GC-MS ANALYSIS OF BIOACTIVE COMPOUNDS FROM ETHANOLIC LEAF EXTRACT OF WALThERIA INDICA LINN. AND THEIR PHARMACOLOGICAL ACTIVITIES

Prabhanna Banakar* and M. Jayaraj

P. G. Department of Botany, Karnataka University, Dharwad - 580003, Karnataka, India.

ABSTRACT: Bioactive compounds are the frontline potent agents in both nutraceutical and pharmaceutical industries. Recently, the bioactive compounds are gaining much importance for their ability in enhancing resistance to various diseases and to improve the health of people both by traditional and modern ways of administrations. Therefore, in the present investigation was undertaken to identify the compounds present in the leaf of Waltheria indica Linn. by Gas Chromatography-Mass Spectrometry (GC-MS) analysis by ethanol extract. The GC-MS analysis showed 27 bioactive compounds. The major compounds identified are 2,3-Dihydroxy-6-methyl-4-h-pyran-4-one, tetracosenone, nonadecane, tetracosane, 9-phytol and squalene. In a mass spectrum, each compound were identified based on their retention time and peak area. Pharmacological activities of these compounds indicated that the compounds present in the leaf of said plant can be used as a crude drug and can also be used to develop a novel drug.

INTRODUCTION: The pharmaceutical industries are using bioactive compounds as potent agents in treatment of many diseases. In the recent days bioactive compounds are gaining importance for their ability to cure many diseases. India has a rich heritage of medicinal plants. Plants have the capacity of synthesizing the organic compounds and are called as secondary metabolites, they have unique and complex structures. The secondary metabolites are used in the treating of chronic as well as infectious diseases. Waltheria indica L. is a highly valued medicinal plant used to treat the cancer, malaria and viral diseases. It also possesses the antimicrobial, antioxidant, anti-inflammatory and analgesic actions. Traditionally it is also used to treat wound healing, sleeping sickness and cough. Overall the plant is considered as febrifugal, purgative and emollient. In the last few years, the GC-MS has become the key tool for secondary metabolite profiling in plants. However, there are few reports are available on the pharmacological properties of the plant. Keeping this in view, the present study is undertaken to establish the GC-MS profile of ethanolic leaf extract of Waltheria indica Linn. and their pharmacological activities.

MATERIALS AND METHODS: Material: Waltheria indica Linn. belongs to the family Sterculiaceae. It is an under shrub growing in sub tropical and tropical regions including in India. It is commonly known as velvet leaf, marsh mallow and monkey bush. The Waltheria indica Linn. (Voucher specimen no: PB/MJ/01) leaves were collected from the plants grown in Karnataka University Campus, Dharwad, Karnataka. The leaves were washed with running tap water and later dried at room temperature.
The dried leaves were powdered using an electric blender. The powder obtained was sieved and stored in an air tight container at room temperature till further analysis\textsuperscript{13}.

**Leaf Extract:** About 10gms of dried and coarsely powdered leaves were subjected to extraction with ethanol (250 ml) using Soxhlet apparatus. The extract was subjected to GC-MS analysis\textsuperscript{13}.

**GC - MS Analysis:** The analysis was performed using a GC-MS (Shimadzu, Kyoto): QP2010S fitted with a 1.4 μm column Rxi-5silMS 30 meter length, 0.25 mm inner diameter and 0.25 μm film thickness. Carrier gas helium with a flow rate of 0.98 ml/min; column temperature 80 °C; initial temperature 70 °C, injector temperature 260 °C and detector temperature 300 °C, followed by a linear programmed temperature from 70 - 280 °C at a rate of 10 °C/min, operating in electron impact mode.

The samples were injected in splitless mode. Interface temperature was kept at 280 °C. Oven temperature programming was done from 80 °C - 260 °C at 10 °C /min. The pressure of the carrier gas was kept at 63.6 kPa. The constituents were identified based on interpretation on mass spectrum of the known components stored in the NIST 11 and WILEY 8 library\textsuperscript{15}. The name, molecular weight and molecular formula of the components of the test materials were tabulated.

**RESULTS:** The GC - MS chromatogram of the ethanolic leaf extract of *Waltheria indica* Linn. showed 27 peaks indicating the presence of twenty seven compounds (Fig. 1).

