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Der Pharma Chemica, 2015, 7(8):21-24
(http://derpharmachemica.com/archive.html)

ISSN 0975-413X
CODEN (USA): PCHHAX

In vitro evaluation of anti-oxidant activity of different extracts of *Justicia gendarussa* leaf

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ABSTRACT

The plant Justicia gendarussa was investigated for their antioxidant activity. The extractions were subjected to assay by Reducing power, Nitric oxide scavenging activity, DPPH methods for evaluation of anti oxidant activity. Among these Petroleum ether extract of Justicia gendarussa revealed strongest antioxidant activity. The results primarily suggest the presence of potent oxidant inhibitory principles in the leaf of Justicia gendarussa.

Key words: Reducing power, Anti oxidant, free radical scavenger & DPPH Method.

INTRODUCTION

There has been a worldwide positive move towards the use of traditional medicines due to the concern over the more invasive, expensive and potentially toxic main stream modern practices. Its popularity is due to desire more for more personalized health care and greater public access to health information [1]. Free radicals play a crucial role in the development of tissue damage in various human diseases like cancer, aging, neuro degenerative diseases, atherosclerosis and pathological events in living organisms. Anti oxidants may an important role in the prevention of these diseases.[2-3] There is an increasing interest in the antioxidant effects of compounds derived from plants, which could relevant in relation to their nutritional incidence and their role in health and disease. A number of reports on the isolation and testing of plant derived antioxidants have been described during the past. We studied different extracts of leaf of *Justicia gendarussa* were investigated for their antioxidant activity by reducing power, Nitric oxide scavenging activity, DPPH Method.

MATERIALS AND METHODS

Plant Material

The plant specimens for the proposed study were collected from ken to select healthy plants and for normal organs. The required samples of different organs were cut and removed from the plant and fixed in FAA (Farmalin-5ml + Acetic acid-5ml + 70% Ethyl alcohol-90ml). After 24 hrs of afixin Medicinal plant garden, Anatomy Research Centre, Chidambaram, Chennai**.** Care was tag, the specimens were dehydrated with graded series of tertiary Butyl alcohol as per the schedule given by Sass, 1940. Infiltration of the specimens was carried by gradual additional of paraffin wax (melting point 58-60 C) until TBA solution attained super saturation. The specimens were cast into paraffin blocks.

Extraction:[4-7] Ten grams of each plant fine powder of indigenous plants weighed into a 250 ml conical flask and 100 ml of methanol and ethanol was added separately for each plant powder then on a rotary shaker at 190 – 220

rpm for 24 hours. This was filtered with whatman No1. filter paper, the residue discarded, and the filter were evaporated to dryness in a water bath temperature at 80 ^{0}C.

Preparation of stock solution

Stock solution was prepared by weighing 10 mg of each dried solvent extract dissolved in 1 ml of dimethyl sulphoxide (DMSO) giving a final concentration of 10,000 µg/ml. The stock solution was kept in screw capped bottles for further analysis [8-10]

Anti oxidant activity:[11-13] The total antioxidant activity of the extract was evaluated by the Nitric oxide scavenging activity, Reducing power & DPPH Method .

NITRIC OXIDE SCAVENGING ACTIVITY

PROCEDURE:

The nitric oxide scavenging activity of *Justicia gendarussa* was determined according to the method. Aqueous solution of sodium nitro prusside spontaneously generates nitric oxide (NO) at physiological pH, which interacts with oxygen to produce nitrate ions and which was measured colorimetrically. 3ml of reaction mixture containing 2ml sodium nitro prusside, 10mM in phosphate buffered saline (PBS) and 1ml various concentrations of the extracts were incubated at 37^{0}C for 4 hours. Control without test compound was kept in an identical manner. After incubation 0.5mL of Griess reagent was added. The absorbance of the chromophore formed was read at 546nm. The percentage inhibition of nitric oxide generation was measured by comparing the absorbance values of control and those of test compounds. Curcumin (50,100,200µg) was used as standard.

REDUCINGPOWER

PROCEDURE:

The reducing power of *Justicia gendarussa* was determined according to the method. Extracts of different concentrations were prepared in 1ml of DMSO and mixed with 2.5ml of phosphate buffer (pH 6.6, 0.2M) and potassium ferricyanide (2.5ml, 10%). The mixture was incubated at 50^{0}c for 20 minutes. Aliquots of TCA (2.5ml, 10%) were added to the mixture, which was then centrifuged at 1500 rpm for 10 minutes. The upper layer of reaction mixture was mixed with distilled water (2.5ml) and freshly prepared $FeCl_3$ solution (0.5ml, 0.1%). The absorbance was measured at 640nm. Increase in absorbance of the reaction mixture indicates the increase in reducing power. Reducing power is given in terms of Ascorbic acid equivalent (As Emg^{-1}).

