GCMS analysis of a Traditional Medicinal Plant *Ficus bengalensis* L. – A fast analytical approach for identification of phytoconstituents

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**Abstract**
GCMS is normally used for direct analysis of component existing in traditional medicines and medicinal plants. A knowledge of the chemical constituents of plants is desirable not only for the discovery of therapeutic agents, but also because such information may be of great value in disclosing new sources of economic phytocompounds for the synthesis of complex chemical substances and for discovering the actual significance of folkloric remedies. Hence in the present study we selected a traditional medicinal plant *Ficus bengalensis*. Traditionally the bark is used in the treatment of diabetes, dysentery and diarrhea but scientific relevance behind this is still unknown. Thus further studies can be conducted to investigate the unexploited potential of *Ficus bengalensis*. Twelve compounds were identified and they were reported as dibutyl phthalate, Phthalic acid, 1,2-benzenedicarboxylic acid, Bis (2- methyl propyl) ester, butyl 2-pentyl ester, Diisoctyl phthalate, Squalene, Trans-Geranyleranion, α-Amyrin, α-Amyrin trimethylsilyl ether, Lup-20(29)-en-3-one and Lupeol. As *Ficus bengalensis* can grow and spread easily and because of its higher biomass availability, it can prove as an effective and cheaper drug for various human diseases.

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1. Introduction

Plants are a rich source of secondary metabolites with interesting biological activities. In general, these secondary metabolites are an important source with a variety of structural arrangement and properties. GCMS is normally used for direct analysis of component existing in traditional medicines and medicinal plants. A knowledge of the chemical constituents of plants is desirable not only for the discovery of therapeutic agents, but also because such information may be of great value in disclosing new sources of economic phyto compounds for the synthesis of complex chemical substances and for discovering the actual significance of folkloric remedies. Hence in the present study we selected a traditional medicinal plant from Atharvaveda - *Ficus bengalensis* L. belonging to family Moraceae. It is a large evergreen tree naturally occurring in tropical forests throughout the subcontinent. It is 23-24m tall with huge spreading limbs supported by aerial roots which later form accessory trunks extending to a large area. The therapeutic potential of *Ficus bengalensis* extracts in herbal medicine has been widely reported. It has been subjected to long term clinical trials in folk medicines. The milky juice is applied externally on pains and rheumatism. The root is used in gonorrhoea, dysentery and inflammation of the liver. The tips of the aerial roots are also used to relieve persisting vomiting and dysentery. The infusion of the bark is supposed to be a powerful tonic and is considered to have specific properties in the treatment of diabetes (Jayaweera, 1982; Subramanian and Misra, 1978). Plants have many phytochemicals with various...
bioactives. Studies have reported that extracts from natural products such as fruits, vegetables and medicinal herbs have positive effects against cancer, compared with chemotherapy or recent hormonal treatments. Therefore, many plants have been examined to identify new and effective antioxidant compounds as well as to elucidate the mechanism of action (Swamy and Tan, 2000). Hence the aim of this study is to determine the phytochemical constituents of *Ficus bengalensis* by GCMS method to ascertain the rationale for its use in traditional medicine.

2. Review of Literature

Plants produce a great array of secondary metabolites as a result of metabolic activities. These compounds either alone or in combination are being responsible for specific physiological changes or the therapeutic action in the human body when administered as a medicament or a health supplement. Hence, it will be practical to subject the individual herb for testing of these compounds. The more precise information in qualitative analysis can be obtained by gas-chromatography coupled with mass spectrometry (GC-MS). It has been seen that most of the work has been done on its antimicrobial activity, but very less information is available regarding the phytochemical analysis through GCMS method.

3. Materials and Methods

3.1 Collection of Plant Material

The leaves of *Ficus bengalensis* collected from different habitat of Pulgaon and Yavatmal.

3.2 Dry Powder Preparation

The plant leaf sample was dried at room temperature and ground into fine powder.

3.3 Sample Preparation for GCMS Analysis

About 15g of powdered material of plant was taken in a clean flat-bottomed glass container and soaked in 150ml of methanol. The container with its content was sealed and kept for 24 hours. The whole mixture then underwent a coarse filtration by a piece of clean, white cotton material. Then it was filtered through Whatman filter paper. The filtrate (methanolic extract) obtained for the plant was evaporated under ceiling fan.

GCMS Analysis: The GCMS analysis was conducted at the Central Instrumentation Laboratory, Punjab University at Chandigarh. 2µL aliquot was injected into a fisons GC8000 series coupled to a TSQ8000 MS (Triplequadrupole) mass analyzer. The chromatography was performed by using the DB5-MS column. Injection temperature was 230°C. Helium flow was 1 mL/min. After a 5 min solvent delay time at 70°C; the oven temperature was increased at 5°C/min to 310°C, 1min isocratic and cooled to 70°C, followed by the additional 5 min delay. The ion trace integration was done using the mass lab find target method for the characteristic fragment of assigned peaks.

3.4 Identification of Components

Interpretation of mass spectrum GCMS was conducted using data base of the Central Instrumentation Laboratory (CIL) spectra Libraries. Spectrum of the unknown component was compared with the spectrum of known components stored in the CIL. The molecular weight, molecular formula and the number of hits used to identify the name of the compound from CIL spectra Libraries were recorded.

