INTERNATIONAL JOURNAL OF PHARMACEUTICAL, CHEMICAL AND BIOLOGICAL SCIENCES

Available online at www.ijpcbs.com

Research Article

EVALUATION OF ANTIOXIDANT ACTIVITY OF ETHANOLIC LEAF EXTRACT OF *FICUS RACEMOSA*

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ABSTRACT

The study was aimed to evaluate the antioxidant activity of ethanolic leaf extract of *Ficus racemosa Linn* belongs to the family Moraceae. In the presents study antioxidant potential of extract was carried out by free radical scavenging activity using 2, 2 diphenyl 1-picryl hydrazyl (DPPH), reducing power assay and phosphomolybdenum assay. Gallic acid was used as a reference standard. The ethanolic leaf extract of *Ficus racemosa* exhibited significant antioxidant activity in all three established methods. Results obtained in the present study reveal ethanolic leaf extract of *Ficus racemosa* possess significant antioxidant activity

Keywords: *Ficus racemosa Linn*, DPPH, Reducing power assay, phosphomolybdenum assay.

INTRODUCTION

The role of oxygen derived free radicals in pathogenesis of number of degenerative disease is well known. Many plants contain phytoconstituents like carotenoids ,flavonoids, phenolics which can be used for scavenging free radicals in the humanbody[1] .Role of free radicals and reactive oxygen species in the pathogenesis of human diseases like cancer aging, liver disease, respiratory diseases[2] and kidney diseases has been widely recognized *Ficus racemosa Linn* is commonly known as cluster fig and udumbara (Sanskrit) belongs to the family Moraceae, is an evergreen, moderate sized, deciduous tree and used as herbal medicine from ancient times. Its leaves are dark green, ovate and having traditional medicinal importance[3]. They are used as mouth washes and used in dysentery, diarrohea, ulcers and menorrhagia[5], effective remedy in glandular swelling, chronic wounds, cervical adenitis haemoptysis[4, 5], and it possess astringent activity.

MATERIALS AND METHODS

The plant was collected from the local area of Pithapuram East Godavari district in month of December- January and the plant was identified by Dr.T.V.Raghava rao, taxonomist.

PREPARATION OF EXTRACT

The leaves were dried under shade and coarsely powdered and macerated for in 95%ethanol. The extract so obtained was concentrated.

ANTIOXIDANT ACTIVITY

DPPH radical scavenging activit[7]

DPPH scavenging activity was measured by the spectrophotometric method. The free radical scavenging activity was followed by preparing 0.002%DPPH solution in methanol. Gallic acid was taken as the reference standard. Different concentration of extract [50,100,200µg/ml] and standard drug[1,2.5,5µg/ml]were prepared using methanol.1ml of 0.002%DPPH solution was mixed with 1ml of all the concentration of extract and standard separately. These mixtures were kept in dark about 30 min and measured the absorbance at 517nm.

DPPH SCAVENGED(%)=$[(A_0-A_1)/A_0]\times100$

A_0 is the absorbance of control and $A_1$is absorbance of test. Antioxidant activity of ethanolic leaf extract of *Ficus racemosa* expressed as IC_{50} values and compared with standard. The IC_{50} values were defined as concentrations (in ig/ml) of extract that scavenges the DPPH radicals by 50%.

REDUCING POWER METHOD[8]

Different concentrations of extract of *Ficus racemosa* (50,100,200µg/ml) in distilled water were mixed with phosphate buffer (0.2M,PH 6.6) and potassium ferricyanide (1%) and incubated at 50^0c for 20 min . A portion (2.5 ml) of trichloroacetic acid (10%) was added and centrifuged at 300 rpm for 10 min. The upper layer of the solution (2.5ml) was mixed with freshly prepared ferric chloride (0.1%) and the absorbance was measured at 700nm. Increased absorbance of the reaction mixture indicates the reducing power.

PHOSPHOMOLYBDENUM ASSAY METHOD[9]

The assay was based on the reduction of MO (VI)-MO (V) by the extract and subsequent formation of a Mo (V) complex (green color) at acidic PH, 0.3ml of extract (50,100,200µg/ml) were combined with 3ml of reagent solution [0.6M] sulphuric acid, 28mM sodium phosphate and 4mM ammonium molybdate] and incubated at 95^0c for 90 min. then the absorbance of solutions were measured at 695nm using spectrophotometer against blank.

