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Preliminary Phytochemical Screening of roots of *Coleus forskohlii*

Kambham Venkateswarlu[1*], N. Devanna[2], K.B. Chandrasekhar[3]

[1]*Department of Pharmaceutics, JNTUA-Oil Technological Research Institute, Anantapur-515001, Andhra Pradessh, India.*

[2]*Director & Professor of JNTUA-Oil Technological Research Institute, Anantapur-515001, Andhra Pradessh, India.*

[3]*Director of Research and Development & Professor of Chemistry, JNTUA, Anantapur-515 002, Andhra Pradessh, India*

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Abstract

The main objective of the present study was to develop the preliminary phytochemical screening of roots of *Coleus forskohlii*. This study has clearly given information about what are the phytoconstituents present in the root. These phytoconstituents also called as active compounds, which may have different pharmacological actions. The main constituent of *Coleus forskohlii* was forskolin.

Keywords: *Coleus forskohlii*, Lamiaceae, Phytoconstituents

Contents

***Corresponding author**
Kambham Venkateswarlu
Department of Pharmaceutics,
JNTUA-Oil Technological Research Institute, Anantapur-515001, A.P, India.
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1. Introduction

1.1. Botanical Classification:

Kingdom	:	Plantae
Phylum	:	Angiospermae
Class	:	Dycotyledons
Order	:	Tubiflora
Family	:	Lamiaceae
Genus	:	*Coleus*
Species	:	*forskohlii*

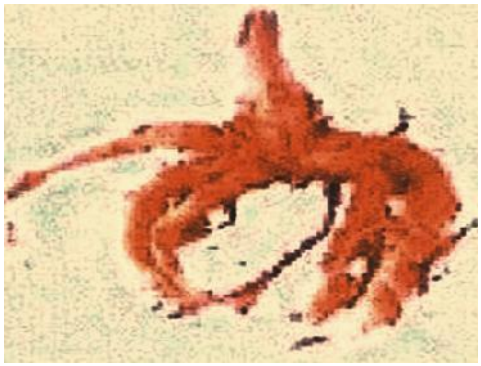

Figure 1*: Coleus forskohlii root*

Coleus forskohlii is a botanical plant that has been used since ancient times in Hindu and Ayurvedic traditional medicine. In olden days, the mixtures of root extract solution (few drops) and milk used for the treatment of abdominal pain and constipation in children. The plant produces the labdane diterpeniod, forskolin in its tuberous roots. Forskolin directly stimulates the catalytic sub-unit of adenylate cyclise and hence affects heart action [positive intropic effect) lowers BP and has anti-inflammatory properties. In vivo testing of forskolin in human cardiac output was eventually used in the treatment of congestive heart failure. This inhibits aggregation of human blood platelets and has the potential to be employed in the treatment of thrombo-embolic platelet disorders. Closely related to this activity would be its employment in the treatment of tumour metastases that apparentlydepend on thrombus formation in establishing colonies in the capillary beds of various organs. Forskolin has been studied as a possible bronchodilator that used in the treatment of Asthma. Topical application of the compound lowered intraocular pressure in human volunteers pointing to the use of forskolin in treatment of glaucoma.

2. Materials and Methods

2.1. Plant collection:

Coleus roots were harvested in autumn.

2.2. Extraction Procedure:

Two types of extracts were prepared, which are namely,

A] Ethanolic extract

B] Aqueous extract

Steps:

a. Remove the foreign matter from the collected root and clean it followed by dry it.
b. Now the roots were made into powder.
c. The powdered crude drug (52 gm) was packed in a thimble or a paper.
d. Then the extraction was carried out by using soxhlet apparatus.
e. 25mg of Coleus root powder was extracted by using suitable solvent of 250ml of 95% absolute alcohol.
f. The solvent used in the extraction was ethanol.
g. Now remove the excess alcohol by using distillation.
h. Store the obtained residue in the dessicator.
i. Finally the obtained extract was used for preliminary phytochemical screening.

2.3. Preliminary Phytochemical Study:

The plants may be considered as biosynthetic laboratories for human consumable chemical compounds [carbohydrates, proteins, lipids] which were taken in the form of food and therapeutically active chemical compounds[like glycosides, alkaloids, volatile oils, tannins which exerts physiological effect]. The compounds which are responsible for therapeutic effects are usually considered as cellular secondary metabolites.

