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Qualitative Determination of Phytochemicals in Anticancer Plant *Catharanthusroseus*(L)

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ABSTRACT

Phytochemical characterization of plants is an important task in medicinal botany and drug discovery. In the current study, *Catharanthusroseus* is considered as medicinal plants by evaluation of phytochemical composition. Methanolic extract of the leaves of *C. roseus* was prepared by soxhlet extraction. The extract was used to detect the presence of different phytochemicals like alkaloids, phenols, flavonoides, tannin, lignin, steroids, glycosides, saponins, terpenoides and anthraquinone by their biochemical tests. Alkaloids, tannin, steroids, glycosides were found to be present in methanolic extract of *C. roseus*.

Keywords: *Catharanthusroseus*, phytochemicals, alkaloids, tannin, steroids, glycosides

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INTRODUCTION

Catharanthus a genus of flowering plant in the dogbane family, apocynaceae like genus vinca, and commonly known as periwinkle. It shows anti-diabetic, anti-bacterial, anti-oxidant, anti-ulcer and anti-diarrheal properties. *C. roseus* is an ornamental shrub that grows up to 30–100 cm in height. It was previously known as *Vincarosea* and commonly known as Madagascar periwinkle. Although this plant originated from Madagascar, it is widely distributed around the World due to its high survivability in a variety of habitats and use as an ornamental plant¹. These are used for various purposes such as pharmaceuticals, food additives; dyes etc.² This plant has a long history as a folk medicine in many countries³ such as South Africa, China, India, Mexico⁴ and Malaysia,⁵ where it is utilized as a remedy to alleviate diabetes complications.⁶ The antioxidant activity of this plant was usually assessed along with their phenolic content as it exhibited high linear correlation with activity.^{7, 8}

Potentially Active Chemical Constituents:

C. roseus possess carbohydrate, flavinoid, saponin and alkaloids. Alkaloids are the most potentially active chemical constituents of *C. roseus*. More than 400 alkaloids are present in the plant, which are used as pharmaceuticals, agrochemicals, flavor and fragrance, ingredients, food additives and pesticides. The alkaloids like actinoptamide, vinblastin, vincristine, vindesine, vindelinetabersonine etc. are mainly present in aerial parts whereas ajmalicine, vinceine, vineamine, raubasin, reserpine, catharanthine etc. are present in roots and basal stem. Rosindin is an anthocyanin pigment found in the flower of *C. roseus*.⁹

Therapeutic Properties/Uses:

Anti-cancer property

The anticancer alkaloids Vinblastine and Vincristine are derived from stem and leaf of *C. roseus*. These alkaloids have growth inhibition effect to some human tumors. Vinblastine is used experimentally for treatment of neoplasmas and is recommended for Hodgkins disease, chorio carcinoma. Vincristine another alkaloids is used to treat leukemia in children. Vinblastine is sold as Velban or Vincristine as oncovin.¹⁰

Anti bacterial property

Crude extracts from different parts of the plant was tested for anti-bacterial activity. Extract from leaves showed significantly higher efficacy. The anti bacterial activity of the leaf extract of the plant was checked against micro organism like *Pseudomonas aeruginosa* NCIM2036, *Salmonella typhimurium* NCIM2501, *Staphylococcus aureus* NCIM5021 and it was found that the extracts

could be used as the prophylactic agent in the treatment of many of the disease.

Anti-helminthic property

Helminthes infections are the chronic illness, affecting human beings and cattle. *C. roseus* was found to be used from the traditional period as an antihelminthic agent.

Anti ulcer property

Vincamine and vindoline alkaloids of the plant showed anti ulcer property.

Anti diarrheal property

The anti diarrheal activity of the plant ethanolic leaf extracts was tested in the wistar rats with castor oil as experimental diarrhea inducing agent in addition to the pretreatment of the extract. The anti diarrheal effect of ethanolic extracts *C. roseus* showed the dose dependent inhibition of the castor oil induced diarrhea.¹¹

Anti-oxidant property

The antioxidant potential of the ethanolic extract of the roots of the two varieties of *C. roseus* namely rosea (pink flower) and *C. alba* (white flower) was determined by using different system of assay. It was found that *C. roseus* possess more antioxidant activity than that of *C. alba*.¹²

Anti diabetic property

Traditional plant treatment have used throughout the world for the therapy of diabetes mellitus. In total 419 useful phyto recipes of 270 plants species belonging to 74 angiospermic families were collected¹³.

MATERIALS AND METHOD

Apparatus:

Test tube, graduated cylinder, whatman filter paper, funnel, bleaker, pestle and mortar, waterbath, soxhlet apparatus, crucible, separator funnel, conical flask, magnetic stirrer, refrigerator.

Plant material:

The present study was carried out on *C. roseus*, traditionally important herb growing in India. Phytochemical and pharmacological activity was identified in *C. roseus* leaf.

METHOD:

Collection of plant:

Mature plant can grow up to 100 cm or more high and the plant session is evergreen. Firstly plant is collected the rod side field. Shade dried the sample and powder was prepared with the help of the blender.

