

In vitro Antioxidant and Antibacterial Activity of Plant Extracts of *Pergularia extensa* Chiov

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ABSTRACT

This present study is undertaken to evaluate the antioxidant and antibacterial activity of various crude extracts of whole plant of *Pergularia extensa* chiov. Methanol and aqueous extract of whole plant of *Pergularia extensa* chiov. were used for this study. The antioxidant potential was determined by 1,1-diphenyl-2-picryl hydrazyl (DPPH) free radical scavenging method. The methanol extract showed significant percentage inhibition in scavenging the DPPH radicals than aqueous extract and standard, ascorbic acid. Also increase in concentration showed increase in percentage of inhibition. For antibacterial activities test, the extract was subjected to its effectiveness against both Gram-positive and Gram-negative bacteria in agar diffusion method. The zones of inhibition produced by the methanol extract against various gram +ive and gram -ive are more significant than aqueous extract and standard, pefloxacin. The obtained results provide a support for the use of this plant in traditional medicine and suggest its further advance investigation.

Key words: *Pergularia extensa*, Antioxidant, Antibacterial.

INTRODUCTION

*Pergularia extensa*¹ commonly called as *Pergularia daemia* or *Daemia extensa* (Fam:- Asclepiadaceae) which is a slender twining perennial herb. It has a foetid smelling laticiferous odour after touch. It has hispid stems, simple, opposite and cordate leaves. Its flowers are greenish or dull white tinged with purple colour. It has corymbose clusters and having axillary long peduncles. Its fruits have reflexed follicles. Its leaves have densely velvety pubescent on both sides. It grows throughout the hotter parts of India upto 900-1000 meters in Himalayas, Ceylon and Afghanistan. It is also medicinally used in Gold coast, Senegambia and Cameroon. The plant is used as a whole in the indigenous systems of medicines. The leaf and fruit of the plant has medicinal properties such as emetic, anthelmintic, expectorant and antipyretic. The decoction of leaves is used urethrorrhea, amenorrhoea, dysmenorrhoea. The root bark is mixed with cow's milk and employed as a purgative²⁻³.

The present investigation was undertaken in study of *invitro* antioxidant and antibacterial activity of polar extracts obtained from the whole plant.

MATERIALS AND METHODS

Plant material

The whole plant including the root of *pergularia extensa* was collected from the herbal garden of KVSR Siddhartha College of Pharmaceutical Sciences, Vijayawada during the month of February 2008. The plant was identified and authenticated by Dr.S.M.Khasim,

Professor, Department of Botany, Acharya Nagarjuna University in Guntur.

Chemicals

All the reagents used in this study were of analytical grade obtained from S.d. Fine Chemicals Ltd., Mumbai.

Experimental procedure

Antioxidant activity

In this model oxidative radicals were generated by using 2, 2-diphenyl-picrylhydrazil (DPPH)⁴. About 0.1 ml of DPPH in acetate buffer was prepared and 1ml of solution was added to 3 ml of solutions of methanol and aqueous extracts of the plant using Acetate buffer at concentration range of 10-320 µg/ml. Ascorbic acid acted as a standard drug and its concentration was also prepared like the test extracts. The mixtures were shaken vigorously and kept under incubation for 30 minutes. Then absorbances of all the mixtures were measured at 517 nm using UV-Vis Spectrophotometer.

DPPH Scavenging effect (%inhibition) = $1 - \frac{A_{517}(\text{sample}) - A_{517}(\text{control})}{A_{517}(\text{control})} \times 100$

Antibacterial activity

Bacterial strains namely *Bacillus subtilis* (NCIM 2063), *Staphylococcus aureus* (NCIM 2019), *Escherichia coli* (NCIM 2065) and *Klebsiella pneumoniae* (NCIM 2036) had been maintained on Mueller Hinton agar (MHA) medium at 37°C. The anti-bacterial activity of all the extracts was performed by paper disc diffusion assay method⁵. The discs of uniform size (6 mm) were prepared from Whatmann No.1 filter paper and were sterilized in hot air oven at 160°C for 1hr. Then the discs were

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Table: 1 Absorbance of methanol extract, aqueous extract and standard (ascorbic acid)

Concentration of each extract (µg/ml)	Absorbance (nm)		
	Methanol extract	Aqueous extract	Ascorbic acid
Control	0.784	0.978	0.342
10	0.664	0.923	0.246
20	0.612	0.876	0.220
100	0.545	0.765	0.178
200	0.428	0.675	0.127
300	0.326	0.628	0.097
320	0.209	0.566	0.056

