



International Journal of Experimental Pharmacology

www.ijepjournal.com

INVITRO ANTI- OXIDANTS ACTIVITY OF ROOTS OF *MALACHRA CAPITATA L.*

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ABSTRACT

The root of the *Malachra capitata* (L.) is traditional remedies for the many disease condition such as pain, hepatic cirrhosis, inflammation, diarrhea, convulsion, dementia, pyrexia, ulcer, healing of wounds. It is very population perennial herb that is distributed in world wide. The leaves of wood sorrel are quite edible with a tangy taste. In this present study the aqueous extract of roots *Malachra capitata* L. were investigated by using DPPH scavenging test and reducing power method. The preliminary phytochemical screening was performed and the total phenolic content had been estimated. The AMC exhibited a significant dose dependent inhibition of DPPH activity. The IC50 values of the AMC and reference standard ascorbic acid have significant anti-oxidant activity and the similar activity was shown in the other method also.

Keywords: *Malachra capitata* L., Antioxidant, DPPH radical scavenging.

INTRODUCTION

Generation of free radicals or reactive oxygen species (ROS) during metabolism and other activities beyond the antioxidant capacity of a biological system gives rise to oxidative stress [1]. Oxidative stress plays a role in heart diseases, neurodegenerative diseases, cancer and in the aging process [2]. This concept is supported by increasing evidence that oxidative damage plays a role in the development of chronic, age-related degenerative diseases, and that dietary antioxidants oppose this and lower risk of disease. Antioxidants are the substance that when present in low concentrations compared to those of an oxidisable substrate significantly delays or prevents oxidation of that substance [3].

Apart from their role of health benefactors, antioxidants are added in foods to prevent or delay oxidation of food, initiated by free radicals formed during their exposure to environmental factors such as air, light and temperature [4]. At present most of the antioxidants are

manufactured synthetically. They belong to the class of synthetic antioxidants. The main disadvantage with the synthetic antioxidants is the side effects when taken in vivo [5]. Strict governmental rules regarding the safety of the food has necessitated the search for alternatives as food preservatives [6].

Plants are the potential source of natural antioxidants. Natural antioxidants or phytochemical antioxidants are the secondary metabolites of plants [7]. Carotenoids, flavonoids, cinnamic acids, benzoic acids, folic acid, ascorbic acid, tocopherols, tocotrienols etc., are some of the antioxidants produced by the plant for their sustenance. Beta-carotene, ascorbic acid and alpha tocopherol are the widely used antioxidants.

Malachra capitata (L.) is a herb belongs to family: Malvaceae. Description: Mostly erect, coarse, annual or perennial herb 1-2 m tall, throughout densely whitish- or yellowish-tomentose with stellate hairs and usually also moderately to copiously hispid with simple or stellate hairs to 2 mm long; roots long-petioled; stipules lanceolate, 5-15 mm long; blades orbicular to ovate, 2-10 cm long, palmately sinuate to 3-, 5-, or 7-lobed, lobes mostly obtuse, crenate to serrate, the base obtuse or truncate; flowers in axillary, pedunculate, bracteate heads, bracts 1-2 cm long, stipitate and subtended by paired, filiform bracteoles,

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conduplicate, suborbicular to ovate, obtuse or acute, entire or once or twice dentate, obtuse to cordate at base, prominently veined and whitish basocentrally; involucre bracts wanting; calyx tubular-campanulate, 4-8 mm long, 5-lobed to below middle, lobes ovate-lanceolate, white with brownish or reddish nerves; petals yellow, obovate, 10-15 mm long, slightly exceeding staminal column; mericarps 3-3.5 mm long, mucous, reddish veined, puberulent; seed obovoid-cuneate, about 2.5 mm long, black, whitish-pubescent about hilum. The root of the *Malachra capitata* (L.) is traditional remedies for the many disease condition such as pain, hepatic cirrhosis, inflammation, diarrhea, convulsion, dementia, pyrexia, ulcer, healing of wounds [8-11]. Literature survey revealed that the plant extract has yet not been screened for its traditional anti-oxidant activity. Therefore the present study was carried out to provide pharmacological evidence for the folklore medicinal consideration of roots as natural anti-oxidants.

MATERIALS AND METHODS

Collection and authentication of plant material

The Plant material of *Malachra capitata* (L.) roots was collected from Tirunelveli District, in the Month of August 2011. The plant was authenticated by Dr.V.Chelladurai, Research Officer Botany. C.C.R.A.S., Govt. of India. The voucher specimen of the plant was deposited at the college for further reference.

Preparation of plant extract

The roots of the *Malachra capitata* (L.) are properly washed in tap water and then rinsed in distilled water. The rinsed roots are dried in an oven at 35°C for 4 days. The dried roots of *Malachra capitata* was crushed to obtain powder. These powdered samples are then stored in airtight polythene bags protected from sunlight until use. The aqueous extract of each sample was prepared by soaking 10g of powdered sample in 200ml distilled water for 12h. The extracts are then filtered using Whatmann filter paper. Percentage yield of aqueous extract of *Malachra capitata* was found to be 10.5 % w/w.

Phytochemical Screening

The phytochemical examination of aqueous extract of *Malachra capitata* (L.) was performed by the standard methods [12].

