Research article Available online www.ijsrr.org ISSN: 2279–0543

International Journal of Scientific Research and Reviews

Evaluation of antioxidant activity of aerial parts of hibiscus vitifolius linn

A.Santhi Priya

Department: pharmacology, Assistant professor, Sir C.R.Reddy college of pharmaceutical sciences, Shanti Nagar, Eluru, Andhra Pradesh, India,
Email litishkumarvudata@gmail.com, Mob.9441610750.

ABSTRACT:

The present study indicating an antioxidant effect in ethanolic extract of *Hibiscus vitifolius Linn.* It is also having antiurolithiatic effect & anti diuretic effect. The extract was screened for possible antioxidant activities by 2,2'-azino-bis-3-ethyl benzthiazoline-6-sulphonic acid (ABTS) assay & free radical scavenging activity(DPPH), The results indicate that the administration of ethanolic extract of Hibiscus vitifolius Linn, Significantly shows anti oxidant activity by using ABTS method and DPPH method.

KEYWORDS: Anti oxidant activity, antiurolithiatic effect, Hibiscus vitifolius Linn., free radical scavenging activity(DPPH), 2,2'-azino-bis-3-ethyl benzthiazoline-6-sulphonic acid (ABTS) assay.

***Corresponding author:**

A.Santhi Priya

Department: pharmacology,

Assistant professor,

Sir C.R.Reddy college of pharmaceutical sciences,

Shanti Nagar, Eluru, Andhra Pradesh, India,

Email litishkumarvudata@gmail.com, Mob.9441610750.

INTRODUCTION:

Oxidative stress is an important risk factor in the pathogenesis of numerous chronic diseases. Free radicals and other reactive oxygen species are recognized as agents involved in the pathogenesis of sicknesses such as asthma, inflammatory arthropathies, diabetes, Parkinson's and Alzheimer's diseases, cancers as well as atherosclerosis. Reactive oxygen species are also said to be responsible for the human aging[1,2].

An antioxidant can be broadly defined as any substance that delays or inhibits oxidative damage to a target molecule[3.] The main characteristic of an antioxidant is its ability to trap free radicals. Antioxidant compounds like phenolic acids, polyphenols and flavonoids scavenge free radicals such as peroxide, hydroperoxide or lipid peroxyl and thus inhibit the oxidative mechanisms that lead to degenerative diseases[4]. Herbal plants considered as good antioxidant since ancient times. Several plant extracts have been used to treat oxidative stress with promising effects, both in its prevention and treatment. Thus the objective of the present study is to evaluate the Anti-oxidant activity of Hibiscus vitifolius Linn.[5,6,7,8,9.]

MATERIALS AND METHODS:

Plant Extraction The aerial part of Hibiscus vitifolius Linn [10, 11, 12] were collected from the farmlands of West Godavari district, Andhrapradesh. Care was taken to collect only the healthy parts. The collected parts were authenticated at the Department of Botany, Acharya Nagarjuna University, India. The parts were then shade dried, coarsely powdered in such a way that it passed through sieve no. 20 and was retained on sieve no. 40. About 500g of the dry powder was extracted continuously in soxhlet apparatus with 99% ethanol for 72 h. After 72 h, the solvent was evaporated to obtain the crude extract. The extract was then dried under vacuum and suspended in water before use. The preliminary phytochemical screening gave positive results for carbohydrates, alkaloids, flavanoids, steroids, glycosides, saponins, tannins and phenolic compounds[13].

Animal Experimentation:

Animal facility of this institute is approved by CPCSEA, NewDelhi. The experimental protocols for the antioxidant activity have been approved by the Insititutional Animal Ethics Committee (IAEC) and conducted according to the guidelines of Indian National Sciences Academy for the use and care of experimental animals. The animals were maintained at a well ventilated, temperature controlled 30oC±1oC animal room for 7days prior to the experimental period and provided with food and water ad libitum. The animals were acclimatized to laboratory conditions before the test (16) s. Each animal was used only once[14].

