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Phytochemical analysis and Antioxidant activity of *Catharanthus roseus* Flower extract in different solvent

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Abstract

Antioxidant effectiveness of indigenous medicinal plants *Catharanthus roseus* flower extract with solvent of different polarity (Ethanol and Acetone) was assessed for DPPH radical scavenging activity. The *Catharanthus roseus* contained appreciable levels of antioxidant activity of flower of *Catharanthus roseus* at various concentrations. In this present study an attempt was made to investigate the phytochemical analysis in different solvent extract of *Catharanthus roseus*. The qualitative analysis of phytochemical screening reveals the presence of some bioactive compounds.

Key words: *Catharanthus roseus*, Antioxidant activity, phytochemical analysis, DPPH assay

1. Introduction:

Research in the chemistry of natural products has endless potential and is especially important in countries like India which has a rich biodiversity (Jayakumar, 2010). Antioxidants are compounds which act as radical scavengers when added to the food products and prevent the radical chain reaction of oxidation, delay or inhibit the oxidation process and increase shelf life by retarding the process of lipid peroxidation (Young, 2001). The ability of phenolic substances including flavonoids and phenolic acids acting as antioxidants has been reported (Liu, 2003). Tannins have been reported to have strong antioxidant activity (Cai, 2006). There is also growing interest both in industry and in scientific research in spices and medicinal herbs because of their antimicrobial and antioxidant activity (Eyob, 2008).

Medicinal plants contain some organic compounds which provide definite physiological action on the human body as well as their physiological activities due to the presence of bioactive substance include tannins, alkaloids, carbohydrates, terpenoids, steroids and flavonoids (Paikara, 2015). It is cultivated mainly for its alkaloids, which are having anticancer activities (Jaleel, 2009).

Moreover, several species of Apocyanaceae family plants has been widely used as main ingredient in traditional medicine flower extract of *Catharanthus roseus* could be of considered infers to the development of new life saving drugs (Komathi, 2014). The leaves and flowers of this plant are effective for diabetic patients. *Catharanthus roseus* are cultivated two common names, which is named on the basis of their flower colours, Pink: Rosea, White: Alba (Sain, 2007).

Many natural antioxidants have already been isolated from various natural resources, such as oilseeds, cereal crops, vegetables, spices, and herbs (Ramarathnam, 1995). Sea grasses specifically produced bioactive compounds that reportedly have anti-bacterial (Harrison, 1980; Devi, 1999; Bhosale, 2002; Bernard, 1989 and Ragupathi Raja Kannan, 2010a) anti-algal (Harrison, 1982) antifungal (Jensen, 1998 and Ballesteros, 1982) anti-viral (Premanathan, 1992 and Rowley, 2002) anti-protozoal (Orhan, 2006) anti-inflammatory (Hua, 2006) and antidiabetic (Gokce, 2008) activities. More recently, reports have revealed that sea grasses are rich sources of antioxidant compounds (Hasina, 2003; Gokce, 2008; Kolenchenko, 2005; Ragupathi Raja Kannan, 2010b; Ragupathi Raja Kannan, 2010c and Sureda, 2008).

The discovery of vitamins as antioxidants in the nutrition has led to their supplementation to improve health and extend life span. These supplements may include specific antioxidant chemicals, like resveratrol (from grape seeds), combinations of antioxidants, like the 'ACES' products that contain Selenium, carotene (provitamin A), Vitamins C and E or special herbs that are known to contain such antioxidants. Vitamin C is found in all the citrus fruits and Vitamin E from green vegetables, etc. Vitamin A is synthesized in the body by carotene being its precursor (Manas, 2014). DPPH radical-scavenging activity and nitric oxide radical inhibition method.

Antioxidants are radical scavengers which give protection to human body against free radicals by inhibiting the oxidizing chain reactions. When these substances are present at low concentration in body they markedly delay or prevent the oxidation of an oxidizable substrate (Velioglu, 1996). They are also generated through environmental pollutants, cigarette smoke, automobile exhaust, radiation, air pollution, pesticides etc **(**Van Bergen, 1996). Normally there exists a balance between the amount of free radicals generated in the body and the antioxidant defense systems that scavenge/quench these free radicals preventing them from causing deleterious effects in the body (Don, 1999).

