



ANTIOXIDANT POTENTIAL OF METHANOLIC EXTRACT OF *MONSTERA ADANSONII*

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Abstract: Antioxidants prevent oxidative damage caused by free radical and can be used in cardiovascular and anti-inflammatory diseases. The presence of natural antioxidant in plants is well known. The present study was undertaken to find the antioxidant value of the plant *Monstera adansonii*. The amount of total phenols, flavonoids, tannins and radical scavenging activity was studied. The analysis carried out was, DPPH free radical scavenging activity, Ferric reducing antioxidant power assay, Superoxide anion scavenging activity and Nitric oxide scavenging activity. Plant extracts were prepared by maceration method using methanol. Results show that *Monstera adansonii* had highest yield of methanolic extract (3.84%), total phenolic content (10.34mg/g) and antioxidant activity (89.84%) using DPPH method. Increasing the concentration of the extracts resulted in increased ferric reducing antioxidant power for the methanolic extract tested. The plants, *Monstera adansonii*, may be potent source of natural antioxidants.

Key Words: Phenol, *Monstera adansonii*, DPPH, Tannin, Antioxidant

INTRODUCTION

Free radicals (superoxide, hydroxyl radicals and nitric oxide) and other reactive species (hydrogen peroxide, hypochloric acid, and proxynitrite) produced during aerobic metabolism can cause oxidative damage of amino acids, lipids, proteins and DNA. It has been established that oxidative stress is among the major causative factors in the induction of many chronic and degenerative diseases including atherosclerosis, ischemic heart disease, ageing, diabetes, cancer, neurodegenerative disease and others. The most effective way to eliminate free radicals which cause the oxidative stress is with the help of antioxidants. In recent years much attention has been devoted to natural antioxidant and their association with health benefits (Arnous *et al.*, 2001) and probe to be plants are potential sources of natural antioxidants. Natural antioxidants especially phenols and flavonoids are safe and also bioactive. Recently focus has been concentrated on identification of plants with antioxidant ability that may be used for human consumption (Madhavi and Salunkhe, 1995; Kaur *et al.*, 2006; Pourmorad *et al.*, 2006; Hosseinimehr *et al.*, 2007).

Monstera adansonii schott., Adanson's monster, is an evergreen root climbing hemi-epiphyte. The stem, branched, cylindrical, aerial elongated axis with extended internodes, and climbing hemiepiphytes. Leaves simple, large, thick, distichous, juvenile, striking of hetero blast. Petiole geniculate apically, sheath usually long, persistent, blade entire, oblique, oblong to ovate-elliptic, often conspicuously and elaborately perforated, primary lateral veins pinnate, running into marginal vein, Secondary laterals often parallel-pinnate.

As part of our continuing research project on *Monstera adansonii*, an in vitro antioxidant activity of methanolic extract with, DPPH free radical scavenging

activity, Ferric reducing antioxidant power assay, Superoxide anion scavenging activity and Nitric oxide scavenging activity was carried out in the present study.

MATERIALS AND METHODS

Sample Preparation

Fresh samples of the *Monstera adansonii* leaves and stem were collected in Tamil Nadu Agriculture Society, Chennai-04, shade dried, powdered and extracted with 75 ml of the solvent methanol and kept overnight in shaker. The extract was collected after filtration using Whatman No.1 filter paper and was stored. Extraction was repeated three times. The extracts were pooled together and was concentrated using rotary vacuum evaporator (40°) and the residue obtained were designed as crude extracts were labeled and stored at 4°C for further study

DPPH free radical scavenging activity (Harbone *et al.*, 1995): Different concentrations (50, 100 and 150µl) of each extracts were added, at an equal volume, to methanolic solution of DPPH (0.1%). The reaction mixture is incubated for 30min at room temperature; the absorbance was recorded at 517 nm. BHT was used as standard controls. The annihilation activity of free radicals was calculated in % inhibition according to the following formula (% of Inhibition = (A of control - A of Test) / A of control * 100).

Ferric reducing antioxidant power assay (Benzie and Strain 1996): The antioxidant capacity of each sample was estimated according to the procedure described by Benzie and Strain (1996). Briefly, methanol extracts solution (50, 100 and 150µl) was added to 1.5 ml of FRAP reagent. Ferrous sulphate was used as standard control.

