Phytochemical screening to validate the ethnobotanical importance of root tubers of *Dioscorea* species of Meghalaya, North East India

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Qualitative phytochemical analysis was done in root tubers of six species of *Dioscorea* found in Meghalaya. The test confirms the presence of various phytochemicals like flavonoids, saponins, steroids, cardiac glycosides and terpenoids in two aqueous extracts of methanol and ethyl acetate. The results suggest that the methanolic extract shows the presence of maximum phytochemical compounds than ethyl acetate extract during screening. Cholesterol and alkaloid was not detected in present investigation.

**Keyword:** *Dioscorea*, Ethnomedicine, Phytochemical, Solvent extraction, Secondary metabolites.

1. Introduction

Traditional knowledge of medicine has long been used since ages for curing various human ailments. About 60-80% of world populations still rely on plant based medicines [25]. Though the traditional Indian system of medicine has a long history of use, yet they lack adequate scientific documentation, particularly in light of modern scientific knowledge [28]. The medicinal value of plant lies in the bioactive phytochemical constituents of the plant and which shows various physiological effects on human body. So through phytochemical screening one could detect the various important compounds which could be used as the base of modern drugs for curing various diseases. Keeping this in view, the plant *Dioscorea* commonly known as yam has been taken for phytochemical screening. Yam is the leading form of staple food for millions of people in the tropic and subtropical countries. Many yams are of economic importance as tuberous food crops. This tuber contains the plant food reserves, mainly starch, and it is often incorporated in the human diet. The tuber not only stores food but also many of the plants secondary metabolites, which are commonly referred to as antinutritional factors. The study investigates on the qualitative phytochemical screening of few species of *Dioscorea* prepared in two different extracts i.e. methanol and ethyl acetate.

1.1 Food and Economic aspect

The Tubers of several species of Yams (*Dioscorea* spp.) are edible and are counted just after potato in its food value. In fact, species like *D. alata*, *D. pentaphylla* and *D. bulbifera* are the most worldwide cultivated true yams for their tubers which are of rich source of starch that form an important dietary supplement. Apart from starch the root tubers of *Dioscorea* also contain protein, fats, fibers and among minerals nutrients Potassium, Sodium, Phosphorus, Calcium, Magnesium, Copper, Iron, Manganese, Zinc and Sulphur containing amino acids [4]. “Diosgenin” is a pharmacologically active component of *Dioscorea* obtained from root and rhizomes which is one of the most costly and important
steroidal drug used worldwide [27]. Dietary PEs (plant estrogens) of Dioscorea can provide wide range of health benefits including protection against development of some cancers, osteoporosis, cardiovascular disease, and nephritis, and asthma, diabetes, used in Preparation of contraceptives and in the treatment of various genetic disorders. Edible species along with other ethnomedicinal value of Dioscorea found in Meghalaya are presented in Table 1.

<table>
<thead>
<tr>
<th>Table 1: Edible and medicinal uses of six Dioscorea species</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Species</strong></td>
</tr>
<tr>
<td>Dioscorea alata Linn.</td>
</tr>
<tr>
<td>Dioscorea bulbifera Linn.</td>
</tr>
<tr>
<td>Dioscorea pentaphylla Linn.</td>
</tr>
<tr>
<td>Dioscorea pubera Bl</td>
</tr>
<tr>
<td>Dioscorea oppositifolia L.</td>
</tr>
<tr>
<td>Dioscorea glabra Roxb</td>
</tr>
</tbody>
</table>

2. Materials and Methods
2.1 Collection of Plant Material
Six species of Dioscorea tubers (D. pentaphylla, D.alata, D.oppositifolia, D. bulbifera, D. glabra and D. pubera) were collected (fig.1) from the East Khasi Hill District of Meghalaya.

2.2 Sample preparation
The tubers were washed and air dried. After drying, the samples chopped into smaller pieces ground into powder and stored in airtight bottles before analysis.

2.3 Preparation of plant extract
10 gm of air dried powder were taken in 100 ml of methanol and ethyl acetate. Plugged with cotton wool and then kept on a rotary shaker at 199-220 rpm for 24 hours. The supernatant were collected and the solvent were evaporated to the final volume one-fourth of the original volume and stored at 4 °C in air tight containers [21].

2.4 Preliminary Phytochemical Screening
The condensed extracts were used for preliminary screening of phytochemicals such as cholesterol, alkaloid, flavanoids, saponin, cardiac glycosides and terpenoids.

2.5 Screening Procedure
2.5.1 Test for flavanoids
Add a few drops of concentrated HCL and Mg turning to 1 ml of ethanol extract. Appearance of pink or magenta-red colour indicates the presence of flavanoids [17].

2.5.2 Test for cholesterol
To 2 ml of the extract 2 ml of the chloroform was added in a dry test tube. Then 10 drop of acetic anhydride and 2 to 3 drops of con. H₂SO₄ was added. A red colour changed to blue green colour [4].

2.5.3 Test for Alkaloids
To the extract added 1% HCl and 6 drops of
Mayer’s reagent and Dragendorff reagent. Any organic precipitate indicated the presence of alkaloids in the sample [13].

2.5.4 Test for terpenoids
5ml of each extract was added to 2ml of chloroform and 3ml of con.H2SO4 to form a monolayer of reddish brown coloration of the interface was showed to form positive result for the terpenoids [1].

2.5.5 Test for cardiac glycoside
5ml of each extract was treated with 2ml of glacial acetic acid containing one drop of ferric chloride solution. This was underplayed with 1ml of con.H2SO4. A brown ring of the interface indicated a deoxysugar characteristic of cardenolides. A violet ring might appear below the brown ring whereas acid layer, a greenish ring might form just gradually throughout thin Layer [1].

