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Evaluation of anti bacterial activity of *Coccinia grandis* leafs and stem extracts

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Abstract

The *Coccinia grandis* is having anti bacterial activity and every part of this plant is valuable in medicine and various preparations in indigenous system of medicine for various skin disease, bronchitis, psoriasis, smallpox, ulcers and it is having the antilithic, hypolipidemic, antimutagenic and hypoglycemic activities. In this study the *in vitro* antibacterial activity of leafs and stem extracts were tested against gram positive and gram negative organisms bacterial strains by agar well disc plate method. The crude extract showed a broad spectrum of antibacterial activity by inhibiting both the gram positive and gram negative groups. The antibacterial activity of *C. grandis* leaf extract using solvents such as ethanol, ethyl acetate was evaluated against The gram positive strains such as *St. aureus*, *B. subtilis* and in gram negative strains such as *E. coli* (ETEC), *P.a*. Minimal inhibitory concentration of the leaf extract against each test organism was also studied by observing their growth on Mueller Hinton Agar containing the extract at various incremental levels, equivalent to 50 µg/ml to 500 µg/ml of the extract. The highest activity was observed in ethanol stem extract against both against gram positive and gram negative organism's bacterial strains with an inhibitory concentration below 50 µg/ml. The significance of the study was conducted to investigate the *in vitro* antibacterial activity of *Coccinia grandis* plant and to evaluate scientific base of their applications.

Keywords: Antibacterial, *Coccinia grandis*, disc plate method, Minimal inhibitory concentration (MIC)

1. Introduction

Coccinia grandis L., of the family Cucurbitaceae is distributed in tropical Asia, Africa and is commonly found in Pakistan, India and Srilanka. Coccinia is a climber and trailer. The fruit of *Coccinia grandis* is used as (a) vegetable when green and eaten fresh when ripened into bright scarlet colour. Every part of this plant is valuable in medicine and various preparations have been mentioned in indigenous system of medicine for various skin diseases, bronchial catarrh, bronchitis and Unani systems of medicine for ringworm, psoriasis, small pox, scabies and other itchy skin eruptions and ulcers. Oil of this plant is used as an injection into chronic sinuses. The plant is used in decoction for gonorrhoeae, diabetes and also useful in dropsical condition, pyelitis, cystitis, strangury, snake bite, urinary gravel and calculi. It is also useful to induce perspiration in fever and cures sores in the tongue. It has antilithic, hypolipidemic, antimutagenic and hypoglycemic activities. The present study was undertaken to evaluate (the) antibacterial activity of *Coccinia grandis* extracts.

2. Materials and Methods

2.1 Collection of plant material

Coccinia grandis, leaves and stem were collected from the fields of Guntur. During the month Nov-Dec 2013 the material was botanically identified and confirmed by Botanist.

2.2 Preparation of crude extracts

Stems and leaves were separated and dried under shade and powdered. The powdered leaves were extracted with ethanol, ethyl acetate and stems extracted with ethanol for three days by using Sox let apparatus at 30 °C. After that the extracts were concentrated with rotary evaporator and dried in vacuum. The partitioning of Ethanolic extract was first done with ethyl acetyl and water. ethyl acetate was evaporated and dried by means of rotary evaporator under reduced pressure. The extracts and fractions were taken for assays.

Phytochemical analysis of *Coccinia grandis* leaves extract:

Compounds	Ethanol extract
Carbohydrates	+
Proteins& amino acids	+
Glycosides	+
Alkaloids	+
Flavonoids	+
Tannins	+
phenols	+
Sterols	-
Tri terpenoids	+
Acidic compounds	-
Saponins	+

3. Anti Bacterial Activity

The anti bacterial activity was carried out by disc diffusion technique. All the bacterial cultures were obtained from the Microbiology department, Chalapathi institute of pharmaceutical sciences, Guntur. Ciprofloxacin is used as

standard and nutrient agar medium was employed as medium. In this classical method, antibiotics diffuse from a confined source through the nutrient agar medium and create a concentration gradient, Dried and sterilized filter paper discs (6mm diameter) containing the test samples (Leaves Ethanol, Ethyl acetate & stem Ethanol extracts which are dissolved in DMSO as a solvent) of known amounts are placed on nutrient agar medium uniformly seeded with the test microorganisms. Standard antibiotic (Ciprofloxacin) discs and blank discs were used as positive and negative control. these plate were then inverted and incubated at 37 °C for 24 hours for optimum growth of the organisms. The test materials having antimicrobial property inhibit microbial growth in the media surrounding the discs and thereby yield a clear, distinct area defined as zone of inhibition. The antimicrobial activity of the test agent was then determined by measuring the diameter of zone of inhibition expressed in millimeter and compared with the control.