Experimental Animals :

Adult Wistar albino rats weighing 200-220 g were used for the study. In the laboratory, rats were fed with standard rat pellet diet (Lipton India Ltd, Bangalore) and water ad libitum. They were housed in Tarson's polypropylene cages with metal grill tops and acclimated to the laboratory conditions.[15]

Effect of antioxidant activity of Ethanolic Extract of *Hibiscus vitifolius In vitro* evaluation

2,2'-azino-bis-3-ethyl benzthiazoline-6-sulphonic acid (ABTS) assay [16, 17, 18]

Procedure :

The scavenging activity of the test sample was tested using $ABTS^+$ assay. The $ABTS^+$ radical solution was prepared by mixing 14mM ABTS stock solution with 4.9 mM ammonium per sulphate and incubating 16 h in the dark at room temperature until the reaction was stable. The absorbance of the $ABTS^+$ solution was equilibrated to 0.70±0.02 by diluting with ethanol at room temperature. To 1ml of the $ABTS^+$ solution various concentration of the test sample (20-100 µg/ml) was added. The absorbance was measured at 734nm after 6minutes. When ABTS reacts with antioxidants in the sample, it was reduced and the end point was blue in colour. The percentage inhibition was calculated and plotted as a function of the concentration of standard and sample to determine the antioxidant concentration. Ascorbic acid was used as a standard.[19, 20, 21,22.]

DIPHENYL 2-PICRYL HYDRAZYL (DPPH) ASSAY:

Procedure:

The reaction mixture contained methanol-50ml. DPPH (diphenyl 2- picryl hydrazyl radical) 0.3mM. 1ml of 0.3mM DPPH in methanol was added to 100µl of compound with concentrations ranging from 20µg to 100µg. DPPH solution with methanol was used as a positive control and methanol alone acted as a blank. When DPPH reacts with antioxidants in the sample, it was reduced and the colour changed from deep violet to light yellow. This was measured at 517nm. Ascorbic acid was used as a standard.[23,24,25].

RESULTS AND DISCUSSION:

Antioxidant activity of Ethanolic Extract of *Hibiscus vitifolius* (EEHv) :

***Invitro* evaluation:**

a) 2,2′-azino-bis-3-ethyl benzthiazoline-6-sulphonic acid (ABTS) assay :

The *invitro* antioxidant activity of EEHv was determined by ABTS assay. Results are presented in **Table.1&2.** The investigations indicate that the ethanolic extract of *Hibiscus vitifolius* leaves were found to have significant effect on free radicals which was well comparable with

standard drug Ascorbic acid. It was found to exert a beneficial action against superoxides generated by ABTS assay method with an IC_{50} of 18.64 while Ascorbic acid showed IC_{50} of 10.96.

S.No	Concentration (µg/ml)	Absorbance (734nm)	
		EEHv	Standard
1.	20.000	0.6125	0.7019
2.	40.000	0.6721	0.8476
3.	60.000	0.6953	0.8942
4.	80.000	0.7156	0.9062
5.	100.000	0.7299	0.9521

Table 1: Effect of Absorbance on EEHv by ABTS assay

S.No	Concentration (µg/ml)	% of Activity	
		EEHv	Standard
1.	20.000	49.71491	42.28674
2.	40.000	45.37677	30.62763
3.	60.000	43.21595	27.08674
4.	80.000	41.40426	25.65718
5.	100.000	40.55478	21.95409
		IC_{50}=18.64	IC_{50}=10.96

