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A STUDY OF ANTIOXIDANT ACTIVITY, MINERAL AND TOTAL VITAMIN C CONTENTS OF STEM, LEAVES, AND INFLORESCENCE SPIKE OF *ACHYRANTHES ASPERA* VAR. PORPHYROSTACHYA

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ABSTRACT

The present study has been carried out to evaluate antioxidant activity of different extracts of stem, leaves and inflorescence spike of *A. aspera* var. porphyrostachya by using the DPPH assay and reducing power assay. The results were compared with ascorbic acid as a standard. Among the various extracts ethyl acetate, ethyl alcohol, and methyl alcohol extracts were found to be the most effective extracts. In support of this mineral study as well as total vitamin C content was also carried out for stem, spike and leaves. Total vitamin C content in stem, spike and leaves was found 28.125 mg/100g, 50mg/100g and 46.876mg/100g respectively .This study suggests that the *A.aspera* var. porphyrostchya plant could be pharmaceutically exploited for antioxidant properties and ethyl acetate, ethyl alcohol and methyl alcohol are good organic solvents for further exploration.

Key words: *Achyranthes aspera* var. Porphyrostachya, DPPH, Reducing Power assay, Vitamin C, Antioxidant properties.

INTRODUCTION

Reactive oxygen species (ROS) play an important role in oxidative stress which is responsible for the causation of various important diseases (Finkel T and Halbrook NJ, 2000). ROS are generated as a byproduct of biological reactions or from exogenous factors (Maryam Z *et al*., 2009). Antioxidants are compounds that protect cells against the damaging effects of ROS. Thus, compounds that can scavenge free radicals have vital role in the diseased condition (Jaysri MA *et al*., 2009). Most of the antioxidant compounds are flavonoids, isoflavones, caumarins, lignans, anthocyanins, flavones, isocatechins, vitamin C, vitamin E, carotens, tocopherol, etc (Yalla Reddy K *et al*., 2010).

The plant *Achyranthes aspera* var. porphyrostchya is a very rare variety of species *A. aspera* belongs to the family Amaranthacea. This plant is used as folk medicine for the treatment of piles, gynecological disorder, dysentery, fever, etc.

There is no scientific information regarding medicinal use of this plant. The present study aims to evaluate the antioxidant property of *A. aspera* var. porphyrostchya to justify the folklore use of this plant and to find the rout for further phytochemical investigation. Mineral and vitamin C study also supports the antioxidant properties of the plant

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MATERIALS AND METHODS

Plant Material: The whole plant of *A. aspera* var. porphyrostchya was collected from Kasara village of District Nasik Maharashtra (India) in the month of October. The plant authenticated by National Botanical Survey of India (NBSI), Pune. The aerial part of the plant was cut in to stem, inflorescence spike and leaves. Each part then air dried in shadow separately and grounded in to fine powder.

Preparation of plant extracts

15gm of plant material was extracted with 250ml

of different solvents. To prepare plant extract petroleum ether, chloroform, ethyl acetate, ethyl alcohol, and methyl alcohol solvents were used separately (Lalita NR, 2002). Extracts were evaporated under reduced pressure and final residues were used for assessment of antioxidant activity by using DPPH (2, 2-Diphenyl-1-picryl hydrazyl radical) and reducing power methods.

ANTIOXIDANT PROPERTIES

DPPH Method

The free radical scavenging activity of different extracts of *A. aspera* var pophyrosctachya was determined by using 2,2-Diphenyl-1-picrylhydrazyl radical at 517 nm using UV-spectrophotometer (uv-1800, SIMADZU) (Vishal DJ *et al*., 2009; Rumi G *et al*., 2010; Aziz T *et al*., 2007) .0.004% DPPH solution was prepared in methanol. Dried extracts then mixed with methanol to prepare the stock solution of 10mg/100ml concentration. From the stock solution 2ml, 4ml, 6ml, 8ml and 10ml solutions were taken in five test tubes and by serial dilution with methanol were made up the final volume of each test tube up to 10ml, whose concentrations were then 20ug/ml, 40ug/ml, 60ug/ml, 80ug/ml and 100ug/ml respectively. Freshly prepared DPPH solution was then added in each test tube. Each test tube then incubated at 37^{o}C for 30 minutes, the absorbance was taken at 517nm. Ascorbic acid was used as reference standard. The DPPH solution without sample was used a control, methanol used as blank. % scavenging of the DPPH free radical was measured using following formula.