Table 2: Antibacterial activity of polar extracts of *Pergularia extensa*

Name of the extract (µg/ml)	<i>B. subtilis</i> (NCIM 2063)	<i>S. aureus</i> (NCIM 2019)	<i>E. coli</i> (NCIM 2065)	<i>K. Pneumoniae</i> (NCIM 2036)
Methanol extract				
50	08	14	08	09
100	14	18	13	11
200	19	23	19	16
Aqueous extract				
50	23	12	22	17
100	28	16	27	21
200	31	19	34	26
Pefloxacin 100	29	34	31	26
DMF (control)	5	7	6	5

DMF:- dimethyl formamide

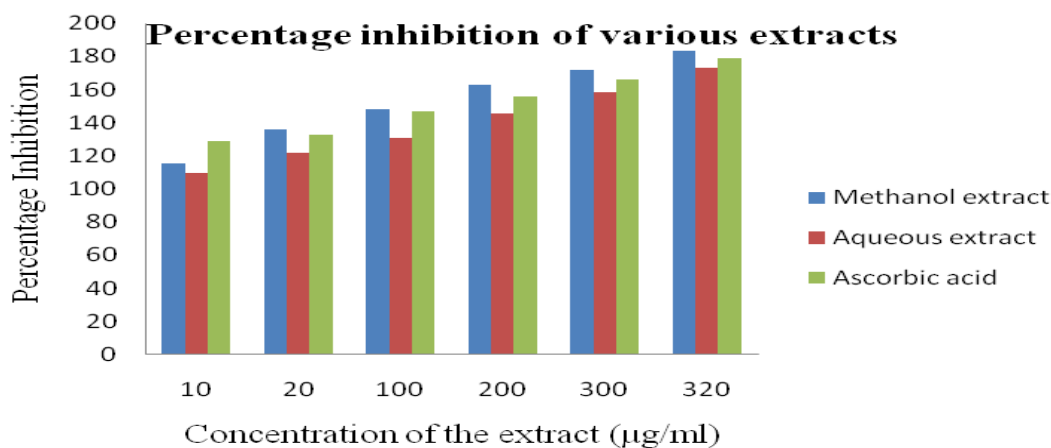


Figure 1: Percentage Inhibition of DPPH radical

impregnated with the MIC (Minimum Inhibitory concentration) of different concentrations (50µg/ml, 100µg/ml and 250µg/ml) of various extracts and standard, Pefloxacin. The solvent DMF is used as a control. The plates were prepared by using MH agar media and the extracts of various dilutions were allowed to solidify and dried. Different impregnated discs in various concentrations of extracts were prepared in the solidified agar plates and were labelled. Then a loopful of bacterial cultures was inoculated with the MH media at the labeled spots. The plates were inoculated at 37°C for 24 hrs and the zone of inhibition was measured.

RESULTS

From the percentage inhibition obtained from the antioxidant activity, it is evident that the methanol extract of *Pergularia extensa* chiov showed significant

antioxidant activity than the standard ascorbic acid (Table 1). Also by increase in concentration of both the extracts and standard showed increase in percentage of inhibition (Fig 1).

From the zone of inhibition (Table 1) obtained from the antibacterial activity, it was confirmed that the methanol extract showed more significant activity against various gram +ive and gram -ive bacterias than aqueous extract and standard, pefloxacin (Fig 2).

DISCUSSIONS

DPPH is the best, easiest and widely used method for testing preliminary free radical scavenging activity of a compound or a plant extract⁶. In present study, methanol extracts of the whole plant of *P.extensa* possess strong antioxidant activity. However the aqueous extract showed mild antioxidant activity. The free radical scavenging

property may be one of the mechanisms by which the drug is effective as a traditional medicine. Most of the antioxidant compounds such as alkaloids, triterpenes, tannins, flavonoids, etc reduce the free radical formation. This plant is also reported for antioxidant compounds⁷⁻⁸ as given above. Also these compounds have the effect in inhibiting the bacterial growth. The antimicrobial activity is probably due to the membrane disruption by terpenes and their activity might be due to their ability to form complex with extra cellular, soluble proteins and bacterial cell walls and disrupt microbial membrane⁹. So, this activity is also may be due to the presence of phenolic compounds (tannins and flavonoids) present in the extract. Further researches are needed to find new clinically effective antibacterial compound from the plant.

CONCLUSION

Hundreds of plant species have been tested for antimicrobial properties, the vast majority has not been adequately evaluated. From the above studies it is concluded that the traditional plants may represent new sources of anti-microbials with stable, biologically active components that can establish a scientific base for the use of plants in modern medicine. Further researchers are needed to find new clinically effective antibacterial compound from the plant.

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