In the recent years interest in the study of antioxidant activity of plant extracts and isolation from plants has grown due to the fact that the free radicals have been related to degenerative diseases (Willcox, 2004). *Vinca rosea* has a variety of medicinal properties such as antibacterial (Carew, 1970) antifungal (Jaleel, 2007) antiviral (Fransworth, 1968) anticancer (Ram, *2001).* *Calotropis gigantea* has been reported to possess a number of medicinal properties and is used in toothache, earache, sprain, anxiety, pain, epilepsy, mental disorder and also it possesses antidiarrheal, analgesic and CNS activity (Pathak, 2007). An extensive literature survey indicates antioxidant and antimicrobial activity in *Calotropis gigantea* and *Vinca rosea*. But only scanty information is available on such potential regarding the individual plant parts concerned (root, stem, leaf, flower and seed).

In the present study, the experiment was carried out in the flower of *Catharanthus roseus* so as to compare the phytochemical and antioxidant activities of the two coloured flower of *Catharanthus roseus* was selected Indian medicinal plants and the values were compared with that of the previous reports. The concentration dependency of the antioxidant was also investigated (Jayakumar, 2010).

2. Materials and Methods:

2.1 Collection of plant materials:

The *Catharanthus roseus* flower sample were collected from the area Anna agar in Thoothukudi district, India and taken to the laboratory. Flower was washed separately using running tap water, followed by rinsing using sterilized distilled water. Excess of water was removed from the plant materials using filter paper before they were used for extraction (Pankaj Goyal, 2008).

2.2 Extract preparation:

Flower samples of *Catharanthus roseus* was thoroughly washed and dried in hot air oven at 100^0 C for 1 hour. Then its weight was noted before drying and after drying. The dried sample was then crushed in pestle and motor into fine powder (Sonia Chaman, 2013).

2.3 Solvent Extraction:

The Acetone and ethanol extract of flower were prepared by following the methodology of (Alam, 2010). Dried powder of plants was taken and solvent was added to it in the ratio of 1:4. The powder of the solvent is evaporation of solvent at room temperature. Extracts were stored at 4°C until further use.

2.4 Qualitative Analysis of Phytochemical:

Catharanthus roseus with acetone and ethanol were subjected to various qualitative tests for the identification of plant constituents present in this species (Nandkarni, 2004 and Khare, 2007).

2.5 Antioxidant Activity

2.5.1 Chemical Reagents:

2, 2-Diphenyl-1-picrylhydrazyl (DPPH) and ascorbic acid was purchased from Sigma-Aldrich. Phosphate buffer (pH- 7.4) and Methanol. **A**ll the chemicals and solvents used were of analytical grade.

2.5.2 DPPH Free Radical Scavenging Activity:

DPPH solution (0.004 %), sample extracts and standard (vitamin C) was prepared in methanol. Sample extract and standard (vitamin C) solution

were prepared in different concentrations 20, 40, 60, 80 and 100 µg/ml. 0.5ml of different concentrations of standard solution or sample extracts was taken in different test tubes and then 0.5 ml of DPPH (0.004 %) solution was added and kept in dark for 30 min. and absorbance was recorded at 517 nm. The decrease in absorbance of the DPPH radical caused by antioxidant was due to the scavenging of the radical by hydrogen donation. It was visually noticeable as a colour change from purple to yellow (Majo, 2008). The percentage inhibition activity was calculated using the formulae below (Blois, 1958).

% DPPH free radical scavenging = (Absorbance of control – Absorbance of test) X 100
Absorbance of control

3. Result And Discussion:

3.1 Qualitative Analysis Of Phytochemical:

The qualitative chemical test for the extracts was performed. The investigation showed that *Catharanthus roseus* contains tannins, alkaloids, flavonoids, terpenoids, and saponins were present in the *Catharanthus roseus* as showed in Table:1. These compounds are described as potent biologically active compounds found in medicinal plant parts which are precursors for clinically useful drugs (Steiling, 1999). The potency of medicinal plants is attributed to the action of the phytochemical constituents. These are actually produced by plants as secondary metabolites in response to environmental pressure or as a defense mechanism to animal or plant diseases. The present study showed that the presence of biologically active secondary metabolites in *Catharanthus roseus*. Plants have long been screened if they contained active compounds with therapeutic activity (Patharajan, 2014).