$$\%\text{DPPH Scavenging} = \frac{\text{Absorbance of Control- Absorbance of test sample}}{\text{Absorbance of control}} \times 100$$

Reducing Power Method

Reducing power method is based on the principle that substances, which have reducing potential react with potassium ferricyanide (Fe^{3+}) to form potassium ferrocyanide (Fe^{2+}) which then reacts with ferric chloride to form a complex. This complex has an absorbance maximum at 700nm. The reducing capabilities of different extracts of stem, spike and leaves were compared with ascorbic acid. 1ml extract solution mixed with 2.5ml phosphate buffer (0.2M, pH 6.6) and 2.5ml 1% potassium ferricyanide ($K_3[Fe(CN)_6]$), the mixture was then incubated at 50^{o}C for 20 minutes, 2.5ml of 10% trichloroacetic acid was added, mixture then centrifuged at 3000rpm for 10 minutes. Finally 2.5ml of the supernatant solution was mixed with 2.5ml of distilled water and 0.5ml 0.1% $FeCl_3$ and absorbance was measured at 700nm on uv-spectrophotometer (uv-1800, SHIMADZO). Ascorbic acid was used as standard and phosphate buffer as blank.

Determination of Vitamin C content

Vitamin C was quantitatively determined according to 2, 6-dichlorophenolindophinol dye method (Ranganna S, 1997; Sharique A *et al*., 2009). Vitamin C content of powdered sample of spike, stem and leaves was done by extracting 10gm of sample using 3% metaphosphoric acid and the extract has made up to a volume of 100 cm^3 .

10cm^3 of this was then titrated against standard 2, 6-dichlorophenolindophenol dye which was already standardized against standard vitamin C solution.

Mineral content

15gm of plant powder of stem spike and leaves taken in labeled crucibles and heated in hot spot furnace at 550^{o}C for 3 hrs. The sample was removed and cooled in a desiccator, 0.5gm of sample was taken in a 250ml beaker and digested in 20ml of aqua-regia, then 10ml of 30% H_2O_2 was added, the beaker was then covered with watch glass and heated on hot plate at 90^{o}Cin a fume cub-board for about 1hr so that the volume reduced to 2 cm^3. This is then diluted using distilled water in 100ml volumetric flask (Madziga HA *et al*., 2010; Sanni S *et al*., 2008). Then samples were analyzed using Inductively Coupled Plasma Atomic Emission (ICP-AES) for Na, K, Mg, Ca, Mn, Fe, Cu, and Zn.

Table 1. Total vitamin C content in spike, leaves and stem

Plant Part	Total Vitamin C Content in mg/100gm
Spike	50.000
Stem	28.125
Leaves	46.876

Table 2. Elemental analysis in spike, stem and leaves

Sample	Na	K	Mg	Ca	Mn	Fe	Cu	Zn
	ppm	ppm	ppm	ppm	ppm	ppm	ppm	ppm
Blank	0.442	ND*	0.279	0.646	ND*	ND*	ND*	ND*
Stem	13.018	>1645.53	100.259	294.165	1.381	9.715	0.358	1.055
Leaves	21.022	>1268.17	100.633	760.216	3.784	19.388	0.136	0.68
Spike	14.397	1116.51	100.739	608.557	3.533	74.414	1.165	1.613

