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Der Pharma Chemica, 2013, 5(6):246-249
(http://derpharmachemica.com/archive.html)

ISSN 0975-413X
CODEN (USA): PCHHAX

In vitro evaluation of anti oxidant activity of methanolic and ethanolic leaf extracts of five indigenous plants in south India

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ABSTRACT

Five indigenous plants, Piper betle, Punica granatum, Psidium guajava, Gloriosa superb, and Mangifera indica were investigated for their antioxidant activity. The extractions were subjected to assay by Phospho-molybdenum antioxidant assay method for evaluation of anti oxidant activity. The IC_{50} values are ranging from 20-60 µg/ml. Among the indigenous medicinal plants methanol extracts Psidium guajava L. revealed strongest antioxidant activity than vit C. The results primarily suggest the presence of potent oxidant inhibitory principles in the leaf of Psidium guajava.

Key words: Phospho-molybdenum antioxidant assay, anti oxidant, free radical scavenger, vitamin C.

INTRODUCTION

There has been a world wide 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 methanolic and ethanolic leaf extracts of *Piper betle, Punica granatum, Psidium guajava, Gloriosa superb,* and *Mangifera indica* were investigated for their antioxidant activity by Phospho-molybdenum antioxidant assay method.

MATERIALS AND METHODS

Plant Materials: *Piper betle* L. (Betel leaf) - leaf *Punica granatum* L. (Pomegranate) – leaf**,** *Psidium guajava* L. (Guava) - leaf, *Gloriosa superb, L.* (glorylilly) – leaf and *Mangifera indica, L.* (mango), - leaf. The leafs of selected indigenous plants were collected from Prakasam district of Andhra Pradesh, South India. And they were identified and authenticated by K.Babu Rao, Incharge Scientist, Sai Lara Biotechnology, Hyderabad, Andhra Pradesh, India.

The leaves of selected indigenous plants were separated and washed with sterile distilled water and dried using laminar air flow, ground into fine powder using a blender and stored in air tight container till further analysis.

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^{o}C.

Preparation of stock solution for each extract of leaves selected indigenous plants powder
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 phospho-molybdenum assay method (Prieto, *et al.*, 1999). It is based on the reduction of Mo (VI) to Mo (V) by the extract and subsequent formation of a green phosphate-Mo (V) complex in acetic condition. 0.3ml (75mg) of extract was combined with 3ml of reagent solution (0.6M sulfuric acid, 28mM sodium phosphate, and 4mM ammonium molybdate). The reaction mixture was incubated at 95°C for 90min. Then, the absorbance of the solution was measured at 695nm using a UV-visible spectrophotometer against a blank (0.3ml DMSO + 3ml reagent) after cooling to room temperature. The antioxidant activity was expressed as the number of microgram equivalent of ascorbic acid from liner regression curve y = mx + c (y = ascorbic acid concentration µg/ml; m = slope; x = absorbance of compound; c = intercept) obtained after assessing the antioxidant activity of ascorbic acid at different concentrations. All the analysis was carried out in triplicate and results were expressed as mean ± SD.

RESULTS AND DISCUSSION

The extractives of *Piper betle, Punica granatum, Psidium guajava, Gloriosa super and Mangifera indica* were assed for anti oxidant activity for methanolic and ethanolic extracts equivalent with ascorbic acid and results are presented in Table-2 and Table-3 respectively. The standard vitamin C values were tabulated in table-1.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 all the extracts significantly posses anti oxidant activity. [14-19]

Table-1: Total Antioxidant activity of Vitamin C

Vit 'C' Concentration (µg/ml	Absorbance(mean of 6 replicates)
20	0.103 ± 0.06
40	0.225 ± 0.04
80	0.49 ± 0.48
160	0.942 ± 0.03

Note: Values are means of triplicates ±SD

Table- 2: In vitro Antioxidant activities of Methanol extract of five indigenous plants expressed as ascorbic acid equivalents

µg/ml equivalent of vitamin C				
Extracts	**20 µg/ml**	**40 µg/ml**	**80 µg/ml**	**160 µg/ml**
PB	4.1202 ± 0.25	19.4307 ± 2.23	36.2436 ± 1.25	49.4202 ± 1.67
PG	2.3214 ± 1.25	13.3048 ± 1.0	24.5332 ± 2.25	33.2441 ± 1.55
PGU	19.0230 ± 1.0	53.2126 ± 0.77	97.4212 ± 1.25	139.2861 ± 0.25
GS	2.3142 ± 1.25	11.5332 ± 1.25	19.2122 ± 1.0	37.6782 ± 1.0
MI	17.2221 ± 1.0	28.2653 ± 1.55	45.2121 ± 1.25	89.4132 ± 1.55

Note: Higher value indicates higher reducing power
PB: Piper betle L. PG: Punica granatum L., PGU: Psidium guajava L. GS: Gloriosa superb and MI: Mangifera indica, .

