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In-vitro Evaluation of *Scindapsus Officinalis* (ROXB.) Schott. Fruit for Antioxidant Potential

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Abstract: In the present study, coarse powder of *Scindapsus officinalis* (Roxb.) Schott. fruit was extracted successively using hexane, chloroform, ethyl acetate and 50% ethanol. The ethyl acetate and 50% ethanolic extracts were investigated for its antioxidant activity by using nitric oxide and DPPH radical scavenging methods. The IC_{50} value was also calculated. Ascorbic acid was used as standard. Both the 50% ethanolic and ethyl acetate extract were found to exert concentration dependent free radical scavenging activity but former extract was more effective than the later on. The highest free radical scavenging activity by *Scindapsus officinalis* fruit extracts was observed at concentration of 1000 µg/ml.

Key words: *Scindapsus officinalis* (Roxb.) Schott · Antioxidant · Free radicals · IC_{50} Value

INTRODUCTION

Oxygen is necessary for the survival of all on this earth. About 5% of oxygen gets univalently reduced to oxygen derived free radicals like hydrogen peroxide, hydroxyl, nitric oxide and superoxide radicals during the process of oxygen utilization in normal physiological and metabolic processes. All these radicals are known as reactive oxygen species (ROS) and exert oxidative stress towards the cells of human body rendering each cell to face about 10,000 oxidative hits per second [1-3]. These free radicals can also damage the cell membranes and intracellular structures such as mitochondria, nucleus and DNA causing mutations. Damaged cell membranes also allow viruses and bacteria to enter the body more freely and asthmatic patients are subjected to continuous oxidative stress associated with viral and bacterial infections [4]. When production of ROS overtakes the antioxidant defense of cells, the free radicals start attacking the cell proteins, lipids and carbohydrates [5-7]. This leads to a number of physiological disorders such as atherosclerosis, hypertension, cancer, diabetes and premature aging [8]. Antioxidant therapy has gained an immense importance in the treatment of these disorders/diseases. Antioxidants have been reported to prevent oxidative damage caused by free radical and may prevent the occurrence of disease, cancer, diabetes, aging as well. It can interfere with the oxidation process by reacting with free radicals, chelating and catalytic metals and also by acting as oxygen scavengers [9, 10].

Presently, available synthetic antioxidants like butylated hydroxy anisole (BHA), butylated hydroxy toluene (BHT), tertiary butylated hydroquinone and gallic acid esters have been supposed to cause or prompt negative health effects. Furthermore, these synthetic antioxidants have also exhibited the low solubility and moderate antioxidant activity. Hence, application of these antioxidants has been strongly restricted. In spite of these, use of naturally occurring antioxidants is getting popularity [11, 12]. The medicinal properties of plants have been investigated in the recent scientific developments throughout the world, due to their potent antioxidant potential, no side effect and economic viability [13]. Many plants often contain substantial amounts of antioxidants including vitamin C and E, carotenoids, flavonoids, phenolics, tannins etc. which have been reported to exert multiple biological effects, including antioxidant, free radical scavenging abilities, anti-inflammatory, anti-carcinogenic and many other activities [14, 15].

About 70% of the people in India still rely on medicinal plants and herbal drugs. In recent times, there has been an increase of interest in the therapeutic potentials of medicinal plants as antioxidants. A large number of medicinal plants and their purified constituents have been reported to exhibit antioxidant activity. In this context, the present study was aimed to evaluate the *Scindapsus officinalis* (Roxb.) Schott. Fruit for *in-vitro* antioxidant property.

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MATERIAL AND METHODS

Plant Material: The fruits of *Scindapsus officinalis* (Roxb.) Schott. for the proposed study were collected from the market [K. Ramaswamy Chetty (KRC) Country Drugs dealer, whole sale and retail, Shop No. 117, Rasappa Chetty Street, Park Town, Chennai, India-600003]. It was identified the help of available literature and authenticated by Dr. P. Jayaraman, [Director, Plant Anatomy Research Center, (PARC) Tambaram, Chennai]. The voucher specimen of the plant and fruit has deposited in departmental herbarium for further reference.

Extraction: The method mentioned by Harbone (1973) [16], Overton KH (1963) [17], Kokate (1994) [18] was followed for extraction of crude drug. The fruit of *Scindapsus officinalis* plant was shade dried and powdered. About 300 gm powdered drug was extracted successively with hexane, chloroform, ethyl acetate and 50% ethanol by cold maceration method. The powdered material was dried each time before extraction with next solvent. After complete extraction, the extracts were concentrated by distilling off the solvent and then evaporated to dryness on water-bath. The ethyl acetate extract of *S. officinalis* fruit (EAESOF) and 50% ethanolic extract *S. officinalis* fruit (EESOF) were subjected to *in-vitro* antioxidant studies.

