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Preliminary Phytochemical Tests and Antioxidant Potential of *Catharanthus Roseus* Extracts

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Abstract **- Periwinkle (nayantara) is the common name for a pair of perennial flowering shrubs belonging to the *Apocynaceae* family. The herb has been used for centuries to treat a variety of ailments and was a favourite ingredient of magical charms it was in the middle ages. The latin name for this herb is *Catharanthusroseus*, but it was classified as *Vincarosea,* and is still called by that name in some of the herbal literature. The present study explores the various phytochemicals present in the plant so as to substantiate its antioxidant property and also shows the several phytoconstituents that contribute to its various medicinal properties. Preliminary phytochemical screening of the crude extracts revealed the presence of alkaloids,flavanoids, tannins, saponins and phenolics. During DPPH assay of methanol extract of aerial part of *Catharanthusroseus*shows highest antioxidant activity of 92.04% in 200(µg/ml). The results confirm that aerial part of *Catharanthusroseus*can be used as source of drugs to fight infections caused by susceptible bacteria.**

Keywords: **DPPH Assay, Phytochemicals, Antioxidant, *C.roseus***

I. INTRODUCTION

Most part of the medicinal plants including leaves, roots, stems, flowers, fruits and twigs are used for extract as raw drugs. While some of these raw drugs are collected in smaller quantities by the local communities and folk healers for local uses, many other raw drugs are collected in larger quantities and traded in the market as the raw material for many herbal industries [1]. Plants used for traditional medicine contain a wide range of substances that can be used to treat chronically infectious diseases.

The antimicrobial activities of plant extracts may reside in a variety of different components, including aldehydes and phenolic [2]. The beneficial medicinal effects of plant materials typically result from *the* combinations of secondary products present in the plant. In plants, these compounds are mostly secondary metabolites such as alkaloids, steroids, tannins, phenolics, flavonoids, steroids, resins, and fatty acids, which are capable of producing definite physiological action. The development of drug resistance in human pathogens against commonly used antibiotics has necessitated a search for new antimicrobial substances from other sources including plants. Screening of medicinal plants for antimicrobial activities and phytochemicals is important for finding potential new drugs for therapeutic use.

Catharanthusroseus(Apocynaceae) a perennial plant is commonly seen in tropical countries. It is more commonly known as Madagascar periwinkle. This plant produces beautiful flowers with a variety of colours such as purple, pink and white and commonly planted for decorative purposes [3]. Historically, Madagascar periwinkle had been used for various treatments, e.g., diabetes mellitus, high blood pressure and infection. Leaf part of the plant contains 90 different alkaloids. The most abundant ones are the monomers like catharanthine and vindoline. Two derivativesof vincamine widely used as medicine is known as ethyl-apovincaminate or vinpocetine. It has vasodilating, blood thinning, and memory-enhancing actions, antherosclerotic plaques [4]. Extracts of *Vinca* have significant anticancer activity against numerous cell types.

*Catharanthusroseus*produces more than 100 mono-terpenoidsindole alkaloids (TIA) in different organs [5]. The leaves and stems are the sources of dimeric alkaloids, vinacristine and vinblastine that are indispensable cancer drugs, while roots have antihypertensive, ajmalicine and serpentine [6]. The leaves are used traditionally in various regions of the world including India, West Indies as well as Nigeria to control diabetes [7]. The leaves have been known to contain 150 useful alkaloids among other pharmacologically active compounds. Significant antihyperglycemicandhypotensive activity of the leaf extracts (hydroalcoholic or dichloromethanes) has been reported to reduce blood glucose in normal and alloxan diabetic rabbits [9]. This study elaborates the phytochemical constituents of the plant so as to ascertain the chemical constituents of the plant that attribute to its free radical scavenging property and various medicinal uses.

II. EXPERIMENTAL PROCEDURE

A. Plant Materials and Chemicals

The aerial part of *C.roseus*was collected from sandy beaches of Kanyakumari, Tamilnadu, India, in the month of March 2012 and authenticated by the Departmentof Botany, Dr.Jeeva, Scott Christian College (Autonomous),Nagercoil, Kanyakumari, Tamil Nadu.

All chemicals and solvents were of analytical grade (RANKEM). The aerial part of plant was washed and air dried over a period of one month. The dried samples were milled into fine powder by poundingmanually with a clean sterile mortar, stored in sterile cellophane bags in a cool dry place till further use.

