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PHYTOCHEMICAL EVALUATION OF THE ETHANOLIC AND AQUEOUS LEAF EXTRACTS OF *ERYTHROXYLUM MOONII* (HOCHR), *HENCKELIA HUMBOLDTIANA* (GARDNER) A.WEBER & BURTT AND *CIPADESSA BACCIFERA* (ROTH) MIQ

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ABSTRACT

Qualitative phytochemical screening of *Erythroxylum moonii*, *Henckelia humboldtiana* and *Cipadessa baccifera* were carried out in ethanolic and aqueous media. The study confirms the presence of secondary metabolites such as saponins, terpenoids, steroids, tannin, alkaloids, flavonoids and phenols. The result suggests that the phytochemical properties for curing various ailments possess potential, antimicrobial and antioxidant and leads to the isolation of new and novel compounds.

Key words: Phytochemicals, Antimicrobials, Antioxidant, Secondary Metabolites.

INTRODUCTION

Medicinal plants are the richest bio-resources of traditional systems of medicine, food supplements, nutraceuticals, pharmaceutical industries and chemical entities for synthetic drugs [1]. Modern medicine has evolved from folk medicine and traditional system only after through chemical and pharmaceutical screening [2]. India is a birth place of renewed system of indigenous medicine such as Siddha, Ayurveda and Unani. The traditional systems of medicines are prepared from a single plant or combinations of number of plants. The efficacy depends on the use of proper plant part and its biological potency which in turn depends upon the presence of required quantity and nature of secondary metabolite in a raw drug [3]. There is growing awareness in correlating the phytochemical constituents of a medicinal plant with its pharmacological activity. The screening of phytochemical constituents from plants has led to the invention of new therapeutic drugs which have efficient protection and treatment roles against various pathogens [4].

Phytochemicals are responsible for medicinal activity of plants [5]. These are non-nutritive chemicals that

have protected human from various diseases.

Phytochemicals are basically divided into two groups as primary and secondary metabolites based on their function in plant metabolism. Primary metabolites comprise of common carbohydrates, amino acids, proteins and chlorophylls while secondary metabolites consist of alkaloids, saponins, steroids, flavonoids and tannins [6]. The phytochemical constituents are the basic source for the establishment of several pharmaceutical industries. And they are playing a significant role in the identification of crude drugs [5].

MATERIALS AND METHODS

Collection and Identification of plant material

The plant materials were collected from the Palni hills of the Western Ghats, Tamilnadu on 11/05/2014. The taxonomic identification of the plant was carried out by S. John Britto, Director, and Head, The Rapinat Herbarium and centre for Molecular systematic, St. Joseph's College Tiruchirappalli, India. Voucher specimens were deposited at the centre (RHT 65283, RHT 65282, and RHT 65280).

Plant extraction

The leaves were shade dried at room temperature and ground into powder. The serial extraction method was followed. First 10gm of plant powder was dissolved in 70ml of chloroform solvent and then it was kept in a rotary shaker for 3 days. The suspension was filtered by using filter paper of pore size 0.2µm. The antibacterial study was carried out using the crude extract. The same procedure was followed for solvents like acetone, ethanol and aqueous media.

Phytochemical Screening of crude extracts procedure

The leaves of the plant were tested for bioactive compounds according to standard procedure (Harborne, (1998) and Kokate, (1986) [7].

Test for Starch

3 ml of test solution was added with few drops of dilute iodine solution. Blue colour appearance showed the presence of starch.

Test for Glycosides

To the extract, glacial acetic acid and few drops of 5% ferric chloride and conc. Sulphuric acids were added and a reddish brown colouration was observed at the junction of two layers and bluish green colour in the upper layer indicating the presence of glycosides.

Test for Tannin

About 0.5gm of the extract was boiled in 10ml of water in a test tube and then filtered. A few drops of 0.1% ferric chloride were added and observed for brownish green of a blue- black colouration. The brownish green or blue – black colouration indicated the presence of tannin.

Test for Phenol

To 1ml of the extract, 2ml of distilled water followed by a few drops of 10% ferric chloride was added. Formation of blue or green colour indicates presence of phenols.

Test for Saponins

To 0.5gm of extract was added 5ml of distilled water in a test tube. The solution was shaken vigorously and observed for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously after which it was observed for the formation of an emulsion.

Test for Alkaloids

2ml of ethanol extract was measured using a measuring cylinder and equal volume of ethanol containing 3% tartaric acid was added and shaken. The few drops of marquin's reagent were added into the mixture. The formation of precipitate indicated the presence of alkaloids.

