ABSTRACT

Tamilnadia uliginosa (Retz.) Tirveng and Sastre belongs to the family Rubiaceae is a small tree distributed in the moist deciduous forest. In Kerala, the plant is widely seen in Muthanga Wild Life Santuary and Kuruva Island. T.uliginosa used as a drug in Ayurvedic, Siddha and Unani system of medicine. The raw fruits of T.uliginosa are eaten as vegetable and fruit extract is used against diarrhoea and dysentery. Fruit pulp is applied on boils. The fruits are rich in carbohydrate and possess piscical and insecticidal properties. Ripe fruit contains glycosides randioside-A, mollisidal triterpenoid glycosides and randianin, etc. It cures abscess, ulcers, inflammations, wounds, tumors, skin disease. In the present study, methanol extracts of T.uliginosa fruit have been analyzed by GC-MS. Ten compounds were identified in fruit extracts. Phytol and n- Hexadecanoic acid are the major phytocomponents. The identified compounds have a broad spectrum of many medicinal properties and anti-oxidant activities were identified. There is no previous report on GC-MS of this plant. The result confirmed the presence of bioactive compounds in T.uliginosa help to drug production and also these compounds are responsible for many pharmacological activities.

KEYWORDS: Tamilnadia uliginosa, Rubiaceae, anti-oxidant, bioactive components, pharmacology.

INTRODUCTION

Plants have been an important source of quality medicines for thousands of years.[1] The medicinal value of plants lies in certain chemical substances that produce a definite
physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds. Knowledge of the identity and relative amounts of the volatile substances released by plants is of great importance to several fields of basic and applied research in biology, chemistry and many other disciplines. For this, it is essential to overcome many analytical challenges posed by these complex mixtures, because they normally exhibit large variations in amounts, chemical structures and functionalities. Gas chromatography (GC) is recognized as the most suitable technique to find out how many components and in what proportion they are present in a complex mixture of volatile compounds. When GC is coupled to Mass Spectrometry (GC-MS), additional information arises about molecular mass, elemental composition (when high resolution mass spectrometry is used), functional groups and in certain cases, molecular geometry and spatial isomerism of each separated compound. GC-MS is normally used for direct analysis of components existing in traditional medicines and medicinal plants. In recent years GC-MS studies have been increasingly applied for the analysis of Chinese medicinal plants as this technique has proved to be a valuable method for the analysis of non-polar components and volatile essential oils, fatty acids and lipids. Gas chromatography - Mass spectrometry is the very compatible and the most commonly used technique for the identification and quantification purpose. The unknown organic compounds in a complex mixture can be determined by matching the spectrum of test sample with reference spectra.

Tamilnadia uliginosa (Retz.) Tiruveng and Sastre belongs to Rubiaceae is a deciduous, thorny, rigid shrub or tree. The fruits are edible and eaten as vegetable. It is an important medicinal plant and popularly known as emetic nut or divine jasmine. The plant is used in traditional practice in the treatment of liver, brain, skin, heart and other ailments. The fruit extract is used against diarrhoea and dysentery. Fruit pulp is applied on boils. The fruits are rich in carbohydrate and possess piscidal and insecticidal properties. Ripe fruit contains glycosides randioside-A, mollisidal triterpenoidglycosides, randianin etc. It cures abscess, ulcers, inflammations, wounds, tumous, skin disease. So the present study was conducted to determine the volatile compounds present in Tamilnadia uliginosa by GC-MS analysis.

MATERIALS AND METHODS

Collection of plant materials

The fruits of T.uliginosa used for the present study were collected from wayanad wild life sanctuary and identified by Dr. E.S. Santhoshkumar from Jawaharlal Nehru Tropical Botanic
Gardens and Research Institute, Thiruvananthapuram. A voucher specimen has been deposited in the Department of Botany, University of Kerala (RUBH 5810). The fruits were washed well using tap water followed by distilled water, cut into pieces and dried in shade for a period 20-25 days, at an ambient temperature of 25°C. The dried samples were ground initially in a mortar and pestle followed by a mixer grinder to obtain the fine powder.

Gas Chromatography-Mass spectrometric (GC-MS) Analysis

Extract preparation

GC-MS technique was used in this study to identify the volatile compounds present in the plant extract. Gas chromatography was carried out in GC Clarus 500 Perkin Elmer with Column Elite-5ms (5% Phenyl 95% Dimethyl polysiloxane). Mass detector used was Turbo mass-gold-Perkin Elmer and nitrogen was used as a carrier gas. Temperature of column was maintained at 220-280°C. The methanol extract of the fruit was injected into the Gas chromatography-Mass spectrometer. Identification of GC-MS mass spectrum was done using the database which is 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

The GC-MS profile showed the presence of ten compounds in methanol extract of T. uliginosa. The gas-chromatogram showed the relative concentrations of various compounds getting eluted as a function of retention time. The peak area indicated the relative concentrations of the compounds present in the plant. The retention time of the compounds eluted at different times was to identify the nature and structure of the compounds present in the sample. The structures of various compounds eluted are given in Table (1) and Figures 1 and 2.