*less than 0.01ppm

Fig 1. Antioxidant Properties of Spike by DPPH method

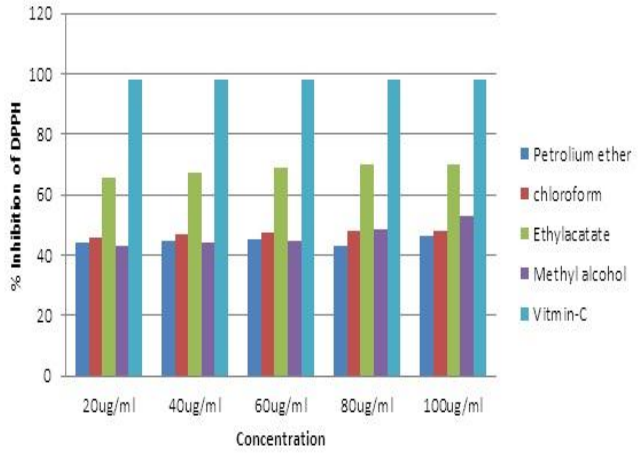


Fig 2. Antioxidant Properties of Stem by DPPH method

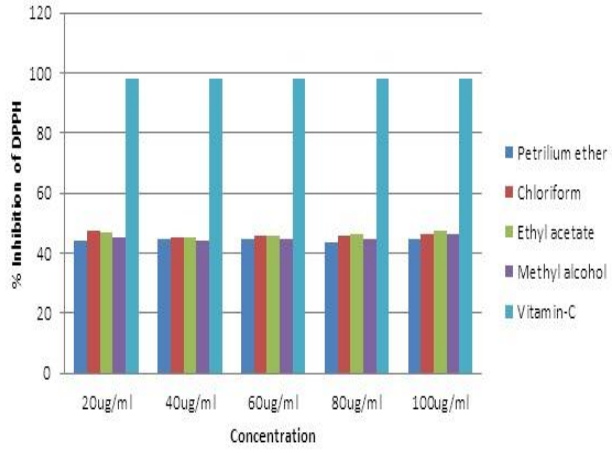


Fig 3. Antioxidant Properties of Leaves by DPPH method

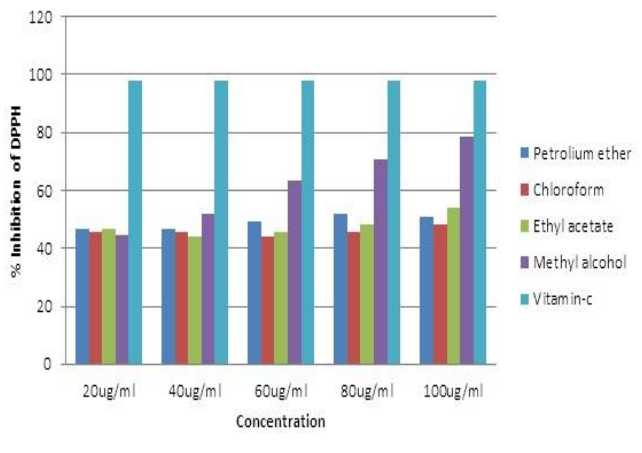


Fig 4. Antioxidant properties of spike by reducing power method

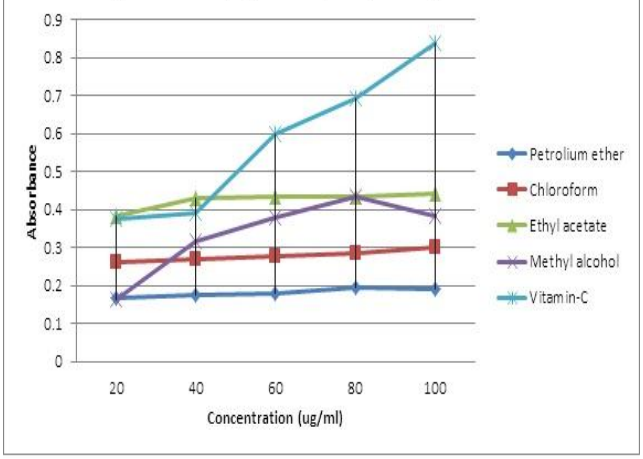


Fig 5. Antioxidant properties of stem by Reducing power method

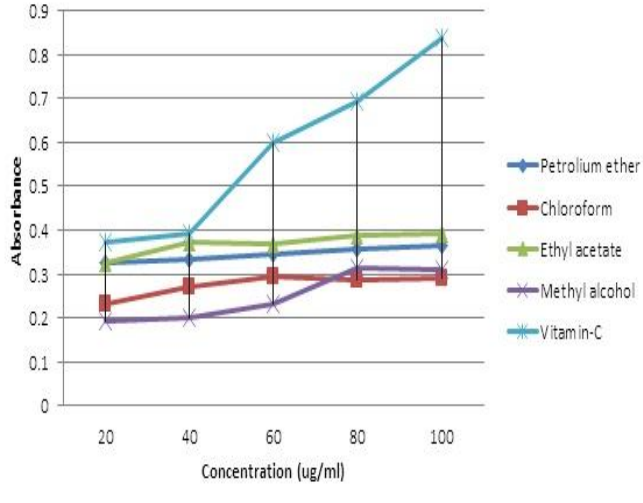
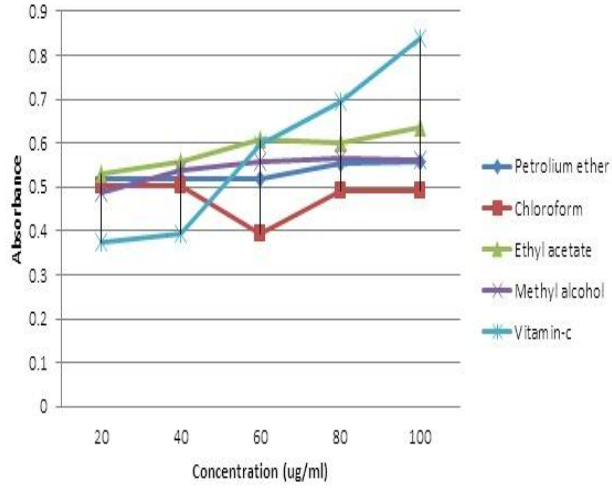


Fig 6.Antioxidant properties of Leaves by Reducing power method



RESULT AND DISCUSSION

Ethyl acetate extract of inflorescence spike, stem and leaves show good antioxidant property in both DPPH and reducing power methods. The results of vitamin C content study shows the maximum vitamin C content in spike. Elemental analysis shows the presence of macro and micro nutrients. Macro nutrients such as Na, K regulate the fluid balance; Ca plays an important role in physiological and biochemical processes like blood coagulation. Elements like Fe, Mn, and Zn are essential because they are important in several enzyme reactions as co-factors.

CONCLUSION

From this experiment it is revealed that ethyl acetate is a good solvent for the isolation of active phytochemicals and spike could be a good source of phytochemicals. Elemental analysis of plant also supports its use as a medicinal plant.

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REFERENCES

Finkel T and Halbrook NJ. Oxidants, oxidative stress and biology of aging. *Nature,* 408, 2000, 239-247.

