

ANTIBACTERIAL AND ANTIFUNGAL EVALUATION OF THE LEAVES OF *RHINACANTHUS NASTUS* LINN

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ABSTRACT: The *in vitro* antibacterial and antifungal activity of the leaves of *Rhinacanthus nastus* was investigated against various strains of bacteria and fungi viz. *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Proteus vulgaris* and *Candida albicans* by disc diffusion method. The chloroform and ethyl acetate extract in a dose of 20 mgml⁻¹ showed significant activity against *Staphylococcus aureus* and *Candida albicans* when compared with the standard antibiotic ciprofloxacin (10 mcg/disc) and ketoconazole (10 mcg/disc) respectively. Methanolic extract showed significant activity against *Staphylococcus aureus*.

Key words: *Rhinacanthus nastus* Linn, Leaves, antibacterial, antifungal.

INTRODUCTION

Rhinacanthus nastus Linn belongs to Acanthaceae family is a shrub and is well known for its medicinal uses, commonly called as Nagamali, Kaligai, anichi Tamil, Nagamella in Telugu, doddapatika in kannada, jupani in Hindi, yuthikaparni in Sanskrit and Gajakarni in Marathi (1-4). The entire plant is has been used as an anthelmintic, antiseptic, aphrodisiac, antiparasitic and as anticancer agent. It is also used for in the treatment of obesity, leprosy, eczema, scurvy and dhobi's itch. Leaves, roots and seeds act as an antidote for snake bites. In Madagascar, the juice of leaves and the root bark were used in the treatment of *Herpes Circinatus* (6-8). The plant has been reported to contain triterpenoids, steroids, coumarins, anthraquinones, glycosides, carbohydrates, rhinacanthone and rhinacanthin (9-11). Since the plant was used as antiseptic, antiparasitic and in ringworm infestation it was thought worthwhile to investigate the antibacterial and antifungal activity.

EXPERIMENTAL

Collection of the plant material

Fresh leaves of *Rhinacanthus nastus* Linn were collected in the month of October 2008 from Madurai, Tamil Nadu and authenticated at Botany department, American College, Madurai, Tamil Nadu. A voucher specimen has been preserved in our herbarium (KMCH/Herb/2008/48).

Preparation of extracts

Five hundred grams of coarse powder of shade dried leaves of *Rhinacanthus nastus* was extracted successively with petroleum ether (60-80°C), chloroform, ethyl acetate and methanol in soxhlet extractor for 48 h. dark green

residues were obtained after concentrating the extract under reduced pressure (yield 5.5%, 6.2%, 5.8% and 6.5% respectively). The aqueous extract was obtained by macerating 500 g of coarse powder of *Rhinacanthus nastus* leaves with 5 L of distilled water for 72 h. the extract was filtered and concentrated under reduced pressure to obtain green colored residue (yield 4.8%). The obtained extracts were stored in dessicator for further phytochemical and antimicrobial investigations. The dried material was tested for its constituents by standard methods (12-13) and the results were tabulated in Table 1.

The plant extracts were diluted with respective solvents to the final concentration of 20 mgml⁻¹. Microorganisms like *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumonia*, *Proteus vulgaris* and *Candida albicans* were used for testing.

ANTIBACTERIAL EVALUATION

Disc diffusion method was adopted for the antibacterial study (14-16). Ciprofloxacin at conc of 10 mcg/disc was used as a standard. The filter paper impregnated with extracts (separately in each extracts at a concentration of 20 mgml⁻¹) and ciprofloxacin disc were placed aseptically on the seeded agar medium (Hi-Media Pvt. LTD, Mumbai) which was already swabbed with the test organisms and incubated at 37°C for 24 h. The zone of inhibition in mm was measured and recorded in Table 2.

ANTIFUNGAL EVALUATION

The antifungal activity of the crude extracts was determined against *Candida albicans* by disc diffusion

method (14-16). Ketoconazole (10 mcg/disc⁻¹) was used as standard. The filter paper disc impregnated with various extracts (20 mgml⁻¹) individually and ketoconazole disc were placed aseptically on the seeded sabouraud dextrose agar medium (Hi-Media Pvt Ltd., Mumbai) which was already swabbed with the test organism and incubated at 37°C for 48 h. the zone of inhibition (in mm) was measured and recorded in Table 2.

RESULTS AND DISCUSSION

Six crude extracts of *Rhinanthus nasutus* were investigated for their potential anti-bacterial and anti-

fungus activities. Standard antibiotics ciprofloxacin (10 mcg/disc) and ketoconazole (10 mcg/disc) showed good inhibitory action on the microorganisms tested. Petroleum ether extracts did not show any interesting inhibitory action. Chloroform and ethyl acetate extracts showed anti-bacterial and anti-fungal activities against all the organisms tested except *pseudomonas aeruginosa* and *proteus vulgaris*. Methanolic extract showed significant activity only against *Staphylococcus aureus*. Aqueous extract was ineffective for the organisms tested. The antibacterial and antifungal activity may be due to the presence of phytosterols and terpenoids in chloroform and ethyl acetate extracts.

Table 1. Phytochemical Screening of *Rhinacanthus naustus* Linn

Phytoconstituents	PE	CL	EA	ME	AQ
Phytosterol	+	+	+	+	--
Terpenoids	+	+	+	+	--
Carbohydrates	--	--	--	+	+
Flavonoids	--	--	--	+	+
Proteins	--	--	--	--	--
Alkaloids	--	--	--	--	--
Glycosides	--	--	--	--	--
Mucilage	--	--	--	--	--

PE - Petroleum ether extract

CL - Chloroform extract

EA - Ethyl acetate extract

ME - Methanol extract

AQ - Aqueous extract

+ present

- absent

Table 2. Evaluation of antibacterial and antifungal activity of *Rhinacanthus naustus* Linn

S.N	Microorganism	Zone of Inhibition (mm)					
		Std	PE	CL	EA	ME	AQ
1	<i>Staphylococcus aureus</i>	32	16	18	20	20	08
2	<i>Escherichia coli</i>	25	10	10	10	--	--
3	<i>Klebsiella pneumonia</i>	24	05	05	20	15	--
4	<i>pseudomonas aeruginosa</i>	23	--	--	--	--	--
5	<i>Proteus vulgaris</i>	32	--	--	--	--	--
6	<i>Candida albicans</i>	23	10	22	20	--	--

PE - Petroleum ether extract

CL - Chloroform extract

EA - Ethyl acetate extract

ME - Methanol extract

AQ - Aqueous extract

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