Table1. Different Phytocomponents identified in the methanol extract of fruit of T. uliginosa

<table>
<thead>
<tr>
<th>Peak name</th>
<th>Molecular weight</th>
<th>Molecular formula</th>
<th>Retention time</th>
<th>% Peak Area</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-Hexanol, 2-ethyl-</td>
<td>130</td>
<td>C_{8}H_{18}O</td>
<td>7.31</td>
<td>1.0680</td>
</tr>
<tr>
<td>Nonanoic acid</td>
<td>158</td>
<td>C_{9}H_{18}O_{2}</td>
<td>12.20</td>
<td>0.3078</td>
</tr>
<tr>
<td>1-Undecanol</td>
<td>172</td>
<td>C_{11}H_{24}O</td>
<td>14.41</td>
<td>2.0199</td>
</tr>
<tr>
<td>Undecanoic acid</td>
<td>186</td>
<td>C_{11}H_{22}O_{2}</td>
<td>17.56</td>
<td>1.7187</td>
</tr>
</tbody>
</table>
Among the plant products, secondary metabolites are responsible for several biological activities in man and animals. Ten compounds identified by the GC-MS analysis of the extracts are medicinally valuable and possess various pharmaceutical applications. The compound 1-Hexanol, 2-ethyl with retention time 7.31 and peak area percentage 1.06 is known to possess antifungal and herbicidal properties. The compound Nonanoic acid, also called
pelargonic acid is a fatty acid, which has antioxidant and herbicidal activities. The compounds 1-undecanol (R.T-14.41) and undecanoic (RT-17.56) with peak area percentage 2.01 and 1.71 respectively are shown to have antioxidant and anti-inflammatory activities, flavour properties\cite{7} and also are used in perfumery. Another compound is 1-Hexadecene, with (RT-17.93) and peak area percentage is 3.54, has antimicrobial and antioxidant properties. The compounds tetradecanoic acid (RT-20.75) and 1-Nonadecene (RT-21.10) with peak area percentage 2.35, and 4.22 respectively possess antioxidant, antimicrobial and anticancer properties. The compound 3, 7, 11, 15-Tetramethyl-2-hexadecen-1-ol, a diterpene derivative also has antioxidant, anticancer and antimicrobial properties.

Another compound phytol (2.12% relative amount with 25.84 retention time) was also detected. Phytol is an acyclic diterpene alcohol that can be used as a precursor for the manufacture of synthetic forms of vitamin E\cite{8} and vitamin K. Phytol is a colourless compound and is a part of chlorophyll. Phytol possesses antimicrobial, anticancer, anti-inflammatory and diuretic properties.\cite{9,10} It also shows antidiabetic activity in type-II diabetic patients.\cite{11}

n-Hexadecanoic or palmitic acid is the most abundant saturated fatty acid in nature, and is the most common in animals, plants and microorganisms.\cite{12} It is the major component of the palm-oil and is found in meat, cheese, butter and dairy products. It possesses antioxidant, hypocholesterolemic, nematicidal, pesticidal, lubricant, haemolytic, cosmetic, antiandrogenic and flavoring properties.\cite{13,14} Reports have showed that secondary plant metabolites exert a wide range of biological activities on physiological systems. Review of literature reported the antimicrobial, anti-inflammatory, antioxidant, hypocholesterolemic, cancer preventive, hepato protective, anti-arthritis, anti-histaminic and anti-coronary activities of flavonoids, palmitic acid (Hexadecanoic acid, ethyl-ester and n-hexadecanoic acid), and an unsaturated fatty acid linolenic acid (docosatetradenoic acid, octadecanoic acid) It is therefore not unlikely that these phytochemicals found in T.uliginosa may play major roles in the reported biological activities and pharmacological properties of the plant. The study thus shows that T.uliginosa fruit is a rich reservoir of medicinally useful phytoconstituents which can be utilized beneficially by isolating these compounds using appropriate methods.

**CONCLUSION**

The present study is the first report on the GC-MS analysis in T.uliginosa fruit extract. Ten
chemical constituents were identified from the methanol extract of the plant. The GC-MS analysis detected the presence of fatty acids and plasticizer compounds. This study concluded that the methanol extract of T.uliginosa is a good source of natural antioxidants. It also possessed significant anticancer activity. Presence of medicinally useful compounds in the plant establishes its pharmaceutical importance. Further studies are however, needed to ascertain the pharmacological activity of the pure compounds.

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REFERENCES