Test for Flavonoids

Three methods were used to test for flavonoids. First, dilute ammonia (5ml) and conc. sulphuric acid (1ml) were added to a portion of an aqueous filtrate of the extract. A yellow colouration was found to disappear indicating the presence of flavonoids. Second, a few drops of 1% aluminium solution were added to a portion of the filtrate. Formation of the yellow colouration indicated the presence of flavonoids. Third, a portion of the extract was heated with 10ml of ethyl acetate over a steam bath for 3 min. The mixture was filtered and 4 ml of the filtrate was shaken with 1ml of dilute ammonia solution. Formation of a yellow colouration indicated the presence of flavonoids.

Test for Steroids

2.0gm extract was dissolved in 10ml chloroform and filtered. 2ml of filtrate was measured and placed in two different test tubes. Two drops of conc. H₂SO₄ were added to one of the test tubes and five drops of acetic anhydride followed by five drops of conc. H₂SO₄ were added to the other test tube for confirmation. A reddish brown colouration of the interface indicated the presence of steroids.

RESULT AND DISCUSSION

The Palni hills have a rich variety of medicinal plants. The inhabitants and herbal practicioners in the study area are aware of the uses of many medicinal plants. The extracts were examined for their physical characterization like colour, odor and consistency. The colour of the aqueous extracts of the experimental samples were yellowish brown and while ethanolic extracts showed the colour of green and dark green. The consistency level of all the extracts were semi-solids and the odor were characteristics. Presence of odor showed the presence of desired phytochemicals. The result of the above study is compiled in Table 1. Different chemical tests were performed to determine the nature of the chemical constituents.

The dual phytochemical screening (aqueous and ethanolic) of the extracts of *Erythroxylum moonii* (leaf) revealed that phenol, tannins and flavonoids were present in both the extracts and while saponin and steroids were present only in aqueous extract and while starch and glycosides were completely absent from both the extracts (Table2).

Henckelia humboldtiana (leaf) in the dual phytochemical screening revealed the presence of phenol, steroids flavonoids and alkaloids in both extracts. Starch and tannins were present only in aqueous extracts and while the ethanolic extract showed the absence of them (Table 2).

Cipadessa baccifera (leaf) in the dual phytochemical screening revealed the presence of glycosides, flavonoids and alkaloids in both extracts. Phenol and saponin were present only in aqueous extracts and while steroids and starch were completely absent from both the extracts (Table 2).

Therefore, the medicinal value of the plants lies in some phytochemical compounds that have a definitive phytochemical action in the human body. Different

phytochemicals have been found to possess a wide range of activities, which may help in protection against human pathogens.

Table 1. Phytochemical Characteristics of the extracts

Name of the Extracts	Name of plant	Parts used	Consistency	Colour	Odor
Ethanolic extract Aqueous extract	<i>Erythroxylum moonii</i>	Leaf	Semi-solid Semi-solid	Dark green Greenish brown	Characteristic Characteristic
Ethanolic extract Aqueous extract	<i>Henckelia humboldtiana</i>	Leaf	Semi-solid Semi-solid	Dark green Greenish brown	Characteristic Characteristic
Ethanolic extract Aqueous extract	<i>Cipadessa baccifera</i>	Leaf	Semi-solid Semi-solid	Green Greenish brown	Characteristic Characteristic

Table 2. Phytochemical tests in the aqueous and ethanolic extracts

S. No.	Phytochemical constituents	<i>E. moonii</i>		<i>H. humboldtiana</i>		<i>C. baccifera</i>	
		Ethanol	Aqueous	Ethanol	Aqueous	Ethanol	Aqueous
1.	Starch	-	-	-	+	-	-
2.	Glycosides	-	-	-	-	+	+
3.	Tannins	+	+	-	+	+	+
4.	Phenol	+	+	+	+	-	+
5.	Saponins	-	+	-	+	-	+
6.	Alkaloids	-	-	+	+	+	+
7.	Flavanoids	+	+	+	+	+	+
8.	Steroids	-	+	+	+	-	-

(+) Presence of active constituent and (-) Absence of active constituent.

CONCLUSION

The presence of phytoconstituents justify the use of selected species for treating different ailments and have a potential of providing useful drugs of human use. In the present study, it is seen that most of the biologically active phytochemicals were present in both ethanolic and aqueous extract of *E. moonii* (leaf), *H. humboldtiana* (leaf) and *C. baccifera* (leaf). Since both the extracts revealed

phytoconstituents, they can be considered beneficial for further investigation. Exploitation of these pharmacological properties involves further investigation of these active ingredients by implementation of techniques like extraction, purification, separation and crystallization. And also a proper documentation of such medicinal plants along with their conservation is a dire need in the study area.

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