2.3.1. Qualitative Chemical Tests:

The *coleus forskohlii* extract was subjected to following qualitative tests for identification of chemical groups present in the extract (See Table No.1).

2.3.1.1. Alkaloids
2.3.1.2. Flavonoids
2.3.1.3. Glycosides
2.3.1.4. Carbohydrates
2.3.1.5. Proteins & Amino acids
2.3.1.6. Phytosterols
2.3.1.7. Saponins
2.3.1.8. Phenolic compounds

2.3.1.9. Fixed oils
2.3.1.10. Gums & Mucilage
2.3.1.11. Aromatic acids
2.3.1.12. Terpenoids

2.3.1.1. Detection of Alkaloids:

A small quantity of extract is treated with few drops of dilute HCl and filtered. The filtrate was used for the following tests.

A) Mayer's test
B) Dragendroff's test
C) Wagner's test
D) Hager's test

A) Mayer's test:

Filtrate was treated with Mayer's reagent. Cream colour precipitate was produced.

B) Dragendroff's test:

The filtrate was treated with Dragendroff's reagent. Orange brown precipitate was obtained.

C) Wagner's test:

The filtrate was treated with Wagner's reagent. Reddish brown precipitate was produced.

D) Hager's test:

The filtrate was treated with Hager's reagent. Yellow colour precipitate was obtained.

E) The filtrate was treated with tannic acid. White precipitate was produced.

2.3.1.2. Detection of Flavonoids:

A) Shinoda test:

The extract was treated with one gram Magnesium turnings and few drops of concentrated HCl and boiled for 5 minutes. Formation of orange colour shows presence of flavonoids.

Small portion of each is dissolved in alkali, yellow colour was produced.

2.3.1.3. Detection of Glycosides:

2.3.1.3.1. Test for Cardiac Glycosides:

A) Keller Killani test
B) Legal's test
C) Baljet test

A) Keller Killani test:

- The Extract was dissolved in 3ml of glacial acetic acid.
- To this about 2 drops of ferric chloride were added.
- The contents are then transferred to a test tube containing 2 ml of concentrated H_2SO_4. Reddish colour was formed at the junction of two layers.

B) Legal's test:

To the extract add 1 ml of sodium nitroprusside and sodium hydroxide. Colour changes from pink to red colour.

C) Baljet test:

To the extract add 1 ml of sodium picrate. A colour change from yellow to orange was observed.

2.3.1.3.2. Test for Anthraquinone Glycosides:

A) Borntrager's test:

About 1 ml extract was boiled with dilute sulphuric acid and filtered. The filtrate was extracted with chloroform. The chloroform layer was separated and equal quantity of dilute ammonia was added. Formation of pink or red colour in organic layer indicates presence of glycosides.

B) A little amount of solution was treated with 5N Sodium hyposulphite. Appearance of red colour indicates the presence of glycosides.

2.3.1.4. Detection of Carbohydrates:

A minimum of various extracts was taken and dissolved in respective solvents and treated with HCl to detect the presence of Carbohydrates by following tests.

A) Molisch's test
B) Fehling's test
C) Benedict's test
D) Barfoed's test
E) Seliwanoff's test

A) Molisch's test:

- The extraction solution was treated with 2-3 drops of alcoholic α-naphthol.
- Along the sides of test tubes, the concentrated H_2SO_4 (2 ml) was added.
- If the violet colour appears, thisindicates the presence of Carbohydrates.

B) Fehling's test:

- The 2 ml of extraction solution was treated with Fehling's solution
- Then it was heat on a water bath.
- If the reddish orange precipitate produced, this indicates the presence of carbohydrates.

D) Barfoed's test:

2 ml of solution was treated with Barfoed's reagent and boiled on water bath. Formation of red colour indicates the presence of carbohydrates.

E) Seliwanoff's test:

2ml solution was treated with few ml of seliwanoff's reagent and boiled on water bath. Red colour was formed.