RESULTS AND DISCUSSION

DPPH RADICAL SCAVENGING ASSAY

The antioxidant activity of *Ficus racemosa* extract was calculated according to the percentage inhibition in DPPH free radical scavenging assay. The assay is known to give reliable information concerning the antioxidant ability of the tested compounds. A freshly prepared DPPH solution exhibits a deep blue color in the medium of methanol. Antioxidant molecules can quench DPPH free radicals (i.eby providing hydrogen atoms or by electron donation, via a free radical attack on the DPPH molecule) and convert them to colorless (i.e2, 2-di phenyl-1-hydrazine or a substituted analogous hydrazine), result in a decrease in absorbance at 517nm. Table-1(fig: 1a, 1b) shows the % inhibition of DPPH radical scavenged by Gallic acid and ethanolic leaf extract of *Ficus racemosa* at various concentrations (µg/ml). The IC_{50} values of Gallic acid and extract were found to be 0.113 and 12.45 µg/ml respectively.

REDUCING POWER ASSAY

Measurement of reducing power gives antioxidant activity in the extract. In this assay the yellow color of the test solution changes to various shades of green and blue depends upon reducing power of each compound. The reducing power of the extract increased with increase in concentration of ethanolic leaf extract of *Ficus racemosa* and the significant increasing absorbance was found to be 0.4240± 0.0004 at 200µg/ml.Table -1 (fig: 2a,2b).

PHOSPHOMOLYBDENUM ASSAY

Phosphomolybdenum assay based on the reduction of MO (VI)-MO (V) by the extract and formation of a MO (V) complex at acidic pH. Increase in Absorbance was observed by standard and extract .The significant increase in absorbance of extract was found to be 0.08385± 0.0001 at 200 µ g/ml. Table 1(fig3: a, b).

Table 1: Effect of ethanolic leaf extract of *Ficus racemosa*

Name of the Drug	Concentration (µg/ml)	DPPH radical scavenging activity	Reducing Power Assay	Phosphomolybdenum Reduction Assay
		% inhibition ±SEM	Absorbance± SEM	Absorbance±SEM
Standard drug Gallic acid	1	70.25±0.36	0.158±0.0001	0.0183±0.0001
	2.5	75.0±1.04	0.1709±0.00003	0.0282±0.06
	5	82.96±0.09	0.1902±0.0005	0.0396±0.09
IC_{50} (µg/ml)		0.113		
Test drug *Ficus racemosa* (leaf extract)	50	82.39±0.23	0.1957±0.0008	0.01075±0.0001
	100	88.88±0.32	0.29975±0.003	0.02475±0.0002
	200	96.95±0.04	0.4240±0.0004	08385±0.0001
IC_{50} (µg/ml)		12.45		