Preliminary screening of secondary metabolites

Extraction

The shade dried leaves material was powdered using mixer grinder and subjected to soxhlet extraction with methanol for 18 hrs. The solvent was evaporated by using steam water bath and extract was weighted. The condensed extract was used for preliminary screening of phytochemical.

Alkaloids^{14, 15, 16}:

Mayers' Reagent:

Dissolve 1.358 g of HgCl₂ in 60 ml of water and pour into a solution of 5 g of KI in 10 ml of H₂O. Add distilled water to make the volume 100 ml. 2 ml of extract was treated with 1 ml of 1 % HCl and boiled for few minutes. 1ml of the above mixture was treated with 6 drops of Mayer reagent/Wagner's reagent /Drangondoff's reagent. The formation of creamish precipitated/orange precipitate indicated the presence of respective alkaloids.

Phenol test^{17, 18, 16}:

The crude extract was mixed with 2 ml of 2 % solution of FeCl₃, a blue green or black coloration indicated the presence of phenols.

Flavonoids

Alkaline reagent test^{17, 18, 16}:

The crude extract was treated with 2 ml 2% solution of NaOH. An intense yellow colour formed which turned colourless on addition of diluted acid, indicated the presence of flavonoids.

Tannin^{17, 14, 15}:

Crude extract was mixed with 2 ml 2 % solution of FeCl₃. A blue green or black coloration indicated the presence of tannins.

Lignins test^{17, 18, 16}:

Add 2% furfuraldehyde in the extract. Formation of red color indicated the presence of lignin.

Steroid:

Salkowski's test^{17, 18, 16}:

The crude extract was mixed with 2 ml of chloroform. Then 2 ml of conc. H₂SO₄ was added carefully and shaken gently. The reddish brown colour indicated the presence of steroids.

Glycolysis^{20, 21, 19}:

Crude extract was mixed with 2 ml of glacial acetic acid containing 1-2 drops of 2% solution of FeCl₃. The mixture was then poured into another test tube containing 2 ml conc. H₂SO₄. Formation of the brown ring at the interphase indicated the presence of glycolysis.

Saponins^{18, 16}:

The crude extract was treated with 5 ml of distilled water in test tube. It was shaken vigorously the formation of stable foam indicated for the presence of saponins.

Terphenoids(Salkoweki test)²²:

The crude extract was mixed with 2 ml of conc. H₂SO₄ and shaken gently. The reddish brown coloration at the interface showed the presence of terpenoids.

Anthraquinone:

The 2 ml of crude extract mixed with the 2 ml of the 25% ammonia solution and shake well. The red colour indicated the positive for the anthraquinone.

Powder analysis:

Vital appearance of plant and its powder to the naked eyes and its sensory characteristic like odour, taste, colour, physical appearance and touch.

RESULTS AND DISCUSSION

Results observed after performing various experiments indicated that *C. roseus* has extreme scope as medicinal use and good source of secondary metabolites. *C. roseus* extract has a pool of phytochemicals, which may have different type of medicinal activities. Phytochemical characteristics verified with various tests. The preliminary phytochemical analysis showed the presence of alkaloids, tannin, steroids and glycosides. But saponins, phenol, flavonoids, anthraquinone, lignin and terpenoid were found to be absent. These secondary metabolites localized in leaf and collected in different parts of plant. Powder of plant has characteristic odour, bitter in taste and smooth in touch.

Table 1: Nature and percentage yield of extract of *C. roseus*

S.No.	Name of extract	Nature	Colour	% yield(w/w)
1	Simple methanolic extract	Shade Dried	Light green	30.5

Table 2: Phytochemical constitute of *Catharanthusroseus*

S.No.	Phytochemical name	Reagent test	Observation	Test Result
A	Alkaloids	Mayer's Reagent	Cremish precipitate /Brownish red precipitate	+++
B	Saponins	Crude extract treated with 5 ml of distilled water shake well	No reaction	-
C	Phenol	2 ml crude extract mixed with 2% solution of $FeCl_3$	No reaction	-
D	Flavonoids	2 ml crude extract treated with 2 % solution of NAOH turned colourless on addition of few drop of diluted acid	No reaction	-
E	Tannins	2ml crude extract mixed with 2% solution of $FeCl_3$	Blue-green or black coloration	+
F	Lignins	Extract was treated with 2% furfuraldehyde	No reaction	-
G	Steroids	Crude extract mixed with 2 ml chloroform then 2 ml of conc. H_2SO_4 shaken gently	Reddish brown color	+
H	Glycosides	Crude extract + 2ml of glacial acetic acid +1-2 drop of 2% solution of $FeCl_3$, poured another test tube contain 2 ml of conc. H_2SO_4	Formation of brown ring at the interphase	++
I	Antraquinone	2 ml extract with 2 ml of 25% ammonia solution shake well	No reaction	-
J	Terpenoids	Crude extract + 2ml chloroform + 1 ml con. H_2SO_4	No reaction	-