Total Antioxidant activity of Vitamin C standard curve

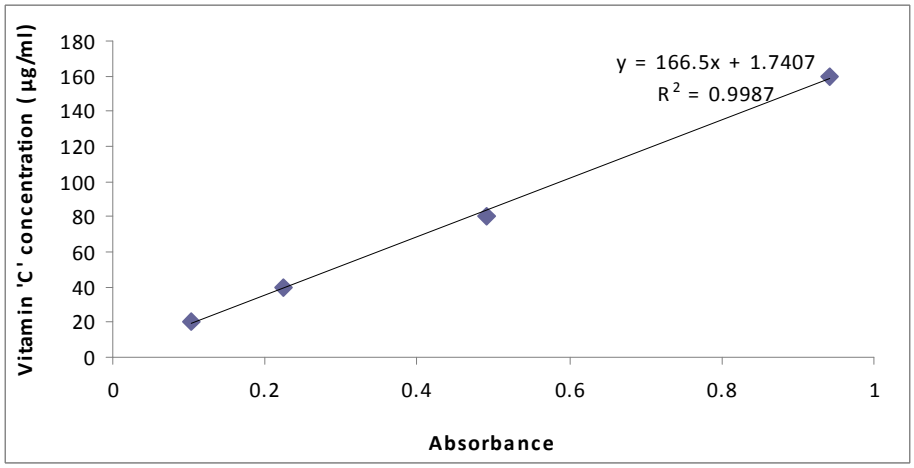

Table-3: In vitro Antioxidant activities of Ethanol extract of five indigenous plants expressed as ascorbic acid equivalents

	µg/ml equivalent of vitamin C			
Extracts	**20 µg/ml**	**40 µg/ml**	**80 µg/ml**	**160 µg/ml**
PB	2.0212 ± 0.5	14.2102 ± 1.25	27.3216 ± 0.25	41.2213 ± 0.25
PG	2.2522 ± 0.25	9.1121 ± 1.25	17.5631 ± 0.5	26.7145 ± 1.15
PGU	12.2115 ± 0.5	41.3123 ± 1.10	78.5213 ± 0.55	101.5315 ± 1.25
GS	1.8212 ± 0.5	7.5724 ± 0.25	12.2231 ± 1.25	22.6514 ± 1.25
MI	9.3115 ± 1.67	19.7223 ± 1.13	31.3233 ± 1.10	52.3352 ± 1.67

Note: Higher values indicates higher reducing power

PB: Piper betle L. PG: Punica granatum L., PGU: Psidium guajava L. GS: Gloriosa superb and MI: Mangifera indica, .

CONCLUSION

In conclusion, in both methanolic and ethanolic extract of *Psidium guajava* showed maximum antioxidant properties in remaining indigious palnt extracts. These revelations are significantly noticeable as *Psidium guajava* induced amelioration of 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. The order of antioxidant activity in Among all the indigenous medicinal plants in both methanolic and ethanolic extracts *Psidium guajava* L. (Guava)- leaf revealed strongest antioxidant potential followed by *Mangifera indica, L.* (mango), - leaf > *Piper betle* L. (Betel leaf) leaf > *Punica granatum* L. (Pomegranate) – leaf > *Gloriosa superb, L.* (glorylilly) – leaf. Antioxidant potential all the plants under study are greater than ascorbic acid.

Acknowledgement

The Author thankful to is guide Dr. B. sitaram. Research adviser, Dept. Of Basic science and Humanity, PVP Siddhartha Institute of Technology, Kanuru, Vijayawada, Andhra Pradesh, India.

REFERENCES

[1] M. Shah, *Bangladesh pharmaceutical journal*, jan-**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] S. M. Newton, *Phytotherapy Research*, **2000**, 14, 303-322.
[13] T. Rabe, *Journal of Ethnopharmacology*, **1997**, 56, 81-87.
[14] F. A. Valter, *Phytotherapy Research*, **2004**, 18, 634-639.
[15] T. Dimo, *Journal of Ethnopharmacology*, **1998**, 60, 179-182.
[16] T. Dimo, *Journal of Ethnopharmacology*, **2001**, 76, 215-221.
[17]C. Abajo, *Journal of Ethnopharmacology*, **2004**, 93, 319-323.
[18] L. P. Yuan, *Journal of Ethnopharmacology*, **2008**, 116, 539-546.
[19] H.Canoa Juan, *Journal of Ethnopharmacology*, **2004**, 90, 293-316.