The active principles with their peak, retention time (RT), area (%), height (%), molecular formula and molecular weight are presented in the Table 1.

The tetradecane, hexadecane, squalene, 2,3-Dihydro-3,5-Dihydroxy-6-methyl-4H-pyran-4-one showed maximum per-centage. The 2-propenoic acid, 10-Henicosenoic (c.t), Nonadecane, 3-Eicosene, (E)-, 1,1 Bicyclo propyl-2-octanoic acid, megastigmatrinone-4 showed a moderate percentage. The minimum percentage compounds are 1-docosanol, 3',5'-Di-methoxyacetophenone, 2-Hexadecen-1-ol, 3,7,11, 15-Tetramethyl-\(\left[R-[R^*,R^*-(E)]\right]\), \(\left[Z,Z-8,10\right]\)-Hexa-decadien-1-ol, phytol, tetracosane, 2-bromodecane, 5,5, Diethyl hepta-decane.

The GC - MS identified compounds shows various pharmacological activities and are ascertained as various Table 2.

![FIG. 1: CHROMATOGRAM OF GC - MS ETHANOLIC LEAF EXTRACT](image)

### Table 1: GC-MS Analysis of Bioactive Compounds in Ethanol Leaf Extract of *Waltheria indica* Linn.

<table>
<thead>
<tr>
<th>Peak</th>
<th>R. Time</th>
<th>Area %</th>
<th>Height %</th>
<th>Molecular formula</th>
<th>Molecular weight (g/mol)</th>
<th>Name of the compound</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4.609</td>
<td>5.65</td>
<td>2.89</td>
<td>(\text{C}_4\text{H}_6\text{N}_6)</td>
<td>126.2</td>
<td>1,3,5-Triazine:2,4,6-Triamine</td>
</tr>
<tr>
<td>2</td>
<td>5.464</td>
<td>7.67</td>
<td>3.21</td>
<td>(\text{C}_6\text{H}_6\text{O}_4)</td>
<td>144.12</td>
<td>2,3-Dihydro-3,5-Dihydroxy-6-methyl-4H-pyran-4-one</td>
</tr>
</tbody>
</table>
TABLE 2: PHARMACOLOGICAL ACTIVITY OF PHYTOCOMPONENTS IN THE ETHANOLIC EXTRACT OF *WALTERIA INDICA* LINN. LEAF

