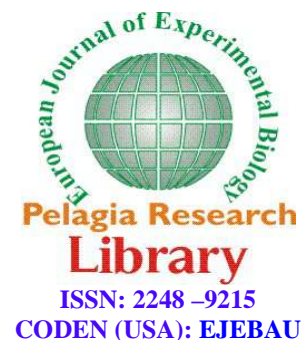




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Preliminary phytochemical screening of *Glochidion ellipticum*

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ABSTRACT

The phytochemical analysis of the plant is very important commercially and has great interest in pharmaceutical companies for the production of the new drugs for curing of various diseases. It is expected that the important phytochemical properties recognized by our said study will be very useful in the curing of various diseases of this region. We have focused on plant in western ghat region of Maharashtra and selected *Glochidion ellipticum* genes from Euphorbiaceae family. The main objective of the research work was to carryout micro macroscopical characterization and preliminary phytochemical test for the said genus.

Key words: *Glochidion ellipticum*, Euphorbiaceae, macro and microscopic characterization

INTRODUCTION

Preliminary screening of phytochemicals is a valuable step, in the detection of the bioactive principles present in medicinal plants and subsequently may lead to drug discovery and development. Medicinal plants have bioactive compounds which are used for curing of various human diseases and also play an important role in healing. Phytochemicals have two categories i.e., primary and secondary constituents. Primary constituents have chlorophyll, proteins sugar and amino acids. Secondary constituents contain terpenoids and alkaloids. Medicinal plants have antifungal, antibacterial and anti-inflammation activities¹.

Herbal medicines have recently attracted much attention as alternative medicines useful for treating or preventing life style related disorders and relatively very little knowledge is available about their mode of action. There has been a growing interest in the analysis of plant products which has stimulated intense research on their potential health benefits. Plants are the essential and integral part in Complementary and Alternative medicine and due to this they develop the ability for the formation of secondary metabolites. Plants are the best source of active secondary metabolites which are beneficial to mankind in treating many diseases. Genus *Glochidion* have been used for a varied of biological activities in traditional medicine and also have been using by many ethnic groups. It is a vast genus in which many plants explored chemically, but most of the species in this genus were not standardized pharmacognostically.²⁻⁵

The main objective of the research work was to check the presence of the phytochemical constituents in all the medicinal plant. The results of the phytochemical analysis of these medicinal plants showed that the terpenoids,

phlobatannins, reducing sugar, flavonoids and alkaloids were found to be present in afore mentioned medicinal plant. Macroscopic and microscopic characterization of *Glochidion ellipticum*.

MATERIALS AND METHODS

Plant materials

The plant was collected from the forest regions of Koyna dam of karad dist, satara. It was authenticated by Dr. Sanjay S. Sathe, Asso. Professor, Dept. of Botany, PDVP, Mahavidyalaya Tasgaon, Dist- Sangli. A herbarium was prepared and deposited in the Dept. of Pharmacognosy for further reference. The plant was identified as *Gochidion ellipticum*. (Euphorbiaceae) and was certified under Voucher No: RCP-SNG/ ph'cog/ 2009-10/003.

Chemicals

All the chemicals and reagent used were of laboratory grade and were procured from manufactures of Research lab fine chemicals, Mumbai., Loba Chemie, Mumbai, Sigma-Aldrich, Mumbai., Hi Media Lab Mumbai, Finar reagents, Ahmadabad, Merck, Mumbai, Genuine Chem., Mumbai, Labin, Mumbai, Moly Chem, Mumbai)

Extraction methods

Preparation of various extract of medicinal plants

- Aqueous extraction

Aqueous extracts *Gochidione lipticum* were carried out by cold maceration. In this process, solid ingredients were subjected to cold maceration with chloroform: Water I.P (2:98) (Indian Pharmacopoeia (I.P.); 1996). Powder was placed in 2 liters round bottom flask for about 7 days at room temperature in a warm place. The flask was securely plugged with absorbent cotton and was shaken periodically with frequent agitation until soluble matter is dissolved. The mixture was filtered and after most of the liquid has drained, the filtrate was concentrated to residue at constant temperature bath at temperature 500C. Note: Chloroform water I.P. 2.5 ml of chloroform was shaken with 900 ml of water until dissolved and diluted to 1000 ml with water.

- Successive solvent extraction

The dried leaves of the plant of *Gochidion ellipticum* were reduced to coarse powder (40 size mesh) and around 200 gm of powder was subjected to successive hot continuous extraction (soxhlet apparatus) with petroleum ether (60-800C), chloroform, ethyl acetate and ethanol to about 10 cycles per batch for 1 batches. The extraction was continued until the solvent in the thimble became clear. Each time before extracting with next solvent the powdered material was dried at room temperature. After the effective extraction, solvent was distilled off using rotary vacuum evaporator and the extracts were concentrated at low temperatures. The dried concentrated extracts were used for phytochemical investigation, isolation, pharmacological activity.^{6,7}

EXPERIMENTAL WORK

- Phytochemical screening⁸

Test for phlobatannins

Plant powder sample was mixed with distill water in a test tube, then shaken it well, and filtered to take plant extract. Then to each plant extract, 1% aqueous hydrochloric acid was added and each plant sample was then boiled with the help of Hot plate stirrer. Formation of red colored precipitate confirmed a positive result

Test for reducing Sugar

An amount of 0.50 g of selected plant sample was added in 5 ml of distilled water. Then 1 ml of ethanol mixed in plant extract. After that we took 1 ml of Fehling solution A and 1 ml of Fehling solution B in a test tube, heated it to boiling and then poured it in the aqueous ethanol extract. When color reaction was observed, it shows a positive result.