***In-vitro* Antioxidant Activity Assays**

Nitric Oxide Radical Scavenging Assay: Nitric oxide scavenging assay was measured by spectrophotometirc method described by Govindarajan *et al.* (2003) [19]. Sodium nitroprusside (5mM) in phosphate buffer saline was mixed with different concentrations (50-1000 µg/ml) of *Scindapsus officinalis* fruit extracts dissolved in methanol and incubated at 25°C for 30 min. After 30 min 1.5 ml of the incubation solution were removed and diluted with 1.5 ml of Griess reagent (1% sulphanilic acid, 2% Phosphoric acid and 0.1% N-1-napthylethylene diamine dihydrochloride). The absorbance was measured at 546 nm. The ascorbic acid was taken as Standard. All determination was performed in triplicate. The percentage of Nitric oxide radical scavenging ability of the sample was calculated by using following formula;

$$\% \text{ Inhibition} = [(\text{Abs}_{\text{Control}} - \text{Abs}_{\text{Sample}})/ \text{Abs}_{\text{Control}}] \times 100$$

Where $\text{Abs}_{\text{Control}}$ is absorbance of control and $\text{Abs}_{\text{Sample}}$ is absorbance of test sample. The IC_{50} Value for extracts was also calculated.

DPPH Radical Scavenging Assay: The effect of various extracts of *Scindapsus officinalis* fruit on DPPH (2, 2-diphenyl-1-picryylhydrazyl) radical was estimated by Rajkumar and Rao. (1993) [20]. In brief, 3mL of 100 µM of DPPH prepared in AR grade methanol was added to various concentrations of *Scindapsus officinalis* fruit extracts (50-1000 µg/ml) and made up the final volume 4 mL with AR grade methanol. The solutions kept for 20 min at room temperatue and then the absorbance of resulting solutions and the blank (With same chemicals except sample) were recorded against the ascorbic acid as standard. The decrease in the absorbance was continuously recorded in a spectrophotometer 515 nm. All determination was performed in triplicate. The percentage of DPPH radical scavenging ability of the sample was calculated by using following formula;

$$\% \text{ Inhibition} = [(\text{Abs}_{\text{Control}} - \text{Abs}_{\text{Sample}})/ \text{Abs}_{\text{Control}}] \times 100$$

Where $\text{Abs}_{\text{Control}}$ is absorbance of control and $\text{Abs}_{\text{Sample}}$ is absorbance of test sample. The IC_{50} Value for extracts was also calculated.

Statical Analysis: The results were expressed as mean ± SD of three independent values.

RESULT AND DISCUSSION

The free radical scavenging activity was evaluated by nitric oxide and DPPH radical scavenging *in-vitro* assays. Nitric oxide (NO) is a potent pleiotropic mediator of physiological processes such as inhibition of platelet aggregation, neuronal signaling, smooth muscle relaxation and regulation of cell mediated toxicity. It is a diffusible free radical which plays many roles as an effector molecule in diverse biological systems including neuronal messenger, vasodilatation antimicrobial and antitumor activities. The scavenging of nitric oxide by was concentration dependent (Table 1). Figure 1 shows the graphical representation of inhibition of nitric oxide generation by *S. officinalis* fruit extracts. The highest nitric oxide radical scavenging of 50% EESOF and EAESOF was found 88.177±0.716 and 77.832±0.424 percent respectively at concentration of 1000 µg/ml. Ascorbic acid was used as standard compound.

In the DPPH radical scavenging assay DPPH radical was used as a substrate to evaluate free radical scavenging activity of *Scindapsus officinalis* fruit extracts. It involves reaction of specific antioxidant with a stable free radical 2, 2-diphenyl-1-picryylhydrazyl

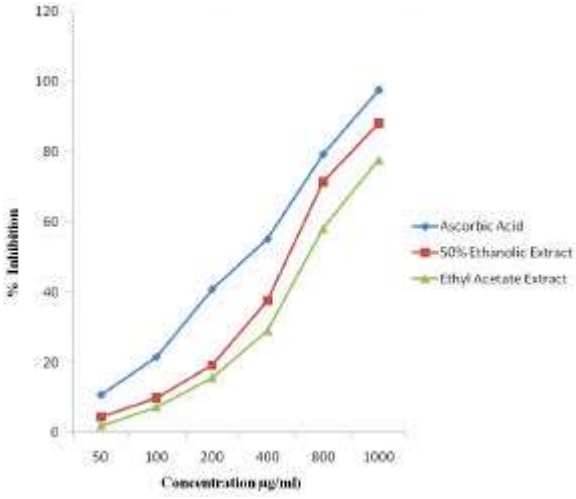

Fig. 1: Nitric oxide Radical Scavenging Activity of *Scindapsus officinalis* Fruit
Results are mean±SD of three values