Table: 1 PHYTOCHEMICAL CONSTITUENTS OF *CATHARANTHUS ROSEUS* STUDIED

Plant	Alkaloids	Tannins	Saponins	Cardiac glycosides	Terpenoids	Phenol	Flavonoids	Carbohydrates
Purple *Catharanthus roseus* flower extracted in acetone	+	+	+	-	+	-	+	-
Purple *Catharanthus roseus* flower extracted in ethanol	+	+	+	-	+	-	+	-
White *Catharanthus roseus* flower extracted in acetone	+	+	+	-	+	-	+	-
White *Catharanthus roseus* flower extracted in ethanol	+	+	+	-	+	-	+	-

3.2 Antioxidant activity of C*atharanthus roseus* flower extracts:

Antioxidants inhibit oxidation of food also quench dreaded free radicals produced due to environmental and physiological stress which leads to aging, atherosclerosis and cancer (Freeman, 1982). The present study indicates that antioxidant activity of *Catharanthus roseus* was determined by DPPH assays at different concentrations (20, 40, 60, 80 and 100 µg). Purple *Catharanthus roseus* flower extracted in acetone 100 µg/ml showed maximum antioxidant activity of 53.71%. Purple *Catharanthus roseus* flower extracted in ethanol 100 µg/ml showed maximum antioxidant activity of 79.95 %. White *Catharanthus roseus* flower extracted in acetone 100 µg/ml showed maximum antioxidant activity of 74.36 %. White *Catharanthus roseus* flower extracted in ethanol 100 µg/ml showed the maximum antioxidant activity of 39.42% as showed in Table: 2

There are many evidences that natural products and their derivatives have efficient anti-oxidative characteristics, consequently linked to anti-cancer, hypolipidemic, anti aging and anti-inflammatory activities (Kapoor, 1969).
Results obtained in this study confirmed that the antioxidant activity of *Catharanthus roseus*. We investigated the free radical scavenging activity of extracts in *Catharanthus roseus* flower (Nasir Rasool, 2011). The role of free radicals reactions in biology has become an area of intense interest. It is generally accepted that free radicals play an important role in the development of tissue damage and pathological events in the living organism (Okwu, 2004). To best of our knowledge no earlier reports are available regarding the DPPH radical

scavenging activity of *Catharanthus roseus* flower of which to compare with the present values. The free radical (DPPH) scavenging activity of *Catharanthus roseus* fractions was found to be comparable to the well known antioxidants such as vitamin C.

Table: 2 Antioxidant effect of *Catharanthus roseus* flower extract on DPPH

Treatment	Dose (µg/ml)	Absorbance @517 nm	% activity against DPPH radicals
Control	----	DPPH control= 0.987	----
Vit C	100	0.168	82.97
Purple *Catharanthus roseus* flower extracted in acetone	100	0.444	53.71
Purple *Catharanthus roseus* flower extracted in ethanol	100	0.185	79.95
White *Catharanthus roseus* flower extracted in acetone	100	0.753	74.36
White *Catharanthus roseus* flower extracted in ethanol	100	0.585	39.42

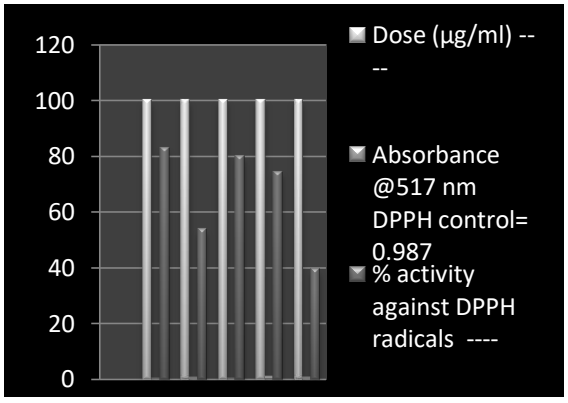

Figure:1 Antioxidant effect of *Catharanthus roseus* flower extract on DPPH.

4. Conclusion:

In this present study *in vitro* antioxidant activity of acetone and ethanol extracts of *Catharanthus roseus* was evaluated. It supports the view that this medicinal plant might be useful as an antioxidant. Therefore extracts from this plant could be seen as a good source for useful drug. The traditional medicine practice is recommended strongly for this plant as well as it is suggested the further work should be carried out to isolate, purify and characterized the active constituents responsible for the activity.

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