Phytochemical analysis of some medicinal plants of district Saharanpur

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Abstract

District Saharanpur is a rich source of medicinal plants because of its physical features, the north and the north-east of the district is surrounded by Shivalik hills and having rich biodiversity. The objective of present study was to investigate the presence of phytochemicals in five medicinal plants such as, Tinospora cordifolia (Thunb.)Miers, Bryophyllum pinnatum (Lam.) Oken, Terminalia belerica Roxb., Xanthium strumarium Linn. and Oldenlandia corymbosa Linn. For the study the solvents used were water, methanol, ethanol and acetone. Soxhlet apparatus was used for the organic solvent extraction. Carbohydrates, proteins, flavonoids, tannins, saponin, and phenols were detected in these selected medicinal plants. Our analysis proved that crude aqueous and organic solvent extracts of these plants having the bioactive compounds which are medicinally important for the use in the treatment of various diseases.

Keywords: Tinospora cordifolia (Thunb.) Miers, Bryophyllum pinnatum (Lam.) Oken, Terminalia belerica Roxb., Xanthium strumarium Linn. and Oldenlandia corymbosa Linn., soxhlet, organic solvents flavonoids.

1. Introduction

It is universal truth that medicinal plants are highly beneficial for human beings on this planet in the treatment of various diseases from ancient time. Drugs from the medicinal plants are easily available with ayurveda. Medicinal plants contain phytochemicals in the form of organic compounds. These phytochemicals are naturally synthesized in plants by primary and secondary metabolism.

Primary metabolism produces primary metabolites which are essential for growth and development. Whereas secondary metabolism produce secondary metabolites which are often colorful and flavored compounds.

1.2 Secondary Metabolites:

These are majority of three types as alkaloids, phenolics and terpenoids. These can be obtained by roots, tubers, stems, leaves, flowers, barks, fruits and seeds. Various secondary metabolites are used as medicines.

In present study, qualitative and quantitative phytochemical study was carried out in five medicinal plants of Saharanpur district as Tinospora cordifolia (Thunb.)Miers, Bryophyllum pinnatum (Lam.) Oken, Terminalia belerica Roxb., Xanthium strumarium Linn. and Oldenlandia corymbosa Linn.
plant materials were taxonomically identified and authenticated by Dr. S.K. Upadhyaya (Well known Botanist of India). The plant materials were shade dried and after drying, we use mechanical blender to ground the plant materials. Fine powder of the leaves with proper labeling was used for analysis.

2.2 Preparation of plant extracts:

2.2.1 Water extraction:
Finely 5gm of dried powdered plant material was taken in a beaker along with 100ml of distilled water and heated with continuous stirring at 30º-40ºC for 20 minutes. Then the water extract was filtered through filter paper and the filtrate was used for the phytochemical analysis.

2.3 Qualitative phytochemical analysis:
The extract was tested for the presence of bioactive compounds by using following standard methods [1-3].

2.3.1 Test for proteins:
Ninhydrin test:
When crude extract and 2ml of 0.2% solution of Ninhydrin boiled then appearance of violet colour show the presence of amino acids and proteins.

Millon’s test:
When crude extract and 2ml of Millon’s reagent mixed then white precipitate appeared which turned red upon gentle heating, which confirmed the presence of protein.

2.3.2 Test for carbohydrates:
Benedict’s test:
When crude extract and 2ml of Benedict’s reagent mixed and boiled, then a reddish brown precipitate indicated the presence of carbohydrates.

Molisch’s test:
Crude extract was mixed with 2ml of Molisch’s reagent and the mixture was shaken vigorously. The development of a violet ring at the interphase indicated the presence of carbohydrate.

Fehling’s test:
Equal volume of Fehling A and Fehling B reagents were mixed together and 2ml of it was added to crude extract and boiled. Presence of brick red precipitate at the bottom of the test tube indicated the presence of reducing sugars.

Iodine Test:
Crude extract was mixed with 2ml of iodine solution. A dark blue or purple color indicated the presence of carbohydrate.

2.3.3 Test for phenols and tannins:
When 3ml of 2% solution of FeCl3 and crude extract was mixed then presence of blue-green or black color indicated the presence of phenols and tannins.

2.3.4 Test for flavonoids:
Shinoda test:
Crude extract was mixed with few fragments of magnesium ribbon and concentrated HCl was added drop wise. Pink scarlet colour appeared after few minutes which indicated the presence of flavonoids.

Alkaline Reagent Test:
Crude extract was mixed with 2ml of 2% solution of NaOH. An intense yellow colour was formed which turned colourless on addition of few drops of diluted acid which indicated the presence of flavonoids.

2.3.5 Test for saponins:
Crude extract was mixed with 5ml of distilled water in a test tube and it was shaken vigorously. The formation of stable foam was taken as an indication for the presence of saponins.

2.3.6 Test for glycosides:
Keller-kilani test:
Crude extract was mixed with 2ml of glacial acetic acid containing 1-2 drops of 2% solution of FeCl3. The mixture was then poured into another test tube containing 2ml of concentrated H2SO4 A brown ring at the interphase indicated the presence of cardiac glycosides.

Liebmann’s test:
Crude extract was mixed with each of 2ml of chloroform and 2ml of acetic acid. A red colour change from violet to blue to green indicated the presence of steroidal nucleus, i.e., glycone portion of glycoside.

2.3.7 Test for steroid:
Crude extract was mixed with 2ml of chloroform and concentrated H2SO4 was added sidewise. A red colour produced in the lower chloroform layer indicated the presence of steroids. Another test was performed by mixing crude extract with 2ml of chloroform. Then 2ml of each of concentrated H2SO4 and acetic acid were poured into the mixture. The development of a greenish coloration indicated the presence of steroids.