ANTIOXIDANT ACTIVITY

DPPH Radical scavenging test

The free radical scavenging activity of the aqueous extracts of *Malachra capitata* L. (AMC) was determined by using 2, 2-Diphenyl-1-picryl hydrazyl radical (DPPH) using UV-Spectrometry [13] at 517nm. The DPPH solution was prepared in 95% aqueous. The AMC was mixed with 95% aqueous to prepare the stock solution (10mg/100ml or 100µg/ml). From the stock solution 2ml, 4ml, 6ml, 8ml and 10ml of this solution were taken in five test tubes and by

serial dilution with same solvent were made the final volume of each test tube up to 10ml whose concentration was then 20µg/ml, 40µg/ml, 60µg/ml, 80µg/ml and 100µg/ml respectively. Freshly prepared DPPH solution (0.004% w/v) was added in each of their test tubes. Containing AMC (20µg/ml, 40µg/ml, 60µg/ml, 80µg/ml and 100µg/ml) and after 10 min, the absorbance was taken at 517nm, using a spectrophotometer (SHIMADZU UV-1700, UV-visible spectrophotometer). Ascorbic acid was used as a reference standard. It is dissolved in distilled water to make stock solution with the same concentration of AMC control sample was prepared without extract and reference ascorbic acid. 95% aqueous was used as blank % scavenging of the DPPH free radical was measured using following equation.

% DPPH radical-scavenging =

$$\frac{(\text{Absorbance of control} - \text{Absorbance of sample})}{\text{Absorbance of control}} \times 100$$

Reducing Power Method

The assay of reducing power method [1,14] is one to determine the antioxidant activity. In this 1 ml of plant extract of AMC solution mixed with 2.5 ml phosphate buffer (0.2M, pH 6.6) and 2.5 ml Potassium Ferricyanide [$K_3Fe(CN)_6$] (10g/l), the mixture was incubated at 50°C for 20 minutes. 2.5 ml of Tri chloroacetic acid (100g/l) was added to mixture. This was centrifuged at 3000 rpm for 10 min. Finally 2.5 ml of the supernatant solution was mixed with 2.5 ml of distilled water and 0.5 ml $FeCl_3$ (1g/L) and absorbance measured at 700nm in UV-visible spectrophotometer (SHIMADZU UV-1700, UV-visible spectrophotometer). Ascorbic acid was used as standard and phosphate buffer used as blank.

RESULTS AND DISCUSSION

Phytochemical investigation

The results of preliminary phytochemical investigation of the aqueous extract of *Malachra capitata* (L.) roots (AMC) shows the presence of carbohydrates, phenols, flavanoids, glycosides, terpenes, alkaloids, tannins, and Saponins.

In this present study the aqueous extract of roots *Malachra capitata* L. (Oxalidaceae) were investigated by using DPPH scavenging test and reducing power method. The roots of AMC showed by their two methods effectively when compared with reference standard ascorbic acid. In the DPPH scavenging method is based on the capability of DPPH radical to decolorize in the presence of antioxidants. The DPPH radical is considered to be model of a stable lipophilic radical a chain reaction. In lipophilic radicals was initiated by the lipid autooxidation antioxidants react with DPPH reducing a number of DPPH molecules equal to number of their hydroxyl groups. Therefore, the absorption at 517 nm was proportional to the amount of residual DPPH

[15]. The AMC exhibited a significant dose dependent inhibition of DPPH activity, the IC50 value of the AMC and reference standard ascorbic acid were found to be 30µg/mL and 37µg/mL respectively. The reducing power method based on the capability of a reducing the compound due to presence of reductants which are breaking the free radical chain by donating hydrogen atom. The roots of AMC exhibited the antioxidant activity due to presence of

reductants (i.e., antioxidants). The reduction of Fe³⁺/Ferricyanide complex to ferrous form, in this main principle is increasing the absorbance of the reaction mixture indicates the antioxidant activity that leads to reducing power of the samples. AMC was very potent and the power of extract was increased with quantity of sample. By comparing the reference standard Ascorbic acid, the AMC showed potent antioxidant activity.

Table 1. Antioxidant activity by DPPH method

S. No.	Concentration (µg/ml)	Absorbance of ascorbic acid	Absorbance of AMC	% scavenging DPPH of Ascorbic acid	%scavenging DPPH of AMC
1	20µg/ml	0.152	0.139	36.14	42.25
2	40µg/ml	0.104	0.090	54.44	60.16
3	60µg/ml	0.088	0.077	62.72	70.42
4	80µg/ml	0.068	0.049	74.24	83.12
5	100µg/ml	0.042	0.021	86.32	92.17

DPPH radical scavenging activity of aqueous extracts of *Malachra capitata* L. (AMC) added to aqueous solution of DPPH and radical scavenging activity was measured as 517 nm as compared to standard Ascorbic acid. Values are the average of triplicate experiments.

Table 2. Antioxidant activity by reducing power method

S. No.	Concentration (mg/ml)	Absorbance of Ascorbic acid	Absorbance of AMC
1	0.1	0.18	0.13
2	0.2	0.24	0.22
3	0.3	0.33	0.32
4	0.4	0.41	0.36
5	0.5	0.51	0.44

Reducing power of aqueous extract of *Malachra capitata* L. (AMC) of as compared to Ascorbic acid. Values are the average of triplicate experiments.

CONCLUSION

It is concluded from the data, that the aqueous extract of *Malachra capitata* L. possess significant Antioxidant activity and may prove to be effective for the treatment of various diseases caused by free radicals. The

antioxidant activity may be rich in flavanoids, glycosides, terpenes in this plant. However further studies required to elucidate the exact mechanism of action for develop its as potent antioxidant.

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