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Superoxide anion scavenging activity

(Nishikimi et al., 1972): The reaction mixture was prepared and samples at different concentration (50, 100 and 150 μ l) and standard solution (100 μ l) obtained from stock solutions were added and finally the reaction was accelerated by adding 100 μ l phenazinemethosulfate (PMS) solution. The reaction was incubated at 25 $^{\circ}$ C for 5 minutes and absorbance was measured at 560nm against the corresponding blank solutions. Quercetin was used as the standard control. The annihilation activity of free radicals was calculated in % inhibition according to the following formula:

$$\text{Inhibition \%} = \frac{(\text{Absorbance of control} - \text{Absorbance of sample}) * 100}{\text{Absorbance of control}}$$

Nitric oxide scavenging activity (Ebrahim zadeh

MA, et al., 2009): The reaction mixture contains 2ml of sodium nitroprusside (10mM), 0.1ml of phosphate buffer saline and samples at different concentration (50, 100 and 150 μ l) obtained from stock solution, were incubated at 25 $^{\circ}$ C for 150 min. After incubation, 0.5ml of the reaction mixture was mixed with 1ml of sulphanilamide (1%) and allowed to stand for 5 min for completing diazotization. Then, 1ml of naphthyl ethylene diaminedihydrochloride (0.1% W/V) was added, mixed and allowed to stand for 30min at 25 $^{\circ}$ C. A pink colored chromophore was formed in diffused light. The absorbance of these solutions was measured at 540nm against the corresponding blank solutions. Ascorbic acid was used as the standard control. The annihilation activity of free radicals was calculated in % inhibition according to the following formula:

$$\text{Inhibition \%} = \frac{(\text{Absorbance of control} - \text{Absorbance of sample}) * 100}{\text{Absorbance of control}}$$

RESULTS AND DISCUSSION

DPPH is used as a free radical to evaluate the antioxidant activity of some natural compounds. There was a decrease in concentration of DPPH (Table 1 & Fig 1) with time when the concentration of extract and BHT increased. Extracts of methanol shows higher (89.84%) scavenging activities and showed lower activity than BHT at the same concentration. DPPH free radical scavenging assay can be helpful for primary screening and finding of novel antioxidants. The same experiment was carried out on ascorbic acids, butylated hydroxylansole (BHA) and α -Tocopherol which are known antioxidant agents. All test and analysis were run in triplicates and the results obtained were averaged (Koleva et al., 2002 and Oloyede, 2010).

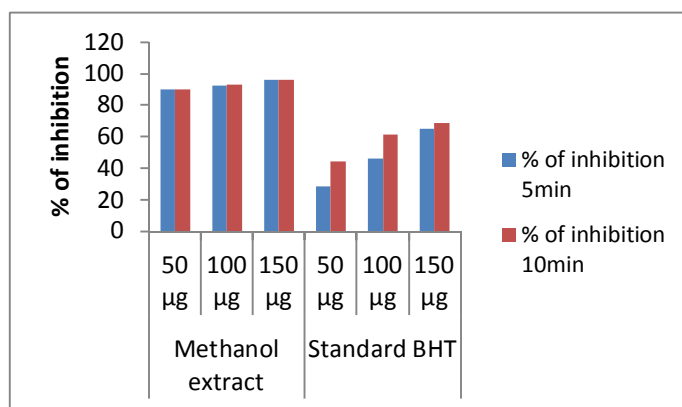


Figure 1: DPPH scavenging activity of methanolic extract of *M. adansonii*

Table 1: DPPH scavenging activity of methanolic extract of *M. adansonii*

Extract / standard	Concentration of extracts (µg/ml)	% of inhibition	
		5min	10min
Methanol extract	50 µg	89.84	90.27
	100 µg	92.58	93.35
	150 µg	96.10	96.38
Standard BHT	50 µg	28.77	44.29
	100 µg	46.37	61.22
	150 µg	65.08	68.99

Reducing power is to measure the reductive ability of antioxidant and it is evaluated by the transformation of Fe (III) to Fe (II) in the presence of sample extracts (Gulcin et al., 2003). The reducing power of methanol extracts of *Monstera adansonii* are summarized in Fig.2 (Table 2). Increasing the concentration of the extracts resulted in increased ferric reducing power for the methanolic extracts. This results is similar to that reported by Gulcin et al., (2003) and Noriham et al., (2004), who demonstrated antioxidative activity on *Pimpinella anisum* seed extracts and four types of Malaysian plants. At 200 ppm, BHA had the highest ability to reduce Fe (III) and had no significant difference with *P. minus* ($P > 0.05$). The ability of reducing power of methanolic extract of *Pimpinella minus* showed almost similar with synthetic antioxidant. BHA at 600, 800, 1000 and 1200 ppm. At 1200 ppm, all methanolic plant extracts have higher ability than BHT to reduce Fe (III) to Fe (II) ($P < 0.05$). The ability to reduce Fe (III) may be attributed from hydrogen donation from phenolic compounds (shimada et al., 1992) which is also to presence of reducing agent (Duh, 1998). In addition, the number and position of hydroxyl group of phenolic compounds also rule antioxidant activity (Rice-Evans et al., 1995).

