



Evaluation of antioxidant property of *Desmodium gangeticum* and *Pseudarthria viscida* roots

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ABSTRACT

Various studies have been done to identify antioxidants from plant sources and efforts have been taken to incorporate it in conventional therapy. In our present study, alcoholic and aqueous extracts of *Desmodium gangeticum* (one of the main ingredients of famous Ayurvedic preparations-Dasamularistha, Dashmoola ark, Dashmoola taila, etc.,) and its substituent *Pseudarthria viscida* roots (used in the preparation of Ayurvedic medicines namely Dashamoola, Mahanarayana taila and Dhantara taila) have been evaluated for in vitro antioxidant activity using DPPH radical scavenging, Nitric Oxide scavenging activity and reducing power assay methods. Ascorbic acid (100 µg/ml) was used as standard antioxidant. All the analysis was made with the use of UV-Visible spectrophotometer. The results of the assay showed that the extracts of *Desmodium gangeticum* and *Pseudarthria viscida* roots possess significant free radical scavenging and reducing power properties. Hence, it can be concluded that the *Desmodium gangeticum* and *Pseudarthria viscida* roots could be pharmaceutically exploited for antioxidant properties.

Keywords: DPPH scavenging, Nitric Oxide scavenging, reducing power, *Desmodium gangeticum*, *Pseudarthria viscida*

INTRODUCTION

Oxygen is essential for the survival of all on this earth. During the process of oxygen utilization in normal physiological and metabolic processes approximately 5% of oxygen gets univalent, reduced to oxygen derived free radicals like super oxide, hydrogen peroxide, hydroxyl and nitric oxide radicals [1,2]. All these radicals known as reactive oxygen species (ROS) exert oxidative stress towards the cells to face about 10000 oxidative hits per second [3]. When generation of ROS overtakes the antioxidant defense of the cells, the free radicals start attacking the cell proteins, lipids and carbohydrates and this leads to a number of physiological disorders. Many plants often contain substantial amount of antioxidants including vitamin C and E, carotenoids, flavonoids and tannins etc., and thus can be utilized to scavenge the excess free radicals from human body. The present study was conducted to evaluate the anti oxidant properties of alcoholic and aqueous extracts of *Desmodium gangeticum*- one of the main ingredient of famous Ayurvedic preparations like Dasmularistha, Dasamulakwath, Dasamoola kadha, and several other Ayurvedic formulations [4] and its substituent *Pseudarthria viscida* roots - the drug is used as an ingredient of a number of official preparations of classical remedies such as Agastya haritaki rasayana, Brahma rasayana, Salaparnyadi kwatha, Laghu panchamula kwatha [5].

EXPERIMENTAL SECTION

Plant Material

The roots of *Desmodium gangeticum* and *Pseudarthria viscida* were collected from Tamil University, Thanjavur and Kutralam, Tamilnadu, India respectively. The plants were botanically identified and authenticated by Dr.M.Jegadeesan, Prof. & Head, Department of Environmental and Herbal Sciences, Tamil University, Thanjavur. The voucher specimens have been deposited at the Tamil University Herbarium, Thanjavur (*Desmodium gangeticum* TUH- 274); (*Pseudarthria viscida* TUH-275).

Chemicals

1, 1 - Diphenyl -2 picryl hydrazyl (DPPH), ascorbic acid were purchased from Himedia Ltd., Mumbai. Sodium nitroprusside and sulphanilamide, N-(1-naphthyl) ethylenediamine, were purchased from Loba Chemicals, India. All the reagents used for the study were of analytical grade. A Shimadzu model 2401 double beam UV- visible spectrophotometer with a pair of 10 mm matched quartz cells was used to measure absorbance of the resulting solution.

Assay for Antiradical Activity [6]

Antiradical activity was measured by a decrease in absorbance at 516 nm with the methanol solution of colored DPPH. A stock solution of DPPH was prepared to produce the concentration such that 75 mg of it in 2 ml of methanol gave an initial absorbance of 0.9. This stock solution was used to measure the antiradical activity. Decrease in absorbance in the presence of the test compounds i.e alcoholic and aqueous extracts of *Desmodium gangeticum* and *Pseudarthria viscida* roots at a concentration of 500 µg/ml were noted after 15 minutes. Ascorbic acid was used as standard (100 µg/ml). All tests were performed in triplicate. Radical scavenging activity was expressed as the inhibition percentage of free radical by the sample and was calculated using the formula

$$\% \text{inhibition} = \frac{(\text{Absorbance}_{\text{control}} - \text{Absorbance}_{\text{test}})}{(\text{Absorbance}_{\text{control}})} \times 100 \quad (1)$$

Assay for Nitric Oxide Scavenging Activity [7]

The procedure is based on the principle that, sodium nitroprusside in aqueous solution at physiological pH spontaneously generates nitric oxide which interferes with oxygen to produce nitric ions that can be established using Griess reagent (1% sulphanilamide, 2% phosphoric acid and 0.1% naphthyl ethylene diamine dihydrochloride). Scavengers of nitric oxide compete with oxygen leading to reduced production of nitric ions.

For the experiment, sodium nitroprusside (5mM) in phosphate buffered saline was mixed with a concentration of 500 µg/ml of alcoholic and aqueous extracts of *Desmodium gangeticum* and *Pseudarthria viscida* and incubated at room temperature for 150 minutes. The same mixture without the extracts of the sample but with equivalent amount of water served as control. After incubation period, 0.5 ml of Griess reagent was added. The absorbance was noted at 546 nm. Ascorbic acid was used as positive control. All the tests were performed in triplicate. Percentage inhibition was calculated using Equation 1.