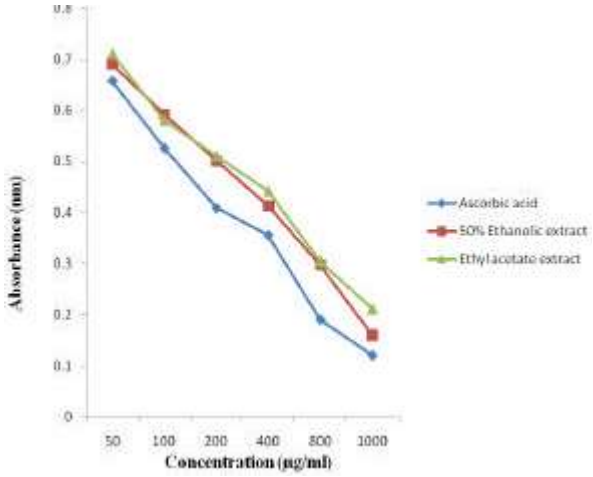

Fig. 2: Effect of *S. Officinalis* fruit on the concentration of DPPH radical
Results are mean±SD of three values

DPPH*. As a result, there is a reduction of DPPH concentration by antioxidant, which decreases the optical absorbance of DPPH; this is detected by spectrophotometer at 517 nm. Figure 2 shows a significant decrease in the concentration of DPPH radical due to scavenging ability of *S. officinalis* fruit extracts. The highest DPPH radical scavenging of 50% ethanolic and ethyl acetate extracts of *S. officinalis* fruit was found 80.199± 0.722 and 73.599±0.606 percent respectively at concentration of 1000 µg/ml (Table 2). It is graphically represented in figure 3. Ascorbic acid is used as standard.

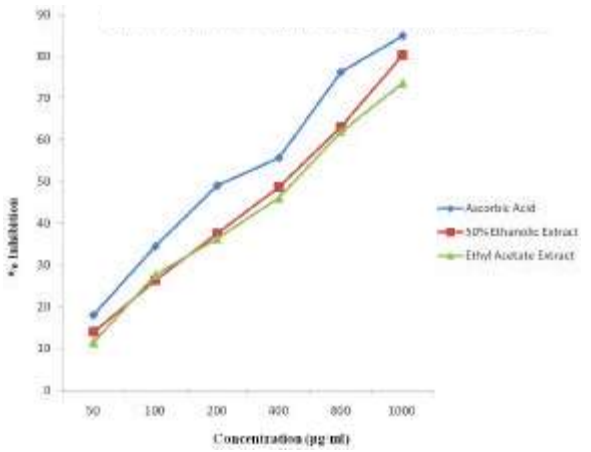

Fig. 3: DPPH Radical Scavenging Activity of *Scindapsus officinalis* Fruit
Results are mean±SD of three values

Table 1: Nitric oxide radical scavenging activity of *Scindapsus officinalis* fruit extracts

	% Inhibition		
Concentration	Ascorbic acid	50% Ethanolic Extract	Ethyl Acetate Extract
50 µg/ml	10.837±0.627	4.443±0.443	1.97±0.085
100 µg/ml	21.675±0.791	9.852±0.495	7.389±0.525
200 µg/ml	40.887±0.858	19.212±0.379	15.763±0.634
400 µg/ml	55.172±0.548	37.574±0.385	29.064±0.518
800 µg/ml	79.31±0.411	71.428±0.555	58.128±0.201
1000 µg/ml	97.537±0.547	88.177±0.716	77.832±0.424

The results were expressed as mean±SD of three values.

Table 2: IC_{50} Values of *Scindapsus officinalis* fruit extracts for nitric oxide radical scavenging activity

	Concentration (µg/ml)		
Parameter	Ascorbic acid	50% Ethanolic Extract	Ethyl Acetate Extract
IC_{50} Value	330 µg/ml	545 µg/ml	690 µg/ml

Table 3: DPPH radical scavenging activity of *Scindapsus officinalis* fruit extracts

Concentration	% Inhibition		
(µg/ml)	Ascorbic acid	50% Ethanolic Extract	Ethyl Acetate Extract
50	18.057±0.627	13.948±0.463	11.457±0.676
100	34.496±0.791	26.276±0.436	27.646±0.404
200	49.066±0.858	37.609±0.616	36.364±0.559
400	55.666±0.548	63.014±0.634	46.077±0.218
800	76.214±0.411	48.568±0.941	2.017±0.423
1000	84.431±0.548	80.199±0.722	73.599±0.606

The results were expressed as mean±SD of three values.

Table 4: IC_{50} Values of *Scindapsus officinalis* fruit extracts for DPPH radical scavenging activity

	Concentration (µg/ml)		
Parameter	Ascorbic acid	50% Ethanolic Extract	Ethyl Acetate Extract
IC_{50} Value	265 µg/ml	297 µg/ml	505 µg/ml

IC_{50} values of 50% ethanolic and ethyl acetate extracts of *S. officinalis* fruit were found to be 545µg/ml and 690µg/ml respectively for nitric oxide radical scavenging activity (Table 2) and 297µg/ml and 505µg/ml respectively for DPPH radical scavenging activity (Table 4).

CONCLUSION

On the basis of results of this study, it was concluded that both the 50% ethanolic and ethyl acetate extracts of *Scindapsus officinalis* fruit were found to be significant but former was more effective than the later. This antioxidant property may be due to the presence of flavonoids and phenolics compounds, which needs further analysis.

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