PHYTOCHEMICAL DETECTION OF WATER AND ETHANOL EXTRACT OF DOLICHANDRON FALCATA AND TINOSORA CORDIFOLIA LEAVES OF NANDURA AND MALKAPUR TAHSIL

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ABSTRACT
Dolichandron falcate (Medshing) and Tinosora Cordifolia (Guduchi) are the important medicinal plants in India. In the present study the water and ethanol extracts of leaves of the plants were prepared by simple reflux concentrated by distillation. This extracts were qualitatively analyzed for alkaloids, terpenoids, flavonoids, saponin, tannin, glycosides, phenols, anthroquinones, carbohydrates etc. Some of the above contents were also checked by TLC for R.F. values.

KEYWORDS: D.Falcata, T.Cordifolia, Phytochemical detection.

INTRODUCTION

Tinospora cordifolia generally known as Guduchi is a plant prescribed in Ayurveda. It belongs to the family Menisparmiaceae, an important drug used by Ayurveda Practitioners. T.Cordifolia is used in ayurvedic, “Rasayanas” to improve the immune system and the body resistance against infections. The root of this plant is known for its antistress, anti-leprotic and anti-malarial activities. The plant is also well known Indian bitter and prescribed for fevers, diabetes, dyspepsia, jaundice, urinary problems, skin diseases chronic diarrhea and dysentery. The plant is known as Amrita and the term is attributed to its ability to impart youthfulness, vitality and longevity to the consumer. The Dolichandrone falcata belongs to family Bignoniaceae is a traditional medicinal plant which is also termed as medshingi. D.Falcata is used by the tribal of Northeast Maharashra to cure stomach problems. It is a traditional medicinal plant of Ayurveda used for the purpose of abortion and fish poisoning. The whole plant is used in traditional medicine and the bark is mentioned to be the most powerful part. D. Falcata containing Chrysins (flavone) was identified and reported for different biological activities such as anti-oxidant, antiallergic, anti-inflamatory, anti-cancer, antiestrogenic and anxiolytic activities. The aim of the present investigation was to evaluate the various constituents from these plants of the selected area.

MATERIALS AND METHODS

Preparation of aqueous and ethanolic extract of D. Falcata and T. Cordifolia
The leaves of plant were collected from Nandura and Malkapur Tahsil area and washed with sterile distilled water. Then shade dried at room temperature for 3 days and grind into fine powder. 25 g of shade dried powdered plant leaf sample was sequentially refluxed in water and ethanol up to 2 h. The extracted samples were concentrated by distillation for distilled out the solvent. The dried extracts were weighed and preserved at 4 °C in refrigerator for further study.

Phytochemical detection of extracts
The aqueous and ethanolic extracts of plant leaves were subjected to preliminary phytochemical analysis. The presence of various groups of phytoconstituents like Alkaloids, Flavonoids, Terpenoids, Saponin, Tannin, Carbohydrate, Anthroquinone, Glycosides, steroids, Phenols, Gums, protein and Amino acids were analyzed by using the standard methods.

- Alkaloids
6 ml of extract was mixed with 6 ml of 1% HCl in steam bath, then it was filtered. 1 ml of Mayer’s reagent was added. Presence of turbidity shows presence of alkaloids. Further addition of a few drops of olive oil to form an emulsion confirmed the presence of alkaloids.
- **Flavonoids**
  5 ml of dilute ammonia solution was added to a portion of the aqueous filtrate of each plant extract followed by addition of concentrated H₂SO₄. A yellow coloration observed in each extract indicated the presence of flavonoids. The yellow colouration disappeared on standing after few minutes.

- **Terpenoids**
  0.5 gm extract was dissolved in 2 ml of chloroform then 3 ml concentrated sulfuric acid was added, a reddish brown color in interphase indicates the presence of terpenoids.

- **Saponins**
  0.5 g of the extract was dissolved in 5 ml distilled water. The mixture was shaken vigorously. Formation of stable persistent froth shows the presence of saponins. A further addition of 6 drops of olive oil while shaking forms an emulsion, confirming the presence of saponins.

- **Tannins**
  0.5 g of the extract was dissolved in 10 ml of distilled water, then a few drops of 1% ferric chloride solution was added to obtain a brownish green or blue black precipitate, which confirms the presence of tannin.

- **Carbohydrates**
  Few drops of Molisch’s reagent were added to 2ml portion of the various extracts. This was followed by addition of 2ml of conc. H₂SO₄ down the side of the test tube. The mixture was then allowed to stand for two-three minutes. Formation of a red or dull violet color at the interphase of the two layers was a positive test. Phenols 2 ml of extract was dissolved in 4 ml of distilled water and added few drops of 10% FeCl₃. Appearance of blue or green color indicates presence of phenols.

- **Anthraquinones**
  2.5 g extract was dissolved in 5 ml of conc. Sulfuric acid and filtered. The filtrate was dissolved in 2.5 ml of chloroform. Chloroform layer was pipette into a tube and 0.5 ml of 10% diluted ammonia was added. Formation of pink red or violet color shows the presence of anthraquinones.

