ABSTRACT

Background: Medicinal plants are important remedy for several communicable and non-communicable diseases and have been used since time immemorial. Euphorbia cotinifolia L. (Family: Euphorbiaceae) is an ornamental plant grows in gardens and in pots.

Purpose of the study: The aim of the current study was to screen the various phytochemicals from the aqueous, methanol, benzene, chloroform and ethanolic extracts of leaves, stems and fruits of E. cotinifolia.

Methods: The extracts were subjected to qualitative phytochemical screening using standard procedures. Results: Five different extracts of leaves, stem and fruits of E. cotinifolia were found to contain various secondary metabolites like aminoacids, anthroquinones, glycosides, saponins, steroids, polyphenol and tannins, triterpenoids and flavonoids. Conclusion: The phytochemicals generated data from the five different extracts of E. cotinifolia could be used as tools for quality control of drugs in the future, for the healing of a diversity of disease conditions.

KEYWORDS: Phytochemical screening, Euphorbia cotinifolia L., Secondary metabolites.
INTRODUCTION
The advantageous of many traditional plants use for various dreaded diseases, which have been depicted by traditional herbal medicinal practitioners since time immemorial. Natural products are the primary foundation of synthetic and conservative herbal medicine. These natural medicines are greatly protective as well as environment friendly. According to WHO, about 75% of the communities throughout the world relies on traditional medicine for their primary health care.[1] These are bioactive constituents of plant origin, which are considering as secondary metabolites. Naturally, these bioactive chemicals are synthesised in all parts of the plant body i.e., bark, leaves, stem, root, flower, fruits and seeds.[2] The presence of bioactive chemicals present in the plant parts, which may vary from one part to another by qualitatively and quantitatively. Moreover, the biochemical activity of medicinal plants are greatly depends on the distribution of active principles, which are more rich in some parts of the plants.[3] The successful screenings of these bioactive compounds isolated from plants are principally dependent on the various solvent used in the extraction methods.[2] Therefore it emphasizes that different solvent attempt are mandatory to screen the plant parts for phytochemicals.

_Euphorbia cotinifolia_ L. (Family: Euphorbiaceae) is an ornamental shrub or tree, grows more than 2 metres in gardens or and in pots. The milky latex found in all parts of the plant is strongly purgative. That from the roots is more poisonous than other parts of the plant. It is employed as a fish and arrow poison. The sap is caustic to the peel, causing rashes and blisters. The whole plant is applied topically to remedy sores. The milky latex is strongly purgative. It is used externally to treat infected nails.[4] The family, Euphorbiaceae encompasses more than 300 genus and 5000 species distributed mainly in America and tropical Africa.[5] This family contains skin irritating and tumour-promoting terpenoids. Various species of this family are used in traditional medicine to treat dermatitis, sexual transmitted diseases, headache, intestinal worms and warts.[6] Moreover, various types of terpenoids have been isolated and characterized from the genus, which are used for antibacterial, anticancer, anti-multidrug-resistant, antifeedant, anti-HIV and analgesic activity.[7] In the present study, various solvent extracts of leaves, stem and fruits of _E.cotinifolia_ were qualitatively screened for phytochemicals using standard tests.
MATERIALS AND METHODS

Collection of Plant Materials
Leaves, stem and fruits of *E. cotinifolia* were collected from the Botanical Garden, Department of Biomedical science, Jimma University, Jimma, Ethiopia. The plant materials were washed with tap water, clean with distilled water and blotted smoothly between the folds of filter paper.

Processing of Plant Materials
Leaves, stem and fruits of *E. cotinifolia* were dried for 12hrs in a hot air oven at 60°C. The dried leaves materials were ground using an electric blender to obtain a fine powder. The powder was additionally passed through a 2mm filter to get fine particles. The powdered samples were stored in a fresh glassware container until required for analysis.

Preparation of extracts
Aqueous, methanol, benzene, chloroform, and ethanolic extracts of leaves of *E. cotinifolia* were prepared in 10g/100 ml. The solvents of organic extracts were dried at 60°C protected from light. The residue was weighed and solubilised in 50ml of dimethyl sulphoxide (DMSO). These extracts were used for the screening of preliminary phytochemical analysis.

Screening procedure
1. *Test for alkaloids*
About two ml of the plant extract was added to 2 ml of hydrochloric acid in a test tube. To this acidic medium, 1 ml of Dragendroff’s reagent was added. An orange or red precipitate produced immediately indicates the presence of alkaloids.

2. *Test for amino acids*
About one ml of the extract was added with few drops of Ninhydrin reagent in a test tube. Appearance of purple colour shows the presence of amino acids.

3. *Test for anthraquinones*
About five ml of the plant extract was hydrolysed with diluted Conc. H₂SO₄ extracted with benzene. 1 ml of dilute ammonia was added to it. Rose pink coloration suggested the positive response for anthraquinones.
4. Test for flavonoids
About one ml of the extract, a few drops of dilute sodium hydroxide was added. An intense yellow colour was produced in the plant extract, which become colourless on addition of a few drops of dilute acid indicates the presence of flavonoids.