DPPH METHOD

PROCEDURE:

DPPH scavenging activity was measured by the spectrophotometric method. A stock solution of 25mg of DPPH (150µM) was prepared in 100ml of ethanol, 0.1ml of extract of different concentrations was dissolved in DMSO and 1.9ml of DPPH was added. 0.1ml of DMSO was added to 1.9ml of DPPH in the case of control and 0.1ml of DMSO was added to 1.9ml of ethanol in the case of blank. The reaction was allowed to be completed in the dark for about 20 minutes. Then the absorbance of test mixtures was read at 517nm. The percentage inhibition was calculated and expressed as percent scavenging of DPPH radical. Curcumin (50,100,200µg) was used as standard.

RESULTS AND DISCUSSION

The extracts of *Justicia gendarussa* were assed for anti oxidant activity for petroleum ether extract and ethanolic extracts equivalent with ascorbic acid and results are presented in Table-2 and Table-3 respectively. The anti oxidants act either by scavenging various types of free radicals derived from oxidative processes, by preventing free radical formation through reduction precursors or by chelating agents .In this study the extracts significantly posses anti oxidant activity. [14-19]

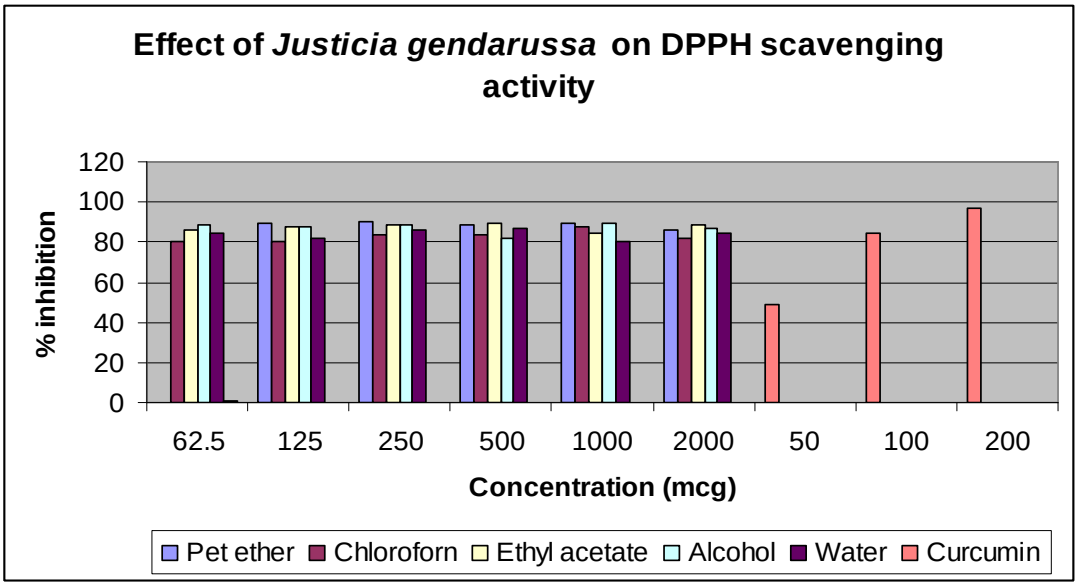
Determination of Reducing power and Total anti-oxidant activity by *In vitro* method.

