself from other organisms within the population and other populations. , during this molecular formula, the morphological characters and phytochemical properties also play a vital role in systematic studies of phytochemistry . In recent times, in addition morphological

markers, anatomical, cytological, biochemical, and molecular markers are also being used to classify the organisms. Understanding the availability of phytochemical compounds and pharmacological properties, Gas Chromatography-Mass Spectrometry (GC-MS) is a valuable tool for reliable and novel identification of phytocompounds(Sampath Kumar 20110). In the present study,30 compounds have been identified from the ethyl acetate extract of the plant flower of *Holarrhena antidysentrica* by Gas Chromatography-Mass Spectrometry analysis. 2,4-Dimethoxybenzene ethanamine, Burpernorphine, .Stigmastane-3,6-dione Among the identified phytochemicals, .

Thus this type of GC-MS analyses is the first step towards understanding the nature of active principles in this medicinal plant (Neeharika V 2012) and this type of study will be helpful for further detailed study. Further investigation into the pharmacological of *Holarrhena antidysentrica* from various solvent extracts and detailed phytochemistry may add new knowledge to the information in the traditional medical systems

Appendix

GC-MS analysis of *Holarrhena antidysentrica*

Wall Flower - Compounds detected Ethyl

Acetate flower extract table

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APPENDIX

Compounds detected Ethyl Acetate flower extract of *Holarrhena antidysentrica* Wall

| S.I | RT | Name of the compound | MF | MW | Peak Area (%) | Activity |
|-----|-------|---|----------|-----|---------------|--|
| 1. | 3.77 | Hexanedioic acid, bis(2-ethylhexyl) ester (CAS) | C22H42O4 | 370 | 2.16 | Antioxidant, antimicrobial, anti-proliferative |
| 2. | 4.65 | Styrene | C8H8 | 104 | 3.81 | Antioxidant, antibacterial, antiasthmatic |
| 3. | 5.71 | (1S,4S,8S)-8-Methoxy-1,8-dimethylbicyclo[2.2.2]oct-5-en-2-one | C11H16O2 | 180 | 1.16 | Antimicrobial |
| 4. | 6.28 | ETHYL HEXANOL | C8H18O | 130 | 1.45 | Antibiotic, antioxidant, anticholesterollemic |
| 5. | 8.03 | Cyclododecane (CAS) | C12H24 | 168 | 1.47 | Antimicrobial, antitumour |
| 6. | 9.09 | CYCLOHEXANE, (4-METHYLPENTYL)- | C12H24 | 168 | 1.17 | Antimicrobial, antidiabetic, sedative |
| 7. | 9.58 | 2-Propenoic acid, 2-ethylhexyl ester | C11H20O2 | 184 | 3.24 | Antimicrobial, antioxidant, hypotensive |
| 8. | 11.23 | 1-Hexadecene (CAS) | C16H32 | 224 | 7.23 | Antioxidant, antiasthmatic, |

| | | | | | | |
|----|-------|---|------------|-----|-------|--|
| | | | | | | antileukemic |
| 9. | 12.1 | Hexanedioic acid, mono(2-ethylhexyl)ester | C14H26O4 | 258 | 1.36 | Antimicrobial, antiulcer, cytotoxic |
| 10 | 12.76 | Cyclohexanecarboxylic acid, octyl ester | C15H28O2 | 240 | 1.48 | Antimicrobial, antiinflamaatory |
| 11 | 14.27 | 2(3H)-Benzo uranone, hexahydro-4,4,7a-trimethyl- | C11H18O2 | 182 | 1.65 | Antimicrobial, antioxidant |
| 12 | 15.02 | 1,2-Dihydronaphtho[2,1-b]furan | C12H10O | 170 | 2.08 | Antimicrobial, antioxidant |
| 13 | 15.31 | Hexadecane | C16H34 | 226 | 9.81 | Antimicrobial, antioxidant, antidiabetic |
| 14 | 16.06 | Di(2-ethylhexyl)adipate | C22H42O4 | 370 | 1.15 | Antimicrobial, anti-inflammatory |
| 15 | 19.97 | 1-Eicosanol | C20H42O | 298 | 11.40 | Antimicrobial, antioxidant |
| 16 | 21.64 | 1,10-Dimethoxy-5-carbonitrile-3-methyl-5H-benzo[b]carbazole-6-dione | C20H14N2O4 | 346 | 1.25 | Antimicrobial, oestrogenic activity |
| 17 | 22.19 | 5,6-dihydro-5,6-dimethylbenzo[c]cinnoline | C14H14N2 | 210 | 1.72 | Antimicrobial, hyaluronidase inhibitors |
| 18 | 23.13 | 10-Nonadecanone | C19H38O | 282 | 1.89 | Antimicrobial |
| 19 | 24.27 | Behenic alcohol | C22H46O | 326 | 7.71 | Antiviral |
| 20 | 25.49 | Phthalic acid, isobutyl pentadecyl ester | C27H44O4 | 432 | 1.99 | Antimicrobial |
| 21 | 28.32 | 1-Octadecene (CAS) | C18H36 | 252 | 4.12 | Anticandida |
| 22 | 30.01 | 1-Tricosanol (CAS) | C23H48O | 340 | 1.50 | Cytotoxic, antitrypanosomal |
| 23 | 30.97 | 9 Buprenorphine | C29H41NO4 | 467 | 2.04 | Antimicrobial, antitumour, anesthesia |
| 24 | 31.37 | 17-Pentatriacontene | C35H70 | 490 | 3.84 | Antibacterial, antiviral |

| | | | | | | |
|----|-------|---|-------------|-----|------|---|
| 25 | 32.69 | Dotriacontane (CAS) | C32H66 | 450 | 1.42 | Antimicrobial, antioxidant, antispasmodal |
| 26 | 33.63 | 2-{2,6-Dimethoxy-4-[(2,2-bis(ethoxycarbonyl)ethyl]phenyl}-1,10-phenanthroline | C28H28N2O6 | 488 | 2.08 | Antimicrobial, antioxidant |
| 27 | 34.21 | Stigmastane-3,6-dione, | C29H48O2 | 428 | 8.26 | Antimicrobial, anti-inflammatory, diuretic |
| 28 | 36.31 | Tricosane | C23H48 | 324 | 2.21 | Antimicrobial, antioxidant |
| 29 | 37.22 | 2,4-Dimethoxybenzeneethanamine | C10H15NO2 | 181 | 2.60 | Antimicrobial, antioxidant |
| 30 | 39.69 | 2-(1'-Naphthyl)-6-[2"-oxo-1",5"-dimethyl-4"-phenyl-(tetrahydro)pyrazol-1"-yl]-5-(hydroxymethyl)-tetrahydropyran | C27H31N2O34 | 431 | 6.74 | Antimicrobial |