2.3.1.5. Detection of Proteins and Amino acids:

A) Biuret test
B) Millon's test
C) Ninhydrin's test

A) Biuret test:

1ml solution was treated with equal volume of 5% sodium hydroxide and 1% copper sulphate solution. A violet colour was produced.

B) Millon's test:

1ml solution was treated with Millon's reagent and heated. A white colour changes into red colour upon heating indicates presence of amino acids.

C) Ninhydrin's test:

- 1 ml of extraction solution was treated with ninhydrine reagent and then heated.
- The Purple colour was produced.

2.3.1.6. Detection of Phytosterols:

A small quantity of extract was dissolved in 5ml chloroform. The solution is subjected to tests like

A) Salkowski's test
B) Libermann-buchards test
C) Zark's test

A) Salkowski's test:

To 1ml solution few drops of concentrated sulphuric acid is added. Brown colour was produced.

B) Libermann-Buchards test:

The prepared chloroform solution was treated with 2 drops of concentrated sulphuric acid followed by 3 drops of acetic anhydride. Emerald green colour formed.

C) Zark's test:

To 1 ml solution mixture of glacial acetic acid, ferric chloride and concentrated sulphuric acid was added. Purple colour is produced.

2.3.1.7. Detection of Saponins:

A) Foam test
B) Haemolysis test

A) Foam test:

a. Dilute the extract with 20ml of distilled water.
b. Then this mixture was transferred into graduated cylinder.
c. This was agitated for about 15 minutes.
d. The appearance of stable foam indicates the presence of saponins.

B) Haemolysis test:

- The thin film was made by spreading the extract on the glass slide.
- Then one drop of human blood was added on to the thin film.
- Then go for microscopic observation for knowing the change in structure of R.B.C. due to the haemolysis.

2.3.1.8. Detection of Phenolic compounds:

A) Test for Phenolic compounds:

- To the extract, the ferric chloride was added.
- The precipitate was turned into a blue colour due to the presence of phenolic compounds.

B) Test for Tannins:

- To the extract the 1% of gelatine and 10% of NaCl were added, now the white precipitate was formed.
- On heating with lead acetate, the presence of white precipitate in test solution indicates the presence of tannins.
- It produces a brown colour on treatment with Dramer's reagent.

2.3.1.9. Detection of Fixed oils:

- Between the two filter papers, small quantity of extracted solution was pressed

- It was treated with Sudan red 3 (or) Tincture of Alkana, which produces red colour.

2.3.1.10. Detection of Gums and Mucilage:

a. To the extract, the Rheuthenium red solution was added.
b. If the pink colour is formed, this indicates the presence of mucilage in the extract.
c. If treated with Methylene blue produces the deep blue colour.
d. If treated with Iodine and sulphuric acid produces the violet colour.

2.3.1.11. Detection of Aromatic acids:

- Tothe extractedsolution (small quantity), Neutral Ferric Chloride was added.
- If the aromatic acid present, a buff colour precipitate was formed.

2.3.1.12. Test for Triterpenoids:

- Few drops of Antimony Chloride were added to the extract.
- If the blue precipitate appears, this denotes the presence of triterpenoids.

3. Results and Discussion

Table 1: Preliminary Phytochemical Screening of *Coleus forskohlii* Extract

S.No	Plant Constituents	Ethanolic Extract	Aqueous Extract
1	Alkaloids	P	A
2	Carbohydrates	A	A
3	Glycosides	A	A
4	Saponins	A	A
5	Proteins & Amino acids	A	A
6	Phenolic compounds & Tannins	P	A
7	Gums & Mucilage	A	P
8	Flavonoids	P	P
9	Fixed oils	P	P
10	Volatile oils	P	A
11	Diterpenoids	P	P
12	Phytosterols	A	A
13	Aromatic acids	A	A

Note: P-Present, **A**-Absent

There was a slight change in presence of chemical constituents in both types of extracts.

4. Conclusion

In the present study an attempt has been made to carryout preliminary phytochemical screening of the alcoholic extract of Coleus forskohlii and shown the presence of Alkaloids, Flavonoids, Phenolic Compounds, Gums, Mucilage, Tannins, and Diterpenoids in the extract. The preliminary phytochemical screening shown the presence of active compounds those have pharmacological action.

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