5. Test for glycosides
The extract was hydrolysed with hydrochloric acid for few hours on a water bath. To the hydrolysate, 1ml of pyridine was added and a few drops of sodium nitroprusside solutions were added and then it was made alkaline with sodium hydroxide solution. Appearance of pink to red colour shows the presence of glycosides.

6. Test for saponins
The extract was diluted with 20 ml of distilled water and it was agitated in a graduated cylinder for 15 minutes. The formation of 1cm layer of foam showed the presence of saponins.

7. Test for steroids
About one ml of the extracts was dissolved in 10 ml of chloroform and equal volume of concentrated sulphuric acid was added by sides of the test tube. The upper layer turns red and sulphuric acid layer showed yellow with green fluorescence. This indicated the presence of steroids.

8. Test for tannins
About two ml of the extract and a few drops of 1% lead acetate were added. A yellow precipitate was formed, indicates the presence of tannins.

9. Test for triterpenoids
About two ml of the extract was dissolved in 1 ml of chloroform; 1 ml of acetic anhydride was added following the addition of 2 ml of Conc. H₂SO₄. Formation of reddish violet colour indicates the presence of triterpenoids.

RESULTS AND DISCUSSION
In the present investigation, preliminary phytochemical investigation has been done in the aqueous, methanol, benzene, chloroform and ethanolic extracts of *E.cotinifolia* leaves, stem and fruits showed the presence of phytochemical constituents namely alkaloids, aminoacids,
anthraquinones, flavonoids, glycosides, saponins, steroids, tannins and triterpenoid shown in Table 1.

The initial phytochemical screening tests may be helpful in the screening of the bioactive compounds and eventually may help to detection and development of new drugs. Further, these tests make easy their qualitative separation and quantitative estimation of pharmacologically active chemical compounds.[8] The phytochemical screening in the present study has publicized the presence of alkaloids, anthraquinones, flavonoids, glycosides, saponins, steroids, tannins and triterpenoids in the leaves extract. Further the presence of different phytochemicals in the five different organic solvent extracts may be responsible for the therapeutic properties of E.cotinifolia.

Tannins and Flavonoids are phenolic compounds that are acting as principal antioxidants or free radical scavengers. Since these phenolic compounds were originated to be present in the extracts, it might be accountable for the potent antioxidant capacity of E.cotinifolia. These phytochemicals of medicinal plants have primarily reported for their medicinal value, which can be valuable for therapeutic index. For instance, saponins and glycosides proved as hypotensive and cardiodepressant properties[9], which are helpful for the treatment of congestive heart failure and cardiac myopathy.[10] The occurrence of saponins and glycosides in aqueous, methanol and ethanolic extracts leaves and stems of E.cotinifolia might play a role in the cardioprotective potential. Tannins and alkaloids have the potential of anti-hyperglycaemic and anti-inflammatory activities.[11] Moreover, the terpenoids have also been revealed to decrease blood sugar level in animal studies.[12] In addition, the steroids and triterpenoids demonstrated the analgesic properties and central nervous system activities.[12-14] Hence the initial phytochemical studies are helpful in finding chemical constituents in the plant material that may help to their quantitative assessment and also in locating the source of pharmacologically active chemical compound.
### Table 1: Phytochemical investigation of leaves, stem and roots of *Euphorbia cotinifolia* L. using aqueous, methanol, hexane and ethanol solvents.

<table>
<thead>
<tr>
<th>Analysis</th>
<th>Aqueous</th>
<th>Methanol</th>
<th>Benzene</th>
<th>Chloroform</th>
<th>Ethanol</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Leaves</td>
<td>Stems</td>
<td>Fruits</td>
<td>Leaves</td>
<td>Stems</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td>Protein and aminoacids</td>
<td>++</td>
<td>+++</td>
<td>+++</td>
<td>-</td>
<td>-</td>
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<tr>
<td>Anthraquinones</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
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<td>Flavonoids</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
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<tr>
<td>Glycosides</td>
<td>-</td>
<td>+++</td>
<td>+</td>
<td>-</td>
<td>-</td>
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<tr>
<td>Saponins</td>
<td>-</td>
<td>+++</td>
<td>-</td>
<td>+</td>
<td>-</td>
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<tr>
<td>Steroids</td>
<td>+++</td>
<td>+++</td>
<td>-</td>
<td>+++</td>
<td>-</td>
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<tr>
<td>Total phenols and Tannins</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>Triterpenoids</td>
<td>+++</td>
<td>+++</td>
<td>-</td>
<td>+++</td>
<td>+</td>
</tr>
</tbody>
</table>

+++ = appreciable amount (positive within 5 mins.); ++ = moderate amount (positive after 5 mins. but within 10 mins); + = trace amount (positive after 10 mins. but within 15 mins); - = completely absent.

**CONCLUSION**

The results of phytochemical screening showed the leaves, fruit and stem extracts of *E. cotinifolia* indicates their potential as a source of bioactive principles that may supply drugs for modern medicines. Further studies are therefore required to validate their antimicrobial, antihyperglycemic, anti-inflammatory and antihelminthic activities. In addition, extraction, cleansing and categorization of the active principles are required to make the plant has novel interesting studies.
ACKNOWLEDGMENT
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REFERENCES