2.3.8 Test for terpenoids:
Crude extract was dissolved in 2ml of chloroform and evaporated to dryness. To this, 2ml of concentrated H2SO4 was added and heated for about 2 minutes. A grayish colour indicated the presence of terpenoids.

2.3.9 Test for alkaloids:
Crude extract was mixed with 2ml of 1% HCl and heated gently. Mayer’s and Wagner’s reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids.

2.4 Quantitative phytochemical analysis:
2.4.1 Estimation of total phenolic content:
The total phenolic content was determined by the spectrophotometric method [4]. In brief, a 1 ml of aqueous
extract (1 mg/ml) was mixed with 1 ml of Folin–Ciocalteu’s phenol reagent. After 5 min, 10 ml of a 7% Na₂CO₃ solution was added to the mixture followed by the addition of 13 ml of deionized distilled water and mixed thoroughly. The mixture was kept in the dark for 90 min at 23°C, after which the absorbance was read at 750 nm.

2.4.2 Estimation of Total flavonoid content:

Aluminium chloride colorimetric method was used with some modifications to determine flavonoid content.

1ml of sample plant extract was mixed with 3ml of methanol, 0.2ml of 10% aluminium chloride, 0.2ml of 1M potassium acetate and 5.6ml of distilled water and remains at room temperature for 30 minutes. The absorbance was measured at 420nm. Quercetin was used as standard (1mg/ml). All the tests were performed in triplicates. Flavonoid contents were determined from the standard curve and were expressed as quercetin equivalent (mg/g of extracted compound) [5].

### Table 1: Qualitative Analysis

<table>
<thead>
<tr>
<th>Name of Plants</th>
<th>Local Name</th>
<th>Family</th>
<th>Carbohydrates</th>
<th>Phenols/ Tannins</th>
<th>Flavonoids</th>
<th>Saponins</th>
<th>Glycosides</th>
<th>Steroles</th>
<th>Terpenoids</th>
<th>Alkaloids</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tinospora cordifolia</td>
<td>Giloy</td>
<td>Minispermaceae</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Bryophillum pinnatum</td>
<td>Pashan bheda</td>
<td>Crassulaceae</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Terminalia bellerica</td>
<td>Baheda</td>
<td>Combertaceae</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Xanthium strumarium</td>
<td>Chota Gokhru</td>
<td>Compositeae</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Oldeniandia corymbosa</td>
<td>Pitt Papda</td>
<td>Rubiaceae</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
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</table>

### Table 2: Quantitative Analysis (Total Flavonoids)

<table>
<thead>
<tr>
<th>Name of Plants</th>
<th>% (mg/g) Total Flavonoids</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tinospora cordifolia</td>
<td>16.5</td>
</tr>
<tr>
<td>Bryophillum pinnatum</td>
<td>8.0</td>
</tr>
<tr>
<td>Terminalia bellerica</td>
<td>38.2</td>
</tr>
<tr>
<td>Xanthium strumarium</td>
<td>25.9</td>
</tr>
<tr>
<td>Oldeniandia corymbosa</td>
<td>5.3</td>
</tr>
</tbody>
</table>

3. Results

The phytochemical characteristics of five medicinal plants tested were summarized in the table-1. The results revealed the presence of medically active compounds in five plants studied. From the table, it could be seen that, proteins, carbohydrates, phenols and tannins, flavonoids and saponins were present in all the plants. Glycosides were absent only from the leaves of Tinospora cordifolia. Steroids were absent only in the leaves of Xanthium strumarium. Terpenoids were absent in the leaves of Xanthium strumarium. Alkaloids were absent in the leaves of Terminalia bellerica and also in the leaves of Tinospora cordifolia.

Total flavonoid contents obtained were 16.5 mg/g, 8.0 mg/g, 38.2 mg/g, 25.9 mg/g, 5.3 mg/g, of the extract for the plants Tinospora cordifolia(Leaves), Bryophillum pinnatum(Leaves), Terminalia bellerica(Leaves), Xanthium strumarium (Leaves) and Oldeniandia corymbosa (Leaves) respectively. (Table-2).

4. Discussion

Phytochemical analysis conducted on the plant extracts revealed the presence of constituents which are known to exhibit medicinal as well as physiological activities [6]. Analysis of the plant extracts revealed the presence of phytochemicals such as phenols, tannins, flavonoids, saponins, glycosides, steroids, terpenoids, and alkaloids.

The phenolic compounds are the groups of plant metabolites [7]. They possess biological properties such as, antiaging, antiinflammation, and cardiovascular protection [8]. Flavonoids are wide range of phytochemical with various pharmacological effects including antioxidant, anti-inflammation, anti-platelet, anti-allergic, cytotoxicity, reduce risk for heart disease or cancer etc. [9]. The plant extracts were also revealed to contain saponins which are known to produce inhibitory effect on inflammation [10]. Saponins have the property of precipitating and coagulating red blood cells. Some of the characteristics of saponins include formation of foams in aqueous solutions, hemolytic activity, cholesterol binding properties and bitterness [11]. Steroids have been reported to have antibacterial properties [12]. Alkaloids have the analgesic [13], antispasmodic and antibacterial [14,15] properties. Glycosides are known to lower the blood pressure according to many reports [16]. The results obtained in this study thus suggest the identified...
Phytochemical compounds may be the bioactive constituents and these plants are proving to be an increasingly valuable reservoir of bioactive compounds of substantial medicinal merit.

5. Conclusion

The medicinal plants can be seen as a potential of drugs in this area. The medicinally important phytochemical constituents in the plants studied, are bioactive in nature. These bioactive compounds contribute medicinally in the treatment of different disease. The present study is suggested that further work should be carried out to isolate, purify, and characterize the active constituents of these plants.

References