

RESEARCH ARTICLE

Hydroxyl radical activity of *Amaranthus retroflexus* leaves

G. Jyoti Jain^{*1} and S. Ramachandra Setty²1.Dept. of Pharmacology, Luqman College of Pharmacy,
Gulbarga, Karnataka, India.2.Dept. of Pharmacology, Govt. college of Pharmacy,
Bangaluru, Karnataka, India.Date Received: 3rd March 2016; Date Accepted: 15th March
2016 Date published: 17th March 2016Email: jyotilcp2@gmail.com**ABSTRACT**

Amaranthus retroflexus is a medicinal plant. Our aim is to investigate its invitro antioxidant property. Hence 70% ethanolic extract of *Amaranthus retroflexus* leaves (AREE) was taken and the parameter studied was hydroxyl free radical scavenging activity. *In-vitro* model was carried out to evaluate its antioxidant activity. Therefore these results concluded that, the ethanolic extract afford significant antioxidant activity which may be attributed due to polyphenols.

Key words : AREE. hydroxyl radical and polyphenols.

INTRODUCTION

Amaranthus retroflexus is a edible plant used as vegetable which is being used by native practitioner as hepatoprotective in treating various types of jaundice. The leaves of this plant contain polyphenolic compounds like tannins and flavonoids. These polyphenolic compounds have antioxidant property. Therefore antioxidants have been known to possess hepatoprotective activity. Keeping the native knowledge and the above mentioned literature information¹, this plant was selected for present study to screen the leaves of this edible plant for the presence of phytoconstituents, and antioxidant activity. This study was carried out by using AREE as antioxidant.

MATERIALS AND METHOD

Collection and identification of plant: The plant was collected from Kusnoor village (Gulbarga district), in the

month of March and was authenticated by Dr. Srinath Rao, chairman, P.G. Department of Studies and Research in Botany, Gulbarga University, Gulbarga, Karnataka. The plant was thoroughly cleaned and the leaves were shade dried and made into a coarse powder by rubbing in the palms.

Extraction

250 gms of shade dried leaf powder of *Amaranthus retroflexus* was extracted in Soxhlet's apparatus using petroleum ether for defatting and then it was extracted with 70% ethanol. This solvent was evaporated on a water bath at a low temperature (50°C) and finally the residue was obtained.

Materials used

All chemicals and reagents used were of analytical grade.

In-vitro* antioxidant activity*Hydroxyl radical scavenging activity**

Hydroxyl radical scavenging activity of AREE was studied by using method of Sasanka et. al.²

In biochemical systems, superoxide radical and H₂O₂ react together to form a hydroxyl radical OH[•], this attacks and destroys almost all known biomolecules. When phenyl hydrazine is added to erythrocytes, it causes peroxidation of endogenous lipids and alteration of membrane fluidity. This peroxidative damage to erythrocytes is probably initiated by active oxygen species like O₂^{•-}, OH[•] and H₂O₂, which are generated in solution from autooxidation of phenyl hydrazine. This forms the basis of this experiment².

Procedure:

Hydroxyl radical generation by phenyl hydrazine has been measured by the 2-deoxyribose degradation assay of Hathwell and Gutteride³. In 50 mM phosphate buffer (pH 7.4) 1 mM deoxyribose, 0.2 mM phenyl hydrazine hydrochloride were prepared. 0.6 ml of 1 mM deoxyribose and 0.4 ml of AREE (varying doses 10, 20, 25, 50 and 100 µg) and sodium metabisulphate (25µg Std.) were taken. 0.2 ml of Phosphate buffer was added to make the volume to 1.6 ml. The reaction mixture was incubated for 10 min and 0.4 ml of 0.2mM phenyl hydrazine HCl was added and incubated for 1 hr and 1 ml each of 2.8% TCA and 1% (w/v) of thiobarbituric acid were added. The reaction mixture was heated for 10 minutes on a boiling water bath. The tubes were cooled and absorbance was taken at 532 nm. by using UV- double beam spectrophotometer. Decrease in the absorbance is indicating the increase in the hydroxyl free radical scavenging activity. The results are compiled in Table No.1.

% Radical scavenging activity = $\frac{\text{control Abs} - \text{sample Abs}}{\text{Control Abs}} \times 100$

Statistical analysis

The data presented in Table No. 1 (n=3) were expressed

as mean \pm SEM. Significant difference among the mean were calculated at the level of $p < 0.001$ and analyzed by one-way analysis of variance by Dunnet's 't' test. A value of $p < 0.05$ was defined as significant.

Table No. 1. Hydroxyl Radical scavenging activity of 70 % ethanolic extract of *Amaranthus Retroflexus* leaves

Groups	Absorbance Mean \pm SEM	% Inhibition
Control	0.313 \pm 0.003	---
Control + Standard 25 μ g	0.103 \pm 0.003***	67.741
Control + 70 % ethanolic extract 10 μ g	0.333 \pm 0.003***	6.451
Control + 70 % ethanolic extract 20 μ g	0.276 \pm 0.003***	12.903
Control + 70 % ethanolic extract 25 μ g	0.263 \pm 0.003***	16.129
Control + 70 % ethanolic extract 50 μ g	0.246 \pm 0.003***	22.580
Control + 70 % ethanolic extract 100 μ g	0.133 \pm 0.003**	58.064

Values are the mean \pm S.E.M., n=3; Significance ***P<0.001, **P<0.01, *P<0.05, compared to standard. Std: Sodium metabisulphate

RESULTS

Hydroxyl radicle is extremely reactive, most common, highly damaging oxygen species which is generated in our body. It affects almost all components of cells. It is clear from results of hydroxyl scavenging activity that this extract showed a good scavenging activity of 58.064%.

DISCUSSIONS

The invitro-antioxidation offered by AREE may be due to the presence of antioxidant phytoconstituents like flavonoids, phytosterols and other polyphenolic constituents. Therefore this extract showed a very good antioxidant activity. These findings add strength to our claim.

CONCLUSION

AREE has good in-vitro antioxidant properties which are attributed due to the presence of antioxidant phytoconstituents. Therefore the above findings reveals that the use of *Amaranthus retroflexus* leaves in our food protects our vital organs from various types of diseases.

SCOPE FOR FUTURE STUDY

As it is a medicinal plant, hence isolation of its phytoconstituents are needed to screen various organ protective potentials.

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REFERENCES

1. www.google.co.in – wikipedia, the free encyclopedia : navigation search.
2. Sasanka Chakrabart, Asha Naik S, Gali Reddy R. Phenylhydrazine mediated degradation of bovine serum albumin and membrane proteins of human erythrocytes. Bioch et Biophy Acta. 1990; 1028: 89-94.
3. Barry Hathwell, John Gutteridge MC. Formation of a thiobarbituric acid reactive substance from deoxyribose in the presence of iron salts. FEBS Letters 1981; 128(2): 347-52.