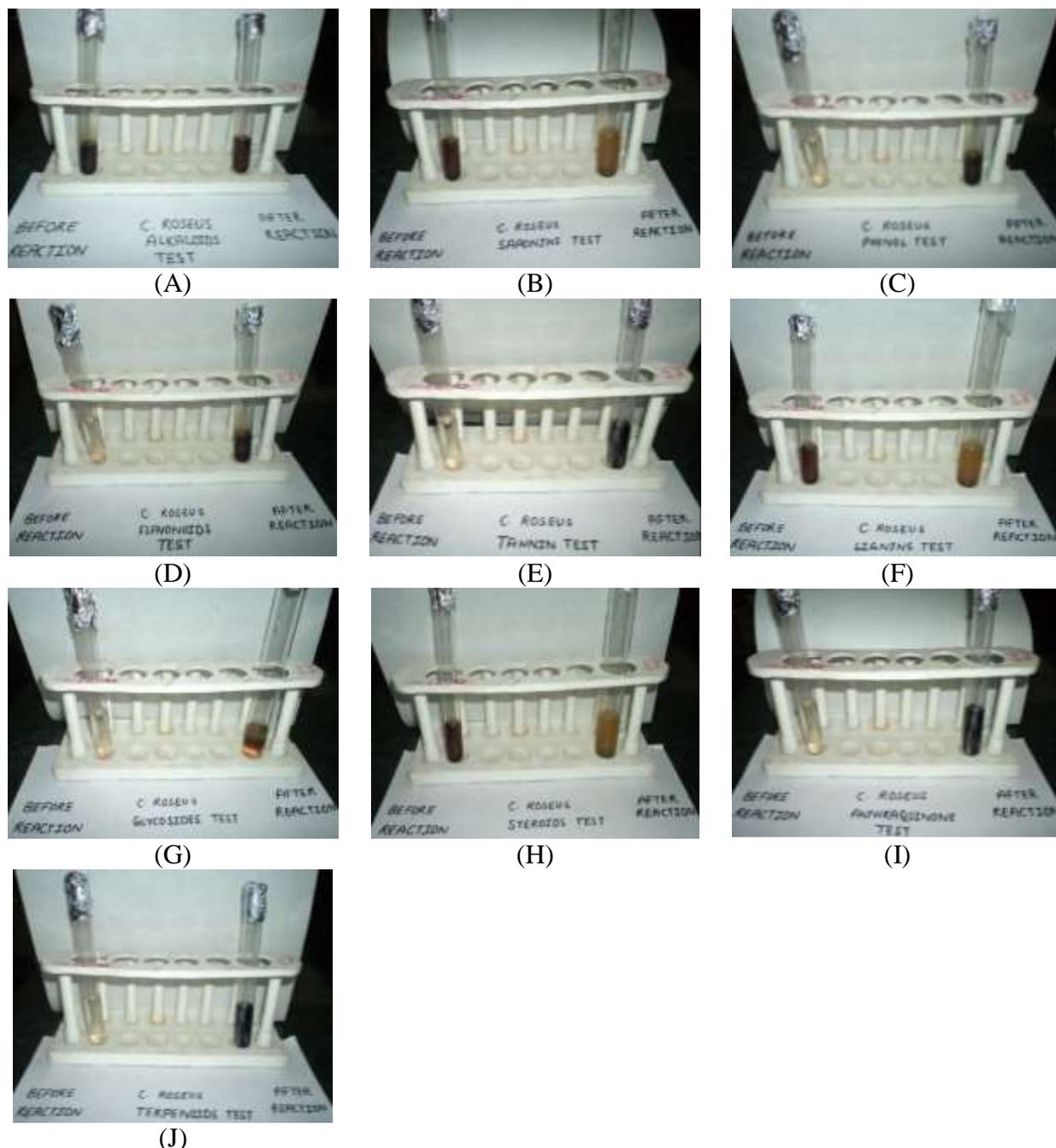


Figure1:Qualitative phytochemical test for estimation of alkaloids (A) and saponin (B), phenol (C) and (D) flavonoids, tannin (E) and lignin (F), steroid (G) and glycosides (H), anthraquinone (I) and terpenoid (J)

Different phytochemicals have been found to possess a wide range of activities, which may help in protection against chronic diseases. For example, alkaloids protect against chronic diseases. Steroids show the analgesic properties. Steroids are also responsible for central nervous system activities. On the basis of presence of these phytochemicals, it is concluded that *C.*

roseus possessed potential secondary metabolites and this plant can also be attempted to derive the drugs of antioxidant properties. However, further *in vivo* studies are still needed to confirm these properties. Author also have various review and research articles on medicinal and Ayurvedic systems of plants, published on *Tribulus terrestris*²³, *Acorus calamus*²⁴, *catharanthus roseus*²⁵, *Oxalis corniculata*²⁶, *Cuscuta reflexa*²⁷, *Solanum nigrum*²⁸, *Murraya koeingii*²⁹, and *Simarouba Glauca*³⁰. These became popular articles for further investigations on particular medicinal herbs. These articles also have been provided very keen interest to students and researchers to make great achievements in medicinal plants research.

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