Table 2: Effect of Antioxidant % of activity on EEHv by ABTS assay

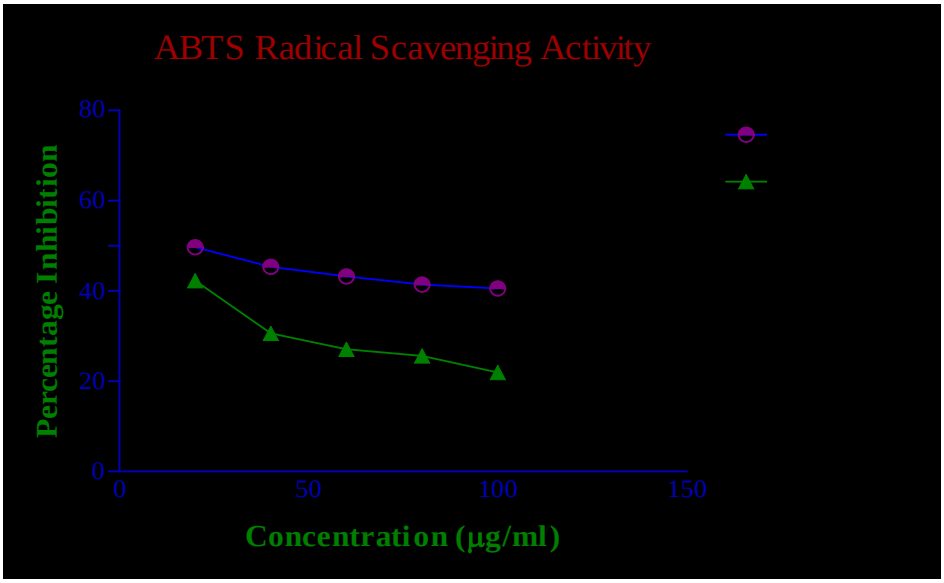

Fig 1: Antioxidant activity of EEHv by ABTS assay

b) Diphenyl 2-picryl hydrazyl (DPPH) assay:

Effect of antioxidant activity of EEHv was determined by DPPH assay.s Results are presented in **Table 3 & 4.** The investigations indicate that the ethanolic extract of leaves of *Hibiscus vitifolius* provides significant cytoprotective effect by exhibiting protection against peroxidative changes by imparting cellular membrane stability and involves inhibition of free radical production along with enhancement of the body defense system.

S.No	Concentration (µg/ml)	Absorbance (517nm)	
		EEHv	Standard
1.	20.000	0.1443	0.213
2.	40.000	0.2333	0.3131
3.	60.000	0.3312	0.382
4.	80.000	0.4376	0.4221
5.	100.000	0.4629	0.441

Table3: Effect of Absorbance on EEHv by DPPH assay

S.No	Concentration (µg/ml)	% of Activity	
		EEHv	Standard
1.	20.000	77.57777	66.69202
2.	40.000	64.58459	51.940
3.	60.000	48.59663	40.54719
4.	80.000	31.47309	33.92912
5.	100.000	28.09803	31.56816
		IC_{50}=53.74	IC_{50}=41.74

Table 4: Effect of Antioxidant % of activity on EEHv by DPPH assay

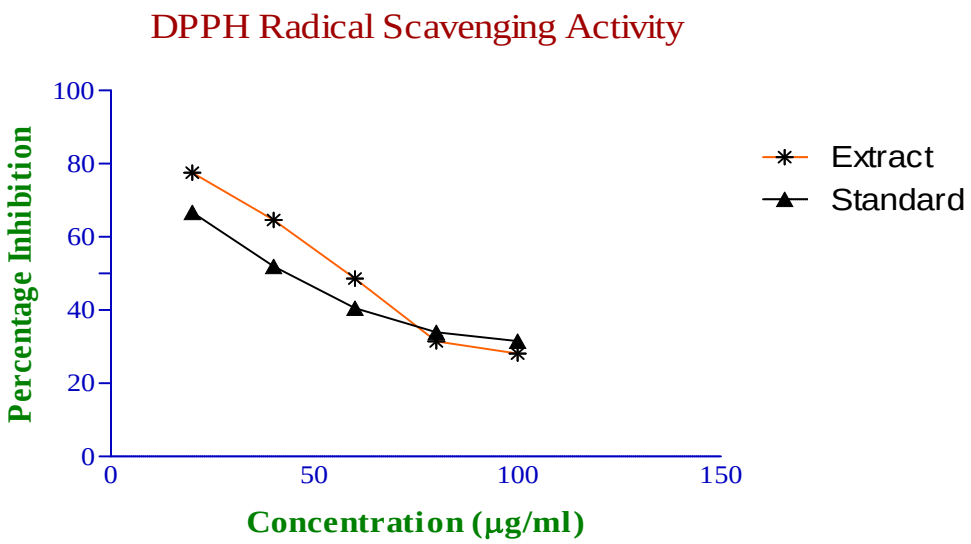

Fig 2: Antioxidant activity of EEHv by DPPH assay

CONCLUSION:

On the basis of result obtained, it can be conclude that the ethanolic extract of aerial parts of *Hibiscus vitifolius* Linn has showed better results in anti oxidant activity when compared with the standard drugs. The findings suggest that *H.vitifolius* could be a potential source of natural anti oxidant that could have a greater importance as therapeutic agent.

ACKNOWLEDGEMENT :

Authors are thankful for the management of Sir C R Reddy College of pharmaceutical sciences, Eluru for providing necessary facilities to carry out the work.