<table>
<thead>
<tr>
<th>Name of the compound</th>
<th>Pharmacological activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,3,5-Triazine-2,4,6-Triamine</td>
<td>Antagonists&lt;sup&gt;16&lt;/sup&gt;, Cytotoxicity against leukemia and adenocarcinoma&lt;sup&gt;17&lt;/sup&gt; Inhibites glucocerebrosidase&lt;sup&gt;18&lt;/sup&gt;, Antileismenial activity&lt;sup&gt;19&lt;/sup&gt;, Anti-HIV&lt;sup&gt;20&lt;/sup&gt;, Antimalarial&lt;sup&gt;21&lt;/sup&gt; Antioxidant, Cancer preventive&lt;sup&gt;22&lt;/sup&gt; Antimicrobial&lt;sup&gt;22&lt;/sup&gt;</td>
</tr>
<tr>
<td>2,3-Dihydro-3,5-Dihydroxy-6-Methyl-4H-Pyran-4-One Tetradecane</td>
<td>Antioxidant&lt;sup&gt;1&lt;/sup&gt;, Antimicrobial&lt;sup&gt;1&lt;/sup&gt;, Automatic nerve activity, Anti-inflammatory&lt;sup&gt;23&lt;/sup&gt;, Antiproliferative&lt;sup&gt;24&lt;/sup&gt;, Anti-arthritis&lt;sup&gt;26&lt;/sup&gt;, Anti-cancer&lt;sup&gt;16&lt;/sup&gt;, Anti-diabetic&lt;sup&gt;1&lt;/sup&gt;, Pro-apoptic effects&lt;sup&gt;24&lt;/sup&gt; Antimicrobial&lt;sup&gt;27&lt;/sup&gt;, Cytotoxicity&lt;sup&gt;28&lt;/sup&gt;, Antipyretic, Anthelmentic, Tumour, Bronchitis, Asthma, Tuberculosis, Dyspepsia, Constipation, Anemia, Throat diseases, Elephantiasis, Antidiabetic, Anti-inflammatory, Antidiarrhoeal&lt;sup&gt;29&lt;/sup&gt;</td>
</tr>
<tr>
<td>Hexadecano, 2-Methyl-3',5'-Dimethoxyacetophenone</td>
<td>Antimicrobial&lt;sup&gt;30&lt;/sup&gt; No activity reported</td>
</tr>
<tr>
<td>Megastigmatrienone - 4</td>
<td>Cytotoxicity&lt;sup&gt;28&lt;/sup&gt; Antimicrobial, Antioxidant&lt;sup&gt;31&lt;/sup&gt;, Antipyretic, Anthelmentic, Tumour, Bronchitis, Asthma, Tuberculosis, Dyspepsia, Constipation, Anemia, Throat diseases, Elephantiasis, Antidiabetic, Anti-inflammatory, Antidiarrhoeal&lt;sup&gt;32&lt;/sup&gt; Aroma Nature&lt;sup&gt;33&lt;/sup&gt;, Cytotoxic Activity&lt;sup&gt;34&lt;/sup&gt; Antidiabetic, Anticancer&lt;sup&gt;35&lt;/sup&gt;, Haemolytic, Pesticide, Skin irritant, Hypcholesterolemic&lt;sup&gt;32&lt;/sup&gt;</td>
</tr>
<tr>
<td>2-Propenoic acid, tridecyl ester 3-Eicosane, (E)-</td>
<td>Antimicrobial, Antihyperglycemic&lt;sup&gt;36&lt;/sup&gt; Cytotoxic Activity&lt;sup&gt;36&lt;/sup&gt;,&lt;sup&gt;28&lt;/sup&gt; Antioxidant, Insecticidal activity&lt;sup&gt;37&lt;/sup&gt; No activity reported</td>
</tr>
<tr>
<td>Octadecane</td>
<td>Antioxidant, Anti-inflammatory&lt;sup&gt;38&lt;/sup&gt; Cough, lung diseases, Cold and fever detoxification&lt;sup&gt;39&lt;/sup&gt; Anti-corrosion agent&lt;sup&gt;36&lt;/sup&gt;, Antisepsis&lt;sup&gt;40&lt;/sup&gt;</td>
</tr>
<tr>
<td>2-Hexadecan-1-ol, 3,7,11,15-Tetramethyl-, [R-[R*,R*]-(E)]-1-Nonadecane</td>
<td>Anti-inflammatory, Antimicrobial, Cancer-preventive&lt;sup&gt;41&lt;/sup&gt; Antitumor, Analgesic, Antibacterial, Anti-inflammatory, Fungicide&lt;sup&gt;42&lt;/sup&gt; Antimicrobial, Antioxidant, Anticancer&lt;sup&gt;43&lt;/sup&gt;</td>
</tr>
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</table>
DISCUSSION: The GC-MS analysis revealed the 27 chemical compounds. The tetradecane (18.21%) is the highest chemical compound and 5,5-Diethylheptadecane (0.34%) as the lowest chemical compound. The compound 2,3-Dihydro-3,5-Dihydroxy-6-methyl-4H-pyron-4-one, tetradecane, Hexadecane, 2-Hexadecen-1-ol, 3,7,11,15-Tetramethyl-, [R-[R*,R*-(E)]]-, showed pharmacological activity as reported in the plants of Hibiscus tiliaceus Linn.\(^1\), Marsilea quadrifolia Linn.\(^2\) and Gymnema sylvestre (Retz) Schult.\(^3\). Similarly, phytol and squalene also showed the various biological activities as reported for Coldenia procumbens Linn.\(^4\). However, isolation and characterization of individual phytochemical constituents may proceed to discover the novel drugs and their pharmacological activities.

CONCLUSION: The ethanolic leaf extract of Waltheria indica Linn. has 27 different chemical compounds and they have different pharmacological activities. Each chemical compound can be extracted individually and can be used in clinical trials to check efficacy, and to develop a novel drug from a crude drug. The GC-MS analysis of leaf extract will also be a part of database of bioactive products of natural drugs.

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CONFLICT OF INTEREST: We have no conflict of interest.

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