4. Results and Discussion

GCMS is one of the best techniques to identify the constituents of volatile matter, long chain, branched chain hydrocarbons, alcohols, acids, esters etc. GCMS analyzed results which include the active principles with their molecular formula are presented in Table 1.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Name of Compound</th>
<th>Molecular Formula</th>
<th>Activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Dibutyl phthalate</td>
<td>C_{5}H_{12}O_{4}</td>
<td>Antimicrobial and Antiulcer</td>
</tr>
<tr>
<td>2</td>
<td>Phthalic acid</td>
<td>C_{4}H_{4}O_{4}</td>
<td>Antimicrobial</td>
</tr>
<tr>
<td>3</td>
<td>1,2-benzenedicarboxylic acid</td>
<td>C_{8}H_{4}O_{4}</td>
<td>Antimicrobial and Antiulcer</td>
</tr>
<tr>
<td>4</td>
<td>Bis(2-ethylhexyl) ester</td>
<td>C_{26}H_{52}O_{2}</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>Butyl 2-ethylhexyl</td>
<td>C_{4}H_{10}O_{2}</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td>Dibucryl phthalate</td>
<td>C_{26}H_{42}O_{2}</td>
<td>Antimicrobial and Antiulcer</td>
</tr>
<tr>
<td>7</td>
<td>Squalene</td>
<td>C_{30}H_{50}</td>
<td>Chemo preventive</td>
</tr>
<tr>
<td>8</td>
<td>Transeosamin</td>
<td>C_{22}H_{24}O_{2}</td>
<td>-</td>
</tr>
<tr>
<td>9</td>
<td>9-aminine</td>
<td>C_{2}H_{7}N</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>L-Homoserine</td>
<td>C_{9}H_{17}N_{2}O</td>
<td>-</td>
</tr>
<tr>
<td>11</td>
<td>L-Amino cyclodextrin</td>
<td>C_{20}H_{32}O_{12}</td>
<td>-</td>
</tr>
<tr>
<td>12</td>
<td>Lupin</td>
<td>C_{22}H_{30}O_{2}</td>
<td>Anti-inflammatory</td>
</tr>
</tbody>
</table>

**Figure 1:** Spectra of Gas Chromatography of leaves of *Ficus bengalensis* in methanol extract showing component peaks at different Retention time
Here twelve compounds were identified and they are reported as dibutyl phthalate, Phthalic acid, 1,2-benzenedicarboxylic acid, Bis (2- methyl propyl) ester, butyl 2-pentyl ester, Disiooctyl phthalate, Squalene, Trans-Geranylgeraniol, á-Amyrin, á-Amyrin trimethylsilyl ether, Lup-20(29)-en-3-one and Lupeol. On further study of each compound, it was found that they individually have its own biological importance.

The more precise information in qualitative analysis can be obtained by gas-chromatography coupled with mass spectrometry (GCMS). For quantitative determination, gas-chromatography with flame ionization detector (GC-FID) and GC-MS are preferred.

The GC-MS analysis of Ficus bengalensis leaves revealed the presence of twelve compounds. The identified compounds possess many biological properties. For instance, 1, 2- benzene dicarboxylic acid and dioctyl phthalate are known to possess antimicrobial and antifouling activity.

Dibutyl phthalate possess antimicrobial and antifouling properties. Ethyl iso-allocholate is reported to possess antimicrobial, diuretic, anti-inflammatory properties.

Lupeol is a pharmacologically active triterpenoid found in a variety of plants, including Mangifera indica and Acacia visco. It has several medicinal properties, one being anti-inflammatory. Study found lupeol to decrease paw swelling in rats by 39%, compared to 35% for the standardized control compound indomethacin (Geetha and Varalakshmi, 2001). Another report states that lupeol has a complex pharmacology in humans, displaying antiprotzoal, antimicrobial, anti-inflammatory, antitumor and chemo-preventive properties.

Squalene is a naturally occurring polyenyl compound primarily known for its key role as an intermediate in cholesterol synthesis. It receives its name because of its occurrence in shark liver oil (Squalus species) which contains large quantities and considered the richest source of squalene. Squalene is a natural antioxidant, a unique oxygen generator, power immune stimulator, antibiotic, anti-coagulant, anti-histamine and anti-allergic (Kelly, 1999). It has been proposed to be an important part of the Mediterranean diet as it may be a chemo preventative substance that protects people from cancer (Smith and Theresa, 2000 and Owen, 2004).

Thus each compound identified in leaf extract of Ficus bengalensis has its own biological importance and further study of this plant phytochemical can prove its medicinal importance in future and can be an effective and efficient drug source in cheaper rate as it has higher biomass availability.

**Conclusion**

The fundamental reason of quality control of herbal medicines is based on the concept of phytoequivalence of herbs, and then to use this conception to identify the real herbal medicine and the false one and further to do quality control. Therefore, GCMS method is a direct and fast analytical approach for identification of phytoconstituents. The importance of the study is due to the biological activity of some of these compounds. The present study, which reveals the presence of components in Ficus bengalensis suggest that the contribution of these compounds on the pharmacological activity can be further evaluated.

**Research Highlights**

GCMS analysis of leaves of Ficus bengalensis will be helpful in standardization of the plant.

Scientific relevance behind the medicinal properties of the plant will be known.

Biological activity of the evaluated compounds will be known.

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**References**