- **Glycosides**
  2.5 g of extract was added to 2.5 ml distilled water. 1 ml glacial acetic acid containing a few drops of ferric chloride was added then 0.5 ml of concentrated sulfuric acid was added. Presence of brown ring at the interphase indicates the presence of deoxy sugar. A violet ring below the brown ring was observed, while a greenish ring also appears above the brown ring, confirming the presence of Cardiac Glycosides.

- **Steroids**
  To the test solution added 10ml of chloroform then filtered. To the 2 ml filtrate added 2 ml of acetic anhydride and conc.H₂SO₄. Blue green ring indicate the presence of steroids in the sample.

- **Amino acids and Proteins**
  2ml of filtrate was treated with 2-5 drops of ninhydrin solution placed in a boiling water bath for 1-2 minutes and observed for the formation of purple colour.

- **Phenols**
  A fraction of the extracts was treated with aqueous 5% ferric chloride and observed for formation of deep blue or black colour.

- **Gums**
  Test for gums were performed by hydrolyzing the 1 ml of extract using dil. HCl (3ml). Then Fehling’s solution was added drop by drop till the appearance of red.

**Thin Layer Chromatography**

TLC plates were prepared for different extracts using silica gel. The plates were placed in developing chamber containing mixture of solvents listed in Table no-3. The Rf Values of some selected constituents like alkaloids, flavonoids, terpenoids and saponin were determined for water and ethanolic extracts were reported in table no-2.

**RESULT AND DISCUSSION**

**Phytochemical Analysis**

The phytochemical analysis of the extracts revealed that D. falcata leaves were found to contain flavonoids, terpenoids,saponins, tannin,amino acids, proteins and glycosides, whereas Carbohydrates and gums were absent. Alkaloid were absent in water extract whereas Anthroquinones, Steroids and Phenols in ethanol extract of D.Falcata.

Alkaloids,flavonoids, Terpenoids,saponins, tannin,amino acids, proteins and glycosides were found to present in ethanol extract of T. Cordifolia leaves. Alkaloids,flavonoids, amino acids, proteins and phenols were found to present in water extract of T. Cordifolia leaves and reported in table no-1.

**Thin Layer Chromatography**

The Rf values of water and ethanol extracts of both plant leaves were found in the range of 0.46-0.86. The highest value (0.86) recorded for saponin in ethanol extract of D.Falcata plant leaves and lowest value (0.46) for terpenoids in ethanol extract of D. falcata leaves.
Table 1: Qualitative analysis of leaves of selected plants.

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>Constituents</th>
<th>Dolichandron Falcata</th>
<th>Tinospora Cordifolia</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Ethanol Extract</td>
<td>Water Extract</td>
</tr>
<tr>
<td>1</td>
<td>Alkaloids</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>Flavonoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>3</td>
<td>Terpenoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>Saponin</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>5</td>
<td>Tannin</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>6</td>
<td>Carbohydrates</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>Phenols</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>8</td>
<td>Amino acids and Proteins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>9</td>
<td>Anthroquinones</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>10</td>
<td>Glycosides</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>11</td>
<td>Steroids</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Gums</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 2: RF Values of some constituents from leaves of selected plants by TLC.

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>Constituents</th>
<th>RF Value Dolichandron Falcata</th>
<th>RF Value Tinospora Cordifolia</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Ethanol Extract</td>
<td>Water Extract</td>
</tr>
<tr>
<td>1</td>
<td>Alkaloids</td>
<td>0.68</td>
<td>0.61</td>
</tr>
<tr>
<td>2</td>
<td>Flavonoids</td>
<td>0.60</td>
<td>0.64</td>
</tr>
<tr>
<td>3</td>
<td>Terpenoids</td>
<td>0.46</td>
<td>0.58</td>
</tr>
<tr>
<td>4</td>
<td>Saponin</td>
<td>0.86</td>
<td>0.78</td>
</tr>
</tbody>
</table>

Table 3: Solvent system and Spraying agents with color developed.

<table>
<thead>
<tr>
<th>Constituent</th>
<th>Solvent system</th>
<th>Spraying Agent</th>
<th>Color developed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>NH₄OH:CH₃OH 3:17</td>
<td>Mayer’s reagent</td>
<td>Yellowish</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>CHCl₃:CH₃OH 18:2</td>
<td>Iodine vapors</td>
<td>Reddish</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>C₆H₆:CH₃COOC₂H₅ 1:1</td>
<td>10% HS₂O₄</td>
<td>Greenish</td>
</tr>
<tr>
<td>Saponin</td>
<td>CHCl₃:CH₃COOH: CH₂OH:H₂O 6:2:1:1</td>
<td>Iodine vapors</td>
<td>Brown</td>
</tr>
</tbody>
</table>

CONCLUSION

The selected plants were found to contain various bioactive constituents which are used for the treatment of various disease and disorders. Hence the further studies like characterization and elucidation of the structure of the bioactive compounds of these plants for industrial drug formulation are needed. Further it is needed to purify proteins from these plants which may act as a drug for the treatment of various diseases.

REFERENCES