2.5.6 Test for steroids
2 ml of acetic anhydride was added to 0.5 g of ethanolic extract of each sample with 2ml of H2SO4. The colour change from violet to blue or green indicated the presence of steroids [8].

2.5.7 Test for Saponins
The extract with 20 ml of distilled water was agitated in a graduated cylinder for 15 minutes. The formation of 1cm layer of foam indicated the presence of saponins [13].

3. Results
The result of the preliminary phytochemical screening from root tubers in table 2 shows the presences of different phytochemicals prepared in two different solvent extracts. Cholesterol and alkaloid were not detected during the present investigation. Quantitative assessments of the different phytochemicals detected during investigation was graded as –ve for 0, +ve for 1, ++ve for 2 and +++ve for 3. The present study reveals the presence of phytochemicals like flavanoids, saponins, cardiac glycosides and terpenoids in methanol extracts. High amount of flavanoid and Terpenoids was also found in species of D.bulbifera as shown in fig 5. D.oppositifolia shows high amount of terpenoids in methanolic extracts. In ethyl acetate extract, terpenoids was high in D.bulbifera as shown in fig. 6.
Fig 1: showing different root tubers of Dioscorea species
Fig 2: Test for flavonoid in Methanol Extract

Fig 3: Test for flavonoid in ethyl acetate extract

Fig 4: Test for saponins in methanol extract
### Table 2: Phytochemical screening of six species of *Dioscorea*

<table>
<thead>
<tr>
<th>Name of the compound</th>
<th>D. pentaphylla</th>
<th>D. alata</th>
<th>D. bulbifera</th>
<th>D. oppositifolia</th>
<th>D. pubera</th>
<th>D. glabra</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cholesterol</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Alkaloid</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Flavanoid</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Terpenoid</td>
<td>+</td>
<td>+</td>
<td>+++</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Cardiac glycoside</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Steroid</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td>Saponin</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

**Ethyl acetate extract**

<table>
<thead>
<tr>
<th>Name of the compound</th>
<th>D. pentaphylla</th>
<th>D. alata</th>
<th>D. bulbifera</th>
<th>D. oppositifolia</th>
<th>D. pubera</th>
<th>D. glabra</th>
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<tbody>
<tr>
<td>Cholesterol</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td>Alkaloid</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Flavanoid</td>
<td>-</td>
<td>-</td>
<td>+++</td>
<td>-</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td>Terpenoid</td>
<td>++</td>
<td>++</td>
<td>+++</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Cardiac glycoside</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Steroid</td>
<td>-</td>
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<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Saponin</td>
<td>+</td>
<td>-</td>
<td>+</td>
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**Methanol Extract**

<table>
<thead>
<tr>
<th>Name of the compound</th>
<th>D. pentaphylla</th>
<th>D. alata</th>
<th>D. bulbifera</th>
<th>D. oppositifolia</th>
<th>D. pubera</th>
<th>D. glabra</th>
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</thead>
<tbody>
<tr>
<td>Cholesterol</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Alkaloid</td>
<td>-</td>
<td>-</td>
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<td>-</td>
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<td>-</td>
</tr>
<tr>
<td>Flavanoid</td>
<td>-</td>
<td>-</td>
<td>+++</td>
<td>-</td>
<td>+</td>
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</tr>
<tr>
<td>Terpenoid</td>
<td>++</td>
<td>++</td>
<td>+++</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Cardiac glycoside</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Steroid</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Saponin</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

Key: - absence, + presence, ++ fairly good amount, +++ good amount

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**Fig 5:** methanol extract of the compounds.

**Fig 6:** Ethyl extract of the compounds.
4. Discussion
The Phytochemical screening and qualitative estimation of root tubers of six species of Dioscorea shows the presence of flavanoids, terpenoids, saponin, steroid and cardiac glycosides. Yams have been well respected by the herbalist community for generations due to their potency in enhancing fertility in males due to the presence of steroidal drug i.e. diosgenin which have been isolated from yam tubers. Diosgenin is used as precursors for the synthesis of hormones and corticosteroids which improve fertility in males [7, 19]. Saponins natural tendency to ward off microbes makes them good candidates for treating fungal and yeast infections. These compounds served as natural antibiotics, which help the body to fight infections and microbial invasion [30]. The biological functions of flavanoids apart from its antioxidant properties include protection against allergies, inflammation, free radicals, platelet aggregation, microbes, ulcers, hepatoxins, viruses and tumors [2]. Cardiac glycosides content was found in methanol extract. Cardiac glycosides have been used for over two centuries as stimulant in case of cardiac failure [31, 18]. Further terpenes or terpenoids are active against bacteria [3, 11, 26]. The presence of terpenoids shows that it could be effective against any bacterial infections. This perhaps could probably support and justify the information about the usage of phytochemicals which were isolated from the solvent extract in the present study.

5. Conclusion
The presence of various phytochemicals such as flavonoids, terpenoids, saponin, steroid and cardiac glycosides in the different species of Dioscorea confirms that this genus is a potent source for modern drugs. The present study not only paves way for preliminary contribution to the medico-botany investigation but also shows a way for pharmacological research in future for the discovery of new sources of drugs from these phytochemicals.

6. Acknowledgement
The authors are thankful to head of Botany Department, NEHU for providing necessary facilities and Director of Botanical Survey of India, Eastern circle, Shillong for allowing library and herbarium consultation. The authors are also thankful to University Grants Commission (UGC) New Delhi for financial support through University with Potentials for Excellence (UPE) Scheme.

7. References
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