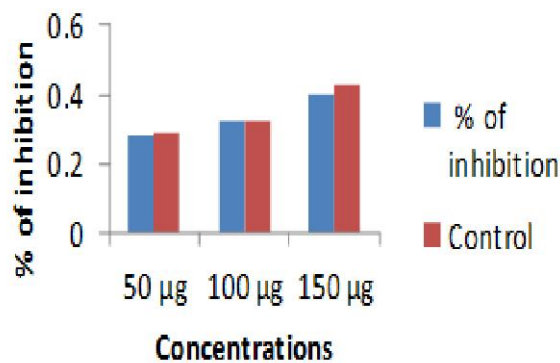


Figure 2: Ferric reducing antioxidant power assay of methanol extracts of *Monstera adansonii*

Table 2: Ferric reducing antioxidant power assay of methanol extracts of *Monstera adansonii*

Concentration of extracts (µg/ml)	% of inhibition	Control
50 µg	0.28	0.29
100 µg	0.32	0.32
150 µg	0.40	0.43

The superoxide radical indirectly initiates lipid oxidation as a result of superoxide and hydrogen peroxide serving as precursors of singlet oxygen and hydroxyl radicals. Therefore, in the present investigation, it was considered important to characterize the scavenging ability of *Monstera adansonii* extracts against the superoxide anion. In the PMS-NADH-NBT system, the superoxide radical derived from dissolved oxygen by PMS-NADH coupling reaction reduces NBT. The decrease in absorbance at 560 nm with antioxidants indicates the consumption of superoxide radicals in the reaction mixture. In this study, the methanol extracts of *Monstera adansonii* was found to be a more potent scavenger of superoxide anion (Table 3; Fig. 3).

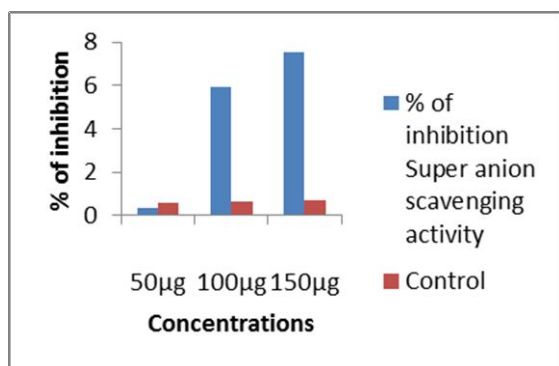


Figure 3: Superoxide anion scavenging activity of methanol extract of *Monstera adansonii*

Table 3: Superoxide anion scavenging activity of methanol extract of *Monstera adansonii*

Concentration of extracts	% of inhibition Super anion scavenging activity	Control
50µg	0.36	0.56
100µg	5.92	0.65
150µg	7.52	0.70

Nitric oxide is a free radical produced in the mammalian cells and is involved in regulation of various physiological processes. However excess production of nitric oxide is associated with several diseases. Methanolic extract of *Monstera adansonii* leaves has demonstrated dose dependent radical scavenging activity against nitric oxide free radicals (Fig.4; Table 4).

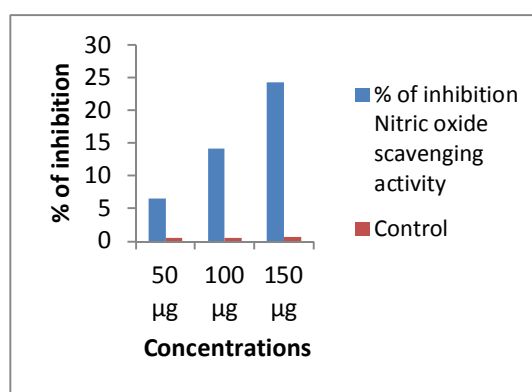


Figure 4: Nitric oxide scavenging activity of methanol extract of *Monstera adansonii*

Table 4: Nitric oxide scavenging activity of methanol extract of *Monstera adansonii*

Concentration of extracts	% of inhibition Nitric oxide scavenging activity	Control
50 µg	6.51	0.45
100 µg	14.19	0.51
150 µg	24.29	0.62

In Recent years, the search for phytochemicals possessing antimicrobial and antioxidant properties have been on the rise due to their potential use in the therapy of various chronic and infectious diseases. Epidemiology and experimental studies have implicated oxidative cellular damage arising from an imbalance between free radical generating and scavenging systems as the primary cause of cardiovascular, diseases, cancer, aging etc. (Halliwell, 1996).

The Plant phenolic compounds have been found to possess potent antioxidants (Adedapo et al., 2009; Adesegum et al., 2009; Lai et al., 2010). The flavonoids from plants extracts have been found to possess antimicrobial and antioxidants properties in various studies (Lin et al., 2008; Lopez-Lazaro, 2009).

The present investigation has shown that the methanol extract of *Monstera adansonii* have active phytochemicals which are able to inhibit free radicals. Strong antioxidant properties were confirmed in the methanol fractions. The results of the present study appear as interesting and promising in the search of potent antioxidant agent and may be effective as potential sources of novel antioxidant drugs.

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