REFERENCES :

1. Kanwar JR, Kanwar RK, Burrow H, et al. "Recent advances on the roles of NO in cancer and chronic inflammatory disorders". 2009;16 https://www.ncbi.nlm.nih.gov/guide/ 16:2373–2394.
2. Chiavaroli V, Giannini C, et al. "Unbalanced oxidant-antioxidant status and its effects in pediatric diseases".2011;16 https://www.ncbi.nlm.nih.gov/guide/ 16:101–107.
3. Yamagishi S, Matsui T. "Nitric oxide, a Janus-faced therapeutic target for diabetic microangiopathy-Friendorfoe? ". 2011;64 https://www.ncbi.nlm.nih.gov/guide/:187–194.
4. Wu YY, Li W, Xu Y, et al . "Evaluation of the antioxidant effects of four main theaflavin derivatives through chemiluminescence and DNA damage analysis". J Zhejiang Univ Sci B. 2011;12:744–751.
5. Coe FL, Evan A, Worcester E. " Kidney stone disease." Curr. Journal of Clinical investigation 2005; 115: 2598-2608.
6. Bahuguna YM, Rawat MS.M, Juyal V, Gnanarajan G. Antilithiatic effect of grains of *eleusine coracana*. Saudi Pharmaceutical Journal 2009; 17:182-8.
7. Moe O W. Kidney stones: pathophysiology and medical management. Lancet 2006; 2:180-84.
8. Abbagani S, Gundimeda S D, Varre S, Ponnala D, Mundluru H P. kidney stone disease: etiology and evaluation. International Journal of Applied Biology and Pharmaceutical Technology 2010; 1:175-82.
9. Herfindal, Gourley. Text book of therapeutic drug and disease management. 7th ed. London: Charcil Livingstone; 2000: 425-36.
10. Urinary Tract Disorders: [online]. [cited 2010 Nov 15]; Available from URL: http://www.mgh.org/education/health/urinary.pdf
11. Data base of Extraction [online] ,http://en.wikipedia.org./Extraction.
12. Database of extraction of Hibiscus vitifolius [online] http://en.wikipedia.org/wiki/Hibiscus.
13. Arthur C Guyton. Text book of medical physiology. 10th ed. Singapore: Harcourt publishers international company; 2000; 264-379.
14. Tortora GJ, Derrickson B. principles of anatomy and physiology. 12th ed. The Urinary system. 26: USA; John Wiley & Sons, Inc 2009; 1018-1061.
15. Database of cancer.org . http://www. Cancer. gov.

16. Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cation Decolorization assay. Free Radical Biology and Medicine. 1999; 26: 1231-1237.
17. Elizabeth K, Rao MNA. Oxygen radical scavenging activity of *Curcumin*. International Journal Pharmaceutics. 1990; 58: 237-240.
18. OECD/OCDE. OECD Guideline no. 423 for Testing of chemicals using acute toxic class method. 2001; 1-14.
19. Pooja C Ochani, Priscilla D Mello. Antioxidant and antihyperlipidemic activity of *Hibiscus sabdariffa* Linn. leaves and calyces extracts in rats. Indian Journal of Experimental Biology. 2009; 47: 276- 282.
20. J. Anbu Jeba Sunilson, P.Jayaraj, et al. "Antioxidant and hepatoprotective effects of the roots of *Hibiscus esculentus* Linn".curr. International Journal of Green Pharmacy. 2008; 200-203.
21. Thabrew M.I, Hughes, R.D., McFarlane, et al. "Antioxidant activity of Osbeckia aspera". Phytotherapy Research, 1998; 12; 288–290.
22. Wangensteen, H., Samuelsen, A.B., Malterud, K.E., 2004. Antioxidant activity from Coriander. Food Chemistry, 88; 293–297.
23. Langley-Evans, S.C., 2000. Antioxidant potential of green and black tea determined using the ferric reducing power (FRAP) assay. International Journal of Food Science and Nutrition, 51; 181–188.
24. Anbu Jeba Sunilson J, Syam mohan , Dinesh kumar chellappan. Hibiscus vitifolius (Linn) root extracts shows potent protective action against anti-tubercular drug induced hepatotoxicity. Journal of Ethnopharmocology, 2012; 141: 396-402.
25. Jayaprakash GK, Jaganmohan Rao L, Sakariah KK. Antioxidant activities of flavidin in different in vitro model systems. Bioorg.Med. Chem, 2004; 12: 5141-5146.