Test for terpenoids

An amount of 0.8 g of selected plant sample was taken in a test tube, then poured 10 ml of ethanol in it, shaken well and filtered to take 5 ml extract of plant sample. Then 2 ml of chloroform were mixed in extract of selected plant sample and 3 ml of sulphuric acid were added in selected sample extract. Formation of reddish brown color indicates the presence of terpenoids in the selected plants.

Test for flavonoids

For the confirmation of flavonoid in the selected plants, 0.5 g of each selected plant extract were added in a test tube and 10 ml of distill water, 5 ml of dilute ammonia solution were added to a portion of the aqueous filtrate of each plant extract followed by addition of 1 ml concentrated H₂SO₄. Indication of yellow color shows the presence of flavonoid in each extract.

Test for alkaloids

For the purpose of phytochemical analysis of the selected plants, 0.2 g of the selected plant samples were added in each test tube and 3 ml of hexane were mixed in it, shaken well and filtered. Then took 5 ml of 2% HCl and poured in a test tube having the mixture of plant extract and hexane. Heated the test tube having the mixture, filtered it and poured few drops of picric acid in a mixture. Formation of yellow color precipitate indicates the presence of alkaloids.

Microscopic Characterization

- T. S. of leaves of *Glochidion ellipticum*
Powder characterization



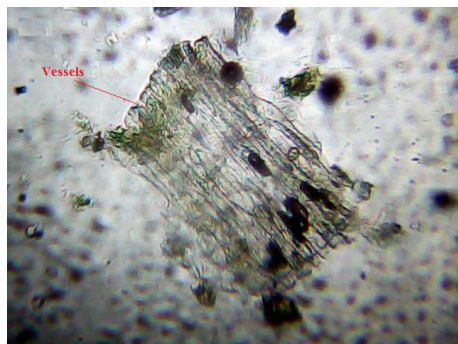
- Ca oxalate crystals and starch grain



- Stomata



- Parenchyma sheet



- Vessels

- Determination of acid value of bark & Total ash value of leaf of *Glochidion ellipticum*

Procedure:-

Proceed as per the steps mentioned in the procedure for determination of total ash value of a crude drug.

1. Using 25 ml of dilute hydrochloric acid, wash the ash from the dish used for total ash into a 100 ml beaker.
2. Place a mere gauze over a Bunsen burner & boil for five minutes.
3. Filter through an 'ashless' filter paper, wash the residue twice with hot water.
4. Ignite a crucible in the flame, cool & weigh.
5. Put the filter paper & residue together into the crucible; heat gently until vapours cease to be evolved & then more strongly until all carbon has been removed.
6. Cool in a desiccator.
7. Weigh the residue & calculate acid – insoluble ash of the crude drug with reference to the air-dried sample of the crude drug.

Calculation:-

- Acid-insoluble ash value of bark of *Glochidion ellipticum*

1. Weight of the empty dish=68.02gm
2. Weight of the drug taken=2 gm
3. Weight of the dish + Ash= 68.42
4. Weight of the residue='a' gm= 68.42 – 68.02=0.4 gm
5. 'y' gm of air dried drug gives 'a' gm of acid-insoluble ash.

Therefore 100 gm of the air-dried drug gives $100 \times \frac{a}{y}$ gm of acid-insoluble ash.

Acid-insoluble ash value of the sample = $100 \times \frac{a}{y} \%$

= $100 \times \frac{0.4}{2} = 20\%$

Acid-insoluble ash value of bark of *Glochidion ellipticum* =20%

Calculation:-

Total ash value of leaf of *Glochidion ellipticum*

1. Weight of the empty dish =x=67.1 gm
2. Weight of the drug taken=y=2gm
3. Weight of the dish + Ash (after complete incineration)=z=68.3 gm
4. Weight of the ash=(z - x)=(68.3 -67.1)=1.2 gm
5. 'y' gm gm of crude drug gives (z - x) gm of the ash.

Therefore 100 gm of crude drug gives $100/y \times (z-x)$ gm of the ash

Total ash value of the sample= $100 (z-x)/y\%$

= $100 (1.2)/2 =60\%$

Total ash value of *Glochidion* leaf is not more than 60%.

RESULTS

This study has revealed the presence of phytochemicals considered as active medicinal chemical constituents. Important medicinal phytochemicals such as terpenoids, reducing sugar, flavonoids, alkaloids and phlobatannins were present in the samples. The result of the phytochemical analysis shows that the plant is rich in at least one of alkaloids, flavonoids, terpenoids, reducing sugars and phlobatannins. Plant *Glochidion ellipticum* having all these phytochemicals. The phytochemical screening and qualitative estimation of plant studied showed that the leaves were rich in phlobatannins, terpenoid, flavonoids, alkaloids and reducing sugar. Phlobatannins are present in *Glochidion ellipticum*. Phlobatannins have been reported for its wound healing properties, these are anti-inflammatory and analgesic⁹ and antioxidant¹⁰. Reducing sugars are present. Terpenoids are present and are reported to have anti-inflammatory, anti-viral, anti-malarial, inhibition of cholesterol synthesis and anti-bacterial¹¹. Flavonoids are also present. Plants having alkaloids are used in medicines for reducing headache and fever. These are attributed for antibacterial and analgesic properties¹². Macro and microscopic characterization was also carried out.

DISCUSSION

The present work was carried out on the *Glochidion ellipticum* which contain flavonoids, alkaloids, reducing sugars and phlobatannins are present. On further study T.S. Showed presence of Parenchyma cell, sclerenchyma tissue powder showed presence of vascular bundles, stomata, calcium oxalate and starch grain also acid-insoluble ash value of bark of *Glochidion ellipticum* was found to be 20% and total ash value was found to be 60%.

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