Reducing Power

Alcoholic and aqueous extracts of *Desmodium gangeticum* and *Pseudarthria viscida* roots (500 µg) in 1 ml of distilled water were mixed with 2.5 ml of phosphate buffer (0.2mM, pH 6.6) and 2.5 ml potassium ferricyanide 1% , and then the mixtures were incubated at 50° for 30 minutes. Subsequently 2.5 ml of trichloroacetic acid (10%) was added to the mixture, which was then centrifuged at 3000 rpm for 10 minutes, finally 2.5 ml of upper layer solution was mixed with 2.5 ml distilled water and 0.5 ml ferric chloride (0.1%) and absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power. [8]

RESULTS AND DISCUSSION

The antioxidant activities as well as reducing activity of alcoholic and aqueous extracts of *Desmodium gangeticum* and *Pseudarthria viscida* evaluated by in - vitro methods exhibited different level of antioxidant activity in models studied. The extracts showed significant antiradical activity of inhibiting DPPH radical at a concentration of 500 µg/ml. Also nitric oxide scavenging activity and reducing powers of the test substances were found to be significant at a dose of 500 µg/ml (Table 1). (P < 0.01).

The percentage inhibition of free radical scavenging activity of alcoholic extract of *Desmodium gangeticum* was slightly higher (88.24 ± 1.64 %) when compared with the alcoholic extract of *Pseudarthria viscida* (84.12 ± 1.08 %) whereas, the aqueous extract of *Pseudarthria viscida* exhibited slightly higher percentage of inhibition (89.18 ± 1.25 %) than that of aqueous extract of *Desmodium gangeticum* (85.21 ± 1.78 %).

The percentage inhibition of nitric oxide scavenging activity were similar for both alcoholic extracts of *Desmodium gangeticum* and *Pseudarthria viscida* whereas, the aqueous extract of *Desmodium gangeticum* exhibited slightly higher percentage of inhibition ($63.72 \pm 1.85\%$) than that of aqueous extract of *Pseudarthria viscida* ($60.58 \pm 2.75\%$).

DPPH is one of the free radicals generally used for testing preliminary radical scavenging activity of a compound of a plant extract [9]. In the present study, alcoholic and aqueous extracts of *Desmodium gangeticum* and *Pseudarthria viscida* showed a good antiradical activity by scavenging DPPH radicals. In addition to this, nitric oxide is also implicated in inflammation, cancer and other pathological condition. The extracts showed nitric oxide scavenging activity.

The reducing capacity of compound may serve as significant indicator of its potential antioxidant activity [10]. The present finding signifies that the extracts of *Desmodium gangeticum* and *Pseudarthria viscida* might be potential sources of natural antioxidants.

Table 1: Antioxidant activity of root extracts of *Desmodium gangeticum* and *Pseudarthria viscida*

Drug treatment	Concentration ($\mu\text{g/ml}$)	Percentage Inhibition	
		Free radical scavenging activity	Nitric Oxide scavenging
Ascorbic acid	100	92.12 ± 1.82	90.66 ± 1.54
DG-alcohol	500	88.24 ± 1.64	74.11 ± 1.32
DG-aqueous	500	$85.21 \pm 1.78^*$	63.72 ± 1.85
PV-alcohol	500	84.12 ± 1.08	$74.92 \pm 1.62^*$
PV-aqueous	500	89.18 ± 1.25	60.58 ± 2.75

$n = 5$

Values are expressed as mean \pm SEM * $P < 0.01$

DG - *Desmodium gangeticum*

PV - *Pseudarthria viscida*

Reducing power of root extracts of *Desmodium gangeticum* and *Pseudarthria viscida*

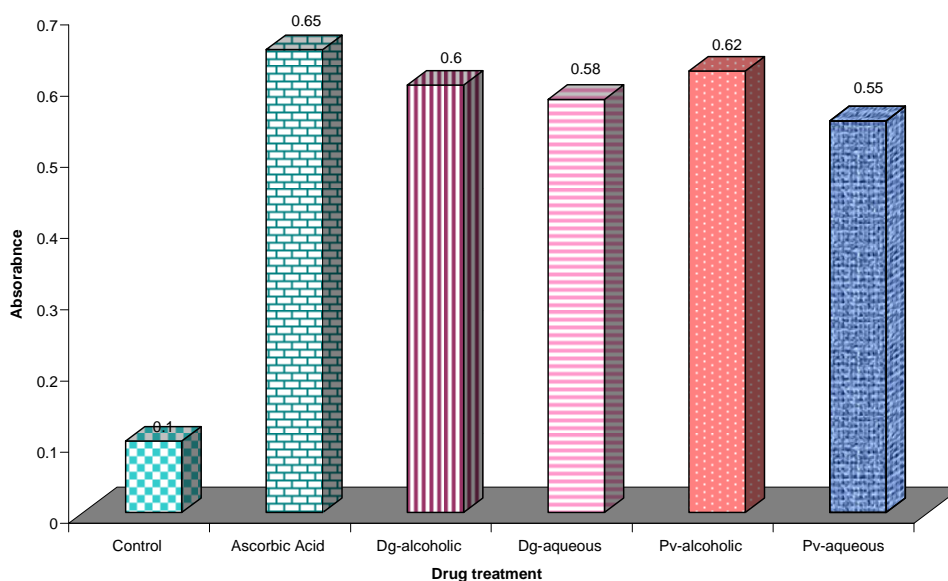


Fig -1 Ferric Reducing power of root extracts of *Desmodium gangeticum* and *Pseudarthria viscida*

CONCLUSION

The findings of this study support the view, that the root extracts of plants *Desmodium gangeticum* and its substituent *Pseudarthria viscida* are promising sources of potential antioxidant and may be efficient as preventive agents in some diseases and general health tonic and can be considered as a natural herbal source in pharmaceutical industry.

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