Values are expressed as Mean ±SEM

DPPH free radicals scavenging activity

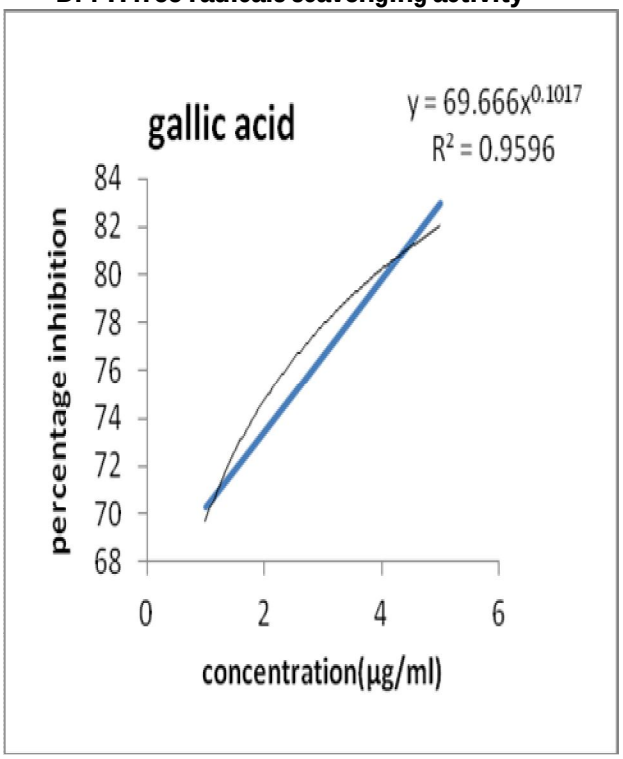

Fig. 1a

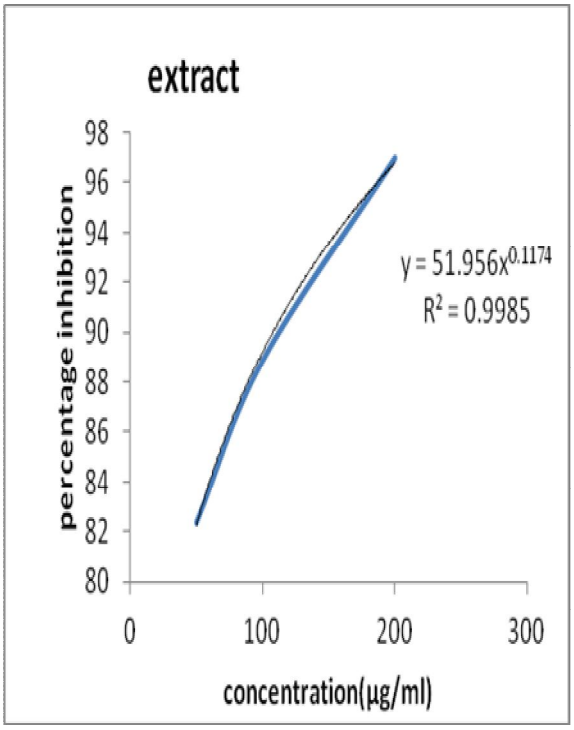

Fig. 1b

Reducing Power Assay

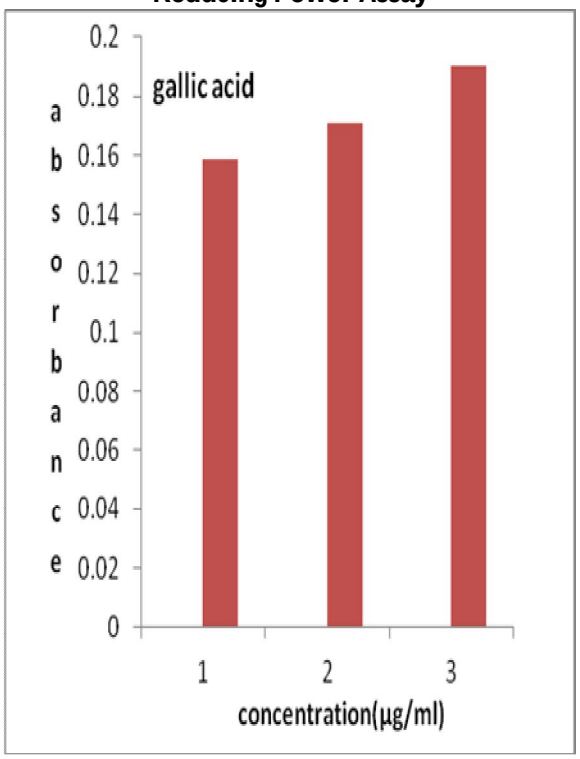

Fig. 2a

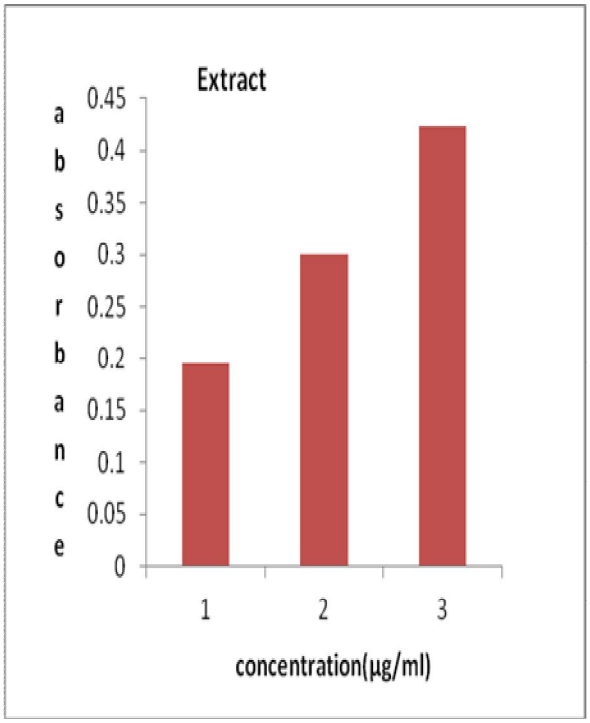

fig. 2b

Phosphomolybdenum Assay Method

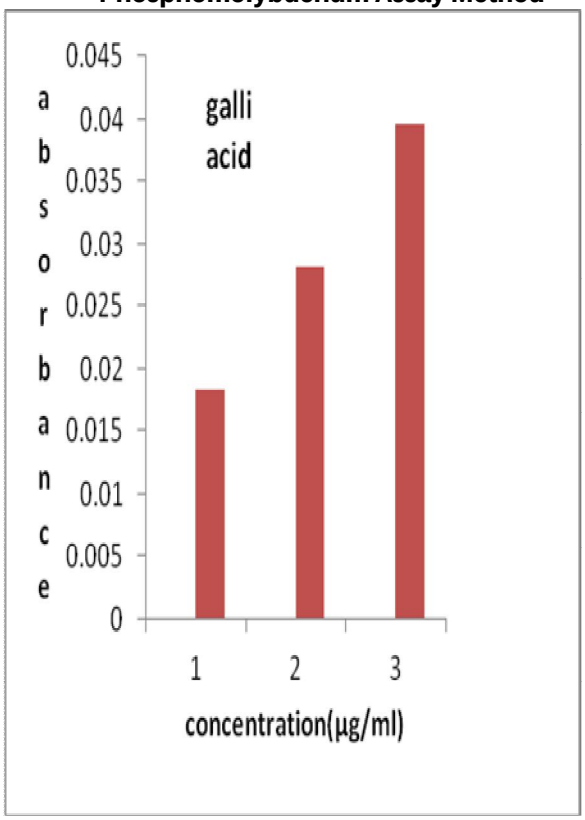

Fig. 3a

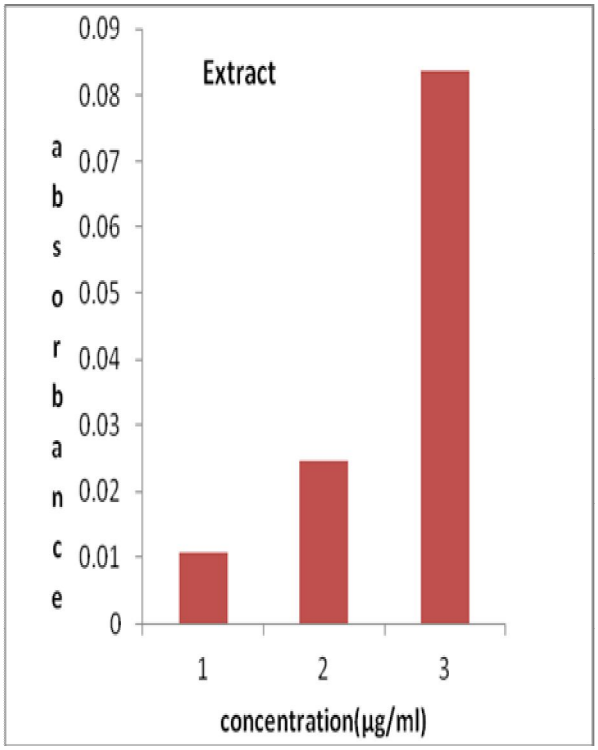

Fig. 3b

CONCLUSION

The results of the present research investigation suggests that the ethanolic leaf extract of *Ficus racemosa* posses good significant antioxidant potential in all three established methods .This activity may be due to its various important phytoconstituents that are present in the plant. Further research can be taken up to isolate the novel molecules present in the plant and biological evaluation of these novel compounds to taken up.

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