4. Results

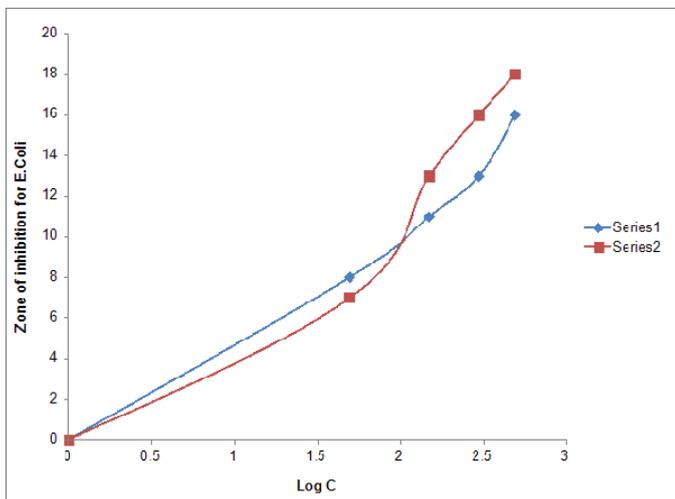
Table 1: Zone of inhibition in mm for Gram Negative Bacteria:

Concentration μ /ml	Ethanol leaf extract		Ethyl acetate Leaf extract		Ethanol stem extract		Ciprofloxacin	
	<i>E. coli</i>	P.a	<i>E. coli</i>	P.a	<i>E. coli</i>	P.a	<i>E. coli</i>	P.a
50mcg	6	7	7	7	8	6	7	9
150mcg	10	11	5	9	11	10	13	11
300mcg	10	12	8	9	13	15	16	14
500mcg	12	15	13	11	16	19	18	16

E. coli :- *Escherichia coli* (ETEC); P.a:-*Pseudomonas aeruginosa*

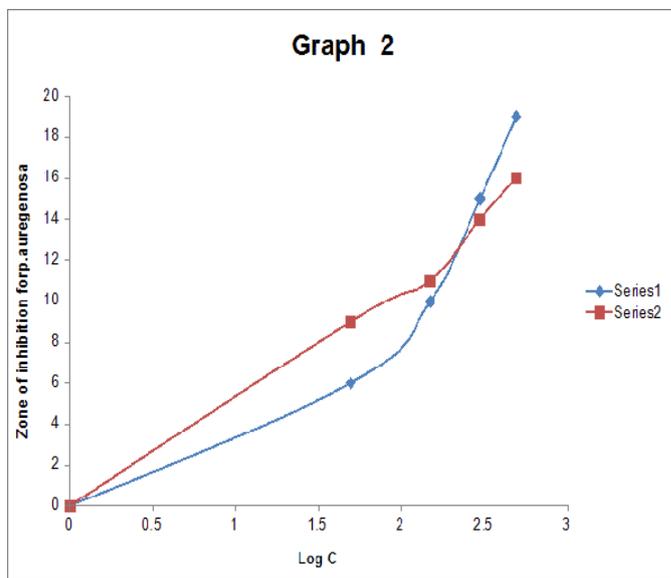
Graphs for Gram negative organisms:

Graph-1



Series- 1 Ethanol stems extract
Series-2 Ciprofloxacin

Graph 2



Series- 1 Ethanol stems extract
Series-2 Ciprofloxacin

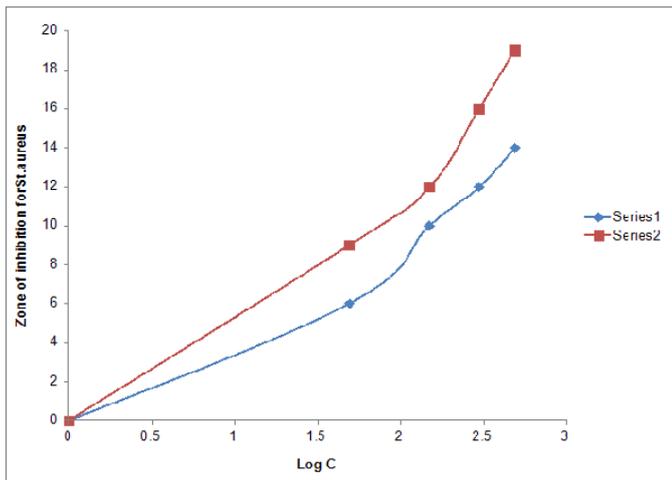
Table 2: Zone of inhibition in mm for Gram positive bacteria:

Concentration μ /ml	Ethanol leaf extract		Ethyl acetate Leaf extract		Ethanol stem extract		Ciprofloxacin	
	<i>St. aureus</i>	<i>B. substillus</i>	<i>St. aureus</i>	<i>B. substillus</i>	<i>St. aureus</i>	<i>B. substillus</i>	<i>St. aureus</i>	<i>B. substillus</i>
50mcg	4	5	5	3	6	4	9	5
150mcg	5	-	6	8	10	7	12	9
300mcg	7	6	9	7	12	9	16	13
500mcg	12	10	11	10	14	11	19	17

St. aureus: Staphylococcus aureus; *B. substillus*:-Bacillus substillus.

Graphs for Gram positive organisms

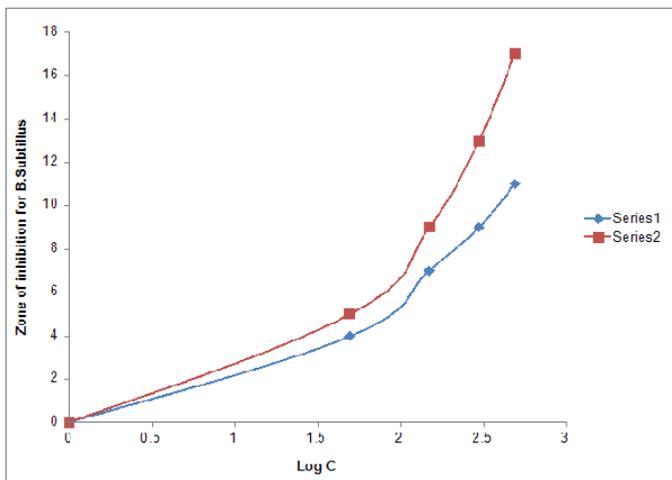
Graph-3



Series- 1 Ethanol stems extract

Series- 2 Ciprofloxacin

Graph-4



Series- 1 Ethanol stems extract

Series- 2 Ciprofloxacin

5. Discussion

The present study was conducted to investigate the *in vitro* antibacterial activity of *Coccinia grandis* plant used by people of India, to evaluate scientific base of their applications. The gram positive strains such as *St. aureus*, *B. subtilis* and in gram negative strains such as *E. coli* (ETEC), *P.a* causing serious infection in human and in other animals including superficial skin lesion, localized abscesses, and food poisoning were in first positions. In this study, the *C. grandis* exerted antibacterial activity against both Gram positive and Gram negative bacteria associated with different type of infections including urinary tract infections Respiratory tract (e.g. pneumonia) Bloodstream (bacteremia) Heart (endocarditis) CNS (e.g. meningitis, brain abscess) Ear (e.g. otitis externa and media) Eye (e.g. bacterial keratitis, endophthalmitis) Bones and joints (e.g. osteomyelitis) GI tract (e.g. diarrhea, enteritis, enterocolitis) Urinary tract Skin (e.g. ecthyma gangrenosum and wound infections). The demonstration of activity against both Gram positive and Gram negative bacteria is an indication of broad spectrum of activity and thus

can be used to source antibiotic substances for drug development that can be used in the control of these bacterial infections. Further investigations of its activity against a wider range of bacteria and fungi, identification and purification of its chemical constituents, and toxicological investigations of the plant extracts should be carried out with a view to developing novel drugs for human consumption. The *Coccinia grandis* plant was used for testing their antibacterial activity and showed high activity against those organisms and these indicate that the herbal preparations could be used for preventing and treat the diseases caused by those selected organisms

6. Conclusion

In the present investigation the antibacterial activities have been observed in case *Escherichia coli*, *Pseudomonas aeruginosa* and *Bacillus subtilis*. *Staphylococcus aureus*; It is evident from the plots that Ethanolic extract possess more predominant antibacterial property in comparison to other extract.

7. Acknowledgement

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8. Reference

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