B. Extraction

100grams of dried aerial part of *C.roseus* was extracted in soxhlet sequentially in 1000ml of hexane, chloroform, ethylacetate, and methanol and aqueous. The process was run for 24hr after which the sample was concentrated using reduced pressure distillation under vaccum pump and freeze dried to powdered form. The dried extracts were weighed and kept in labeled sterile specimen bottles.

C. Antioxidant Activity Assays

DPPH assay: (2, 2-diphenyl-1-picrylhydrazyl)
The Radical Scavenging Activity of different extracts was determined by using DPPH assay according to Chang *et al* (2008) [10]with small modification. The decrease of the absorption at 517nm of the DPPH solution after the addition of the antioxidant was measured in a cuvette containing 2.960 µl of 0.1mmethanolic DPPH solution mixed with 20 to 200µg/ml of plant extract and vortexed thoroughly. The setup was left at dark in room temperature and the absorption was monitored after 20 minutes. Ascorbic acid was used as references. The ability of the plant extract to scavenge DPPH radical was calculated by the following equation:
% of DPPH Radical Scavenging Activity (% RSA) =

$$\frac{\text{Abs. control} - \text{Abs. sample}*100}{\text{Abs.Control}}$$

Abs. control is the absorbance of DPPH radical + ethanol; Abs. sample is the absorbance of DPPH radical + plant extract. Measurements were performed in triplicates. Absorbance values were corrected for radicals decay using blank solutions.

D. Preliminary Phytochemical Investigations

The major secondary metabolites classes such as tannins, saponins, terpenoids, flavonoids, alkaloids and glycosides were screened according to the common phytochemical methods described by Harborne1998. [11]

III. RESULTS AND DISCUSSION

A. Antioxidant Activity of C.roseus

The results of the antioxidant activity of *C.roseus* determined by DPPH assay at different concentrations are given in Table I. The results of the antioxidant activity of best screened methanolic extract of aerial parts of *C.roseus* determined by DPPH assays at different concentrations are given in Table. It was evident that part of *C.roseus* showed moderate antioxidant activity when compared with standard antioxidant L-ascorbic acid whose antioxidant activity at different concentrations like 100 to 200µg were 80%, 82%, 85%, 90% and 92% .In the present investigation, the obtained data shows that methanol extract is free radical scavengers and may act as primary antioxidants which can react with free radicals by donating hydrogen.

TABLE I RADICAL SCAVENGING ACTIVITY (RSA) OF THE SELECTED PLANT EXTRACT

Concentration (µg/ml)	RSA%	
	Aerial	Standard
20	68.56	18.54
40	71.86	35.28
60	73.19	47.76
80	76.24	50.19
100	79.35	55.04
120	83.19	59.06
140	86.74	66.37
160	91.09	72.80
180	91.85	74.46
200	92.23	84.51

B. Preliminary Phytochemical Analysis C.roseus

The preliminary phytochemical screening tests for the crude methanol extract of *C.roseus* revealed the presence of tannins, saponins, flavonoids, alkaloids, glycosides and phenolics (Table II).

TABLE II PHYTOCHEMICAL ANALYSIS OF THE BEST SCREENED EXTRACTS OF AERIAL OF *C.ROSEUS*

Phytochemical	Results
	Aerial
Alkaloids	+
Flavonoids	+
Phenols	+++
Tannins	+
Glycosides	+
Reducing sugars	+
Proteins	-
Saponins	+

\+ Present in minor amounts
++ present in moderate amounts
++ present in higher amounts
\- Not detected

The presence of tannins, saponins, flavonoids, alkaloids, glycosides and phenolics has potentially significant application against human pathogens, including those that cause enteric infections [12]. The presence of alkaloids is interesting as significant quantities are used as antimalarials,

analagesics and stimulants [13]. The presence of glycosides moieties like saponins, glycosides and flavonoids which are known to inhibit tumor growth and also serve to protect against gastro-intestinal infections are one of the pharmacognostic importance and give evidence to the use of the plant in ethnomedicine [14]. Tannins are widely used in traditional medicine in treating wounds and arrest bleeding. Some of these bioactive compounds which are synthesised as secondary metabolites as the plant grows also serve to protect the plant against microbial attacks and predation by animals. Any of these secondary metabolites, singly or in combination with others could be responsible for the anti-oxidant activity of the plant.

IV. CONCLUSION

These findings enrich our knowledge of the chemical constituents that are responsible for the medicinal uses of the plant and the antioxidant potential of *C.roseus*. Hence the present study supports the view that these medicinal plants might be useful as antioxidant agents. The plant showed significant activity, so further the compound isolation, purification and characterization which is responsible for inhibiting activity, has to be done for the usage of antioxidant agent.

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