



ANTI-MICROBIAL ACTIVITY OF *NERIUM OLEANDER* STEM EXTRACT

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Abstract

Nerium oleander) is commonly known as 'kaner' and the plant is toxic. It is used for treatment of heart diseases, respiratory problems, cancer and aids. In the present study we have evaluated the Anti-microbial activity of *Nerium oleander* stem extracts on wistar strain albino rats. All the extracts showed good activity against all the bacteria used. Preliminary chemical studies of the plant reveal the presence of flavanoids which are responsible for the anti microbial activity of the plant. Extracts showed potent anti microbial activity against *Pseudomonas auregnosa* and *B.Subtilis*. Ethanol extract of the plant is more potent than any other extract and is active against all the bacteria tested. None of the extract showed any of the activity against any fungus tested.

Keywords: - *Nerium oleander*, kaner, *Pseudomonas auregnosa*.

Introduction

Nerium oleander L (Apocynaceae) is commonly known as 'kaner' and the flowers are showy, sweet-scented, and double petaled, pink in colour. Despite its well-recognized toxic potential this plant is being used as folklore medicine throughout the world. In ethno botanical literature it is mentioned to be effective in the treatment of **cardiac illnesses, asthma, corns, cancer & epilepsy**[1,2] and also used as **diuretic** .[3] In ancient India it is regarded as *Nighantu ratnakar* which relieves headache and overcomes the ill effect of *Vata* and *Kapha*. Most of the polysaccharides purified from oleander showed anti-tumor and immune-stimulating effects. The whole plant is said to have anticancer properties. Its various parts are reputed as therapeutic agents in the treatment of swellings, leprosy, eye and skin diseases. The plant has been used in the treatment of cardiac illness, asthma, diabetes mellitus, corns, scabies, cancer and epilepsy.[4]

Materials and Methods

Plant material

The *Nerium oleander* (Apocynaceae) stem was collected from the Karnal, India in June 2008. Botanical identification was confirmed by morphological characteristic from Raw Materials Herbarium and Museum, NISCAIR, DELHI) with accession no.

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Niscair/RHMD/Consult/-2008-2009/1080/111. A voucher specimen (no. hcop/pcg/01763/ 08) of same was submitted to Hindu College of Pharmacy, Sonipat, India. Preparation of Extracts

The stem was dried at room temperature under well-ventilated shade by spreading them uniformly. The dried part was powdered, weighed (300gm) and filled in Soxhlet apparatus for successive solvent extraction with different solvents viz. pet-ether, ethanol and water. Before making extraction with next solvent previous one was dried at a temperature below 60°C. Percentage Yield was calculated for each extract after drying it under vacuum.

The microorganisms were availed from M.T.C.C. Institute of Microbial Technology, Sector 39-A, Chandigarh-160036, India. ST No: 02/ST/Sci & Tech/STC/Chd/2002.

Invoice No. MTCC/08/10/5396.

Chemicals

Ciprofloxacin and fluconazole was procured from Ranbaxy research lab., Gurgaon, India. All the chemicals used were of analytical grade and used as received.

Well diffusion method

Test solution of each extract viz. pet. ether (60-80°C), ethanol and aqueous extracts was prepared at a conc. of 25,50,75,100 mg/ml. Ciprofloxacin was taken as standard for antibacterial activity at a conc. 10µg/ml. and fluconazole was taken as standard for fungi at a conc. 10µg/ml. Nutrient agar medium was prepared and sterilized by an autoclaving. In an aseptic room, the medium was poured into sterile petri dishes to a uniform depth and then allowed to cool at room temperature.[7-9]

For bacteria (pour plate method): Before it could solidify the agar medium was mixed with the test organisms (one day old subculture) and allowed to solidify.[10-12]

For fungi (spread plate method): Fungus was spread on the surface of solid agar medium with the help of an L shaped rod by streaking on the solidified agar plates.

Then the wells were made in solidified agar plates with the help of sterile glass borer of size 8 mm and capacity 2 ml in solidified agar in such a way that overlapping of zone of inhibition does not occur. Plates were kept at room temperature for half an hour for diffusion of the sample into agar media. The organism-inoculated petridishes were then incubated as per requirement of micro-organisms. After the completion of incubation period, the zone of inhibition produced by the sample with different organisms in different plates was measured and recorded immediately.[8,10]

Statistical Analysis

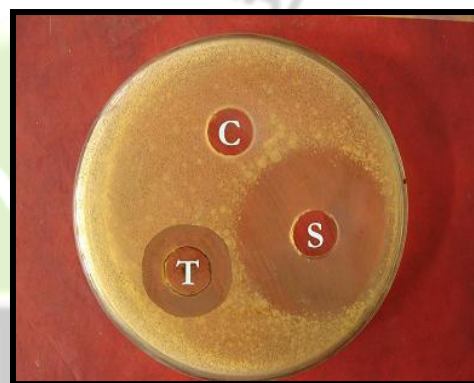
The values of all the above methods are expressed as Mean \pm SEM. Total variation in asset of data was estimated through one way analysis of variance (ANOVA) followed by Dunnett's test. Values of $P < 0.1$ & $P < 0.05$ were considered statistically significant.

Results and Discussion:

Antimicrobial activity of various extracts of stem part of

Nerium oleander was studied by measuring the zone of inhibition formed around the agar well and the results are given in table no. 1. All the extracts showed good activity (Fig 1) against *P. auregenosa* and *B. subtilis*. Ethanol extract has activity against all the four microorganisms tested but have anti bacterial activity against *M. leuteus* only at higher concentration. Aqueous extract showed considerable activity only against *P. auregenosa* and *B. subtilis*. All extracts failed to show any activity against any of the fungi used. Thus the plant shows antimicrobial activity and can be a potent ingredient for herbal products

Fig: 1 antibacterial activity of ethanol extract against *P. auregenosa* (Where T= test, S= standard, C= positive control)



Sr. No.	Microorganism	Zone of Inhibition (mm)													
		Ciprofloxacin (µg/ml)	Fluconazole (µg/ml)	Petroleum ether extract (mg/ml)				Ethanol extract (mg/ml)				Aqueous extract (mg/ml)			
		10	10	25	50	75	100	25	50	75	100	25	50	75	100
1.	<i>S. aureus</i>	23.3 \pm 0.33	-	-	-	-	-	9	14.0 \pm 0.00	16.3 \pm 0.33	18.0 \pm 0.0*	-	-	-	-
2.	<i>B. subtilis</i>	22.6 \pm 0.66	-	-	3 \pm 0.89*	5 \pm 0.00	8.3 \pm 0.33	10	11	12.3 \pm 0.33	14.0 \pm 0.00	12 \pm 0.01	13 \pm 0.078	15 \pm 0.6*	17
3.	<i>M. luteus</i>	20.3 \pm 0.33	-	-	-	-	-	-	-	11.3 \pm 0.33	12.6 \pm 0.33	-	-	-	-
4.	<i>P. aeruginosa</i>	29.3 \pm 0.33	-	8 \pm 1.21	9 \pm 0.00	10.3 \pm 0.33*	11.6 \pm 0.66	5.03 \pm 0.33	8.6 \pm 0.33	11.3 \pm 0.33	12.0 \pm 0.00	14 \pm 0.098	15 \pm 0.06	15 \pm 0.78	15
5.	<i>C. albicans</i>	-	17.3 \pm 0.33	-	-	-	-	-	-	-	-	-	-	-	-
6.	<i>A. niger</i>	-	21.6 \pm 0.33	-	-	-	-	-	-	-	-	-	-	-	-

Values are expressed as mean \pm SEM. * $p < 0.001$ as compared to control group

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