Maryam Z, Farrukh A and Iqbal A. The in-vitro antioxidant activity and total phenolic content of four Indian medicinal plants. *Int. Journal of Pharmacy and Pharmaceutical Sciences,* 1(Supp l), 2009, 88-95.

Jaysri MA, Lazar M, Radha A. A Report on the antioxidant activity of leaves and rhizomes of Costus pictus D. Don. *Int. Journal of Intigrative Biology*, 5(1), 2009.

Yalla Reddy K, Sarvana Kumar A, Mohana Lakshmi S, Surendar A. Antioxidant properties of methanolic extract of Oxalis corniculata. *Int. Journal of Phytopharmacology,* 1, 2010, 43-46.

Lalita NR. Isolation and Identification of Chemicals Constituents from Plant Zanthoxylum budrunga DC Wall, Ph.D.Thesis, University of Mumbai, 2002.

Vishal DJ, Tekeshvar V, Prajwal RS. Antioxidant potential of Bauhinia purpurea Linn. Leaves. *Int. Journal of Pharmaceutical Research,* 1(2), 2009, 51-55.

Rumi G, Parag K, Vilasrao K. *Int. Journal of Pharma Research and Development,* 1(11), 2010, 1-8.

Aziz T, Mehmet ED and Nazime M. Antioxidant and antimicrobial activity of Russula delica Fr: An edible wild mushroom. *Eurasian Journal of Analytical Chemistry,* 2, 2007, 54-67.

Francis OA. Phytochemical screening of some Nigerian woody plants. *Research Journal of Phytochemistry,* 1(1), 2007, 33-39.

Ranganna S. Mannual of analysis of fruit and vegetable products. 9th ed, Tata Mc-Graw Hill, New Delhi, 1997.

Sharique A, Seerat HB. *Journal of Medicinal and Biological Sciences*, 3, 2009, 1-8.

Madziga HA, Sanni S and Sandabe UK. Phytochemical and Elemental Analysis of *Acalypha wilkesiana* leaf. *Journal of American Science*, 6(11), 2010, 510-514.

Sanni S, Onyeyili PA and Sanni FS. Phytochemical analysis, elemental determination and some *in-vitro* antibacterial activity of *Ocimum basilicum* L. leaf extracts. *Research Journal of Phytochemistry,* 2(20), 2008, 77-83.