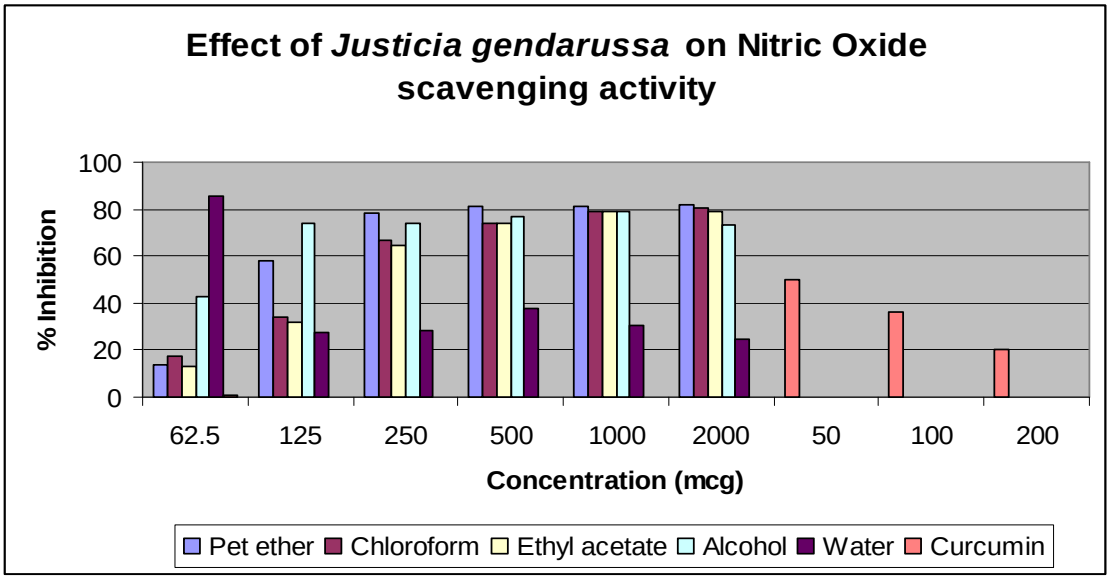
Table -1

Extracts (2000µg)	Reducing power (mg)	Total Anti-oxidant (mg)
Pet ether	0.09	6.13
Chloroform	0.3	3.03
Ethyl acetate	0.07	5.26
Alcohol	0.6	4.88
Water	0.2	3.96

Table-2 *In vitro* Anti-oxidant activity of *Justicia gendarussa* leaves

Extracts	Concentration (µg)	DPPH Method (%Inhibition)	Nitric Oxide (%Inhibition)
Petroleum Ether	62.5	87.3	14.07
	125	89.46	57.69
	250	89.9	78.16
	500	88.5	81.43
	1000	89.62	80.94
	2000	86.2	81.79
Chloroform	62.5	80.0	17.12
	125	80.2	34.19
	250	83.5	66.6
	500	83.5	74.12
	1000	87.46	78.8
	2000	82.29	80.40
Ethyl acetate	62.5	86.17	13.24
	125	88.12	32.11
	250	88.22	64.16
	500	89.41	74.07
	1000	84.51	79.22
	2000	88.67	79.03
Alcohol	62.5	88.70	42.89
	125	87.8	73.99
	250	88.7	73.67
	500	81.60	77.16
	1000	89.7	79.11
	2000	86.96	73.17
Water	62.5	84.25	85.30
	125	81.67	27.47
	250	86.09	28.20
	500	86.92	37.98
	1000	80.50	30.66
	2000	84.59	24.47
Curcumin	50	49.2	50.0
	100	84.7	35.9
	200	97.1	20.5





CONCLUSION

In conclusion, petroleum extract of *Justicia gendarussa* showed maximum antioxidant property. These revelations are significantly noticeable as numerous metabolic disorders and functional defects might be attributed to its antioxidant potential. However, scientific confirmation of traditional claims is necessary for exploiting the therapeutic benefits of this wonder herb.

REFERENCES

[1] M. Shah, *Bangladesh Pharmaceutical Journal*, **2010**, 13(1), 68-70.
[2] NS. Gill, *Journal of Pharmacology Toxicology*, **2011**, 6, 82-89.
[3] SA. Adefegha , *African Journal of Biotechnology*, **2011**,10(4), 632-639.
[4] T. Samatha, *Asian Journal of Pharma and Clinical Research*, **2012**, 5(4), 177-179.
[5] AB. Aliyu, *African Journal of Biotechnology*, **2010**, 9(16), 2437-2441.
[6] RP. Vinay, *Ayuvervedic and Biological Research*, **2010**, 4(1), 23-26.
[7] C. Stanly, *Journal of Medicinal Plant Research*, **2011**, 5(11), 2210-2217.
[8] AAL.Ordonez, *Food Chemical Toxicology*, **2006**, 97, 452-458.
[9] CY. Hsu, *Biological Research*, **2007**, 40, 13-21.
[10] RJ. Ruch, *Carcinogenesis*, **1989**, 10, 1003-1008.
[11] Y. Hsin-Ling, *Food Chemical Toxicology*, **2006**, 44, 1513-1521.
[12] Kumar, *New Botanist*. V (26), **1999**, 41-46.
[13] Owen, *Pharmaceutical Biology*; **2002**; 40 (5); 346 – 357.
[14] Prajapathi, *Indian Pharmaceutical Biology*, **2003**; 41 (3), 166-170.
[15] An unusual secolignan glycoside from *Justicia purpurea. Journal of Natural Products*; **2003**; 66 (8); 113-115.
[16] Leite, *Rev – Bras Farm*. **1993**; 74, (Jan – Mar); 12-15.
[17] Nadkarni, K M., *Indian Materia Medica*, **1927**, Vol (1), pg : 572
[18] Peraza *Pharmaceutical Biology*; **2005**; 43 (7); 594-598.
[19] Perez,.*Chemical and Pharmaceutical Bulletin*; **2004**; 52 (1); 130-131.