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Content

Antimicrobial activity of *Capparis zeylanica* Linn. *roots*

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The present study was designed to screen antimicrobial activity of *Capparis zeylanica* Linn. The coarse material of *C. zeylanica* roots was successively extracted with petroleum ether, chloroform and ethanol using Soxhlet and macerated to form water extract. All extracts were screened for its antibacterial and antifungal activity using agar well diffusion method. The microorganisms used for antibacterial and antifungal activity were *Bacillus pumillus* (NCIM-2752), *Staphylococcus aureus* (NCIM-2901), *Bacillus subtilis* (NCIM-2063), *Escherichia coli* (NCIM-2256), *Klebsiella pneumoniae* (NCIM-2957), *Proteus vulgaris* (NCIM-2027), *Candida albicans* (MTCC-3018) and *Aspergillus niger* (MTCC-404). Gentamicin 5 µg/ml and Clotrimazol 5 µg/ml were used as standards. The extracts showed antimicrobial activity were subjected to minimum inhibitory concentration assay by two-fold dilutions method. Petroleum ether, chloroform, ethanol and water extract exhibited *in-vitro* antibacterial activity. None of the extracts showed antifungal activity.

Key words: *Capparis zeylanica* L, antimicrobial activity, Indian caper

INTRODUCTION

Medicinal herbs are an indispensable part of the traditional medicine practised all over the world because of low costs, easy access and ancestral experience (Machado *et al.*, 2003). *Capparis zeylanica* Linn. (Capparidaceae), commonly known as Indian caper, is a climbing shrub found throughout India and has been used as a 'Rasayana' drug in the traditional Ayurvedic system of medicine. In North India, the leaves are widely used as counter-irritant, febrifuge and as a cataplasm in swellings and piles (Kirtikar and Basu, 1987). *Capparis* species has been reported to have antihelmintic, antimicrobial (Mali *et al.*, 2004) and anti-inflammatory (Chaudhary *et al.*, 2004) activities. We found relevant literature substantiating the uses indicated. Modern phytochemical screening of the plant has shown the presence of fatty acids (Haque *et al.*, 2004), flavonoids (Sabhi *et al.*, 1985) and alkaloids (Cordell, 1981) in its leaves. An attempt was made to evaluate the antimicrobial activity of different extracts of *C. zeylanica* roots.

MATERIALS AND METHODS

Plant Material

The roots of *C. zeylanica* were collected from the local areas of Wardha district, Maharashtra, India, and were authenticated by the authority of Botany Department, Nagpur University, Nagpur, India, where a voucher specimen No. 6581 was deposited.

Preparation of Extracts

The roots were collected, cleaned and shade-dried. The dried roots were pulverized by a mechanical grinder and passed through a 20-mesh sieve. A powdered root (500 g) was extracted successively with petroleum ether, chloroform and ethanol using a Soxhlet apparatus and water extracted by cold maceration. The extraction was carried out for 24 h at room temperature with mild shaking (Chopra *et al.*, 1992). The extracts were filtered and concentrated at 45°C, and the weight of each residue was recorded and percent yield was calculated.

Screening for Antibacterial and Antifungal Activity

The antibacterial and antifungal activity was evaluated by employing 24-h cultures of *Bacillus pumillus* (NCIM-2752), *Staphylococcus aureus* (NCIM-2901), *Bacillus subtilis* (NCIM-2063), *Escherichia coli* (NCIM-2256), *Klebsiella pneumoniae* (NCIM-2957), *Proteus vulgaris* (NCIM-2027), *Candida albicans* (MTCC-3018) and *Aspergillus niger* (MTCC-404). Activity of abovementioned extracts was tested separately using agar well diffusion method. The bacterial strains employed in the study were obtained from National Chemical and Industrial Microorganisms (NCIM), Pune. Antifungal strains were obtained from MTTC. The medium was sterilized by autoclaving at

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120°C (15 lb/in^2). About 30 ml of nutrient agar medium inoculated with the respective strains of bacteria and fungi was transferred aseptically into each sterilized Petri plate. The plates were left at room temperature to allow solidification. In each plate, a single well of 6-mm diameter was made using a sterile borer. The extracts were freshly reconstituted with suitable solvents (dimethyl sulphoxide) and tested at various concentrations. The test sample and the control (0.2 ml) were placed in 6-mm diameter well. Antibacterial assay plates were incubated at 37 ± 1°C for 24 h, whereas antifungal assay plates were incubated at 28 ± 1°C for 48 h. A standard disc (6-mm diameter) with antibiotic Gentamicin (5 µg/ml) was used as positive antibacterial control, whereas Clotrimazole (5 µg/ml) was used as positive antifungal control. Each experiment was carried out in triplicates, and diameter of the zone of inhibition surrounding each well was recorded. Observations and results are shown in Table 1. The extracts that showed antimicrobial activity were subjected to minimum inhibitory concentration (MIC) assay by using serial two-fold dilution method (Florey *et al.*, 1989). MIC was interpreted as the lowest concentration of the sample, which showed clear fluid without development of turbidity; observations and results are shown in Table 2.

RESULTS AND DISCUSSION

Results of antimicrobial assay showed that chloroform, ethanol and water extracts of *C. zeylanica* roots exhibited *in vitro* antibacterial activity against Gram-positive and Gram-negative bacteria, whereas petroleum ether exhibited antibacterial activity against select bacterial strains. None of the extracts exhibited antifungal activity. In terms of specific inhibition, petroleum ether extract inhibited *S. aureus*, *B. subtilis*, *K. pneumoniae* and *P. vulgaris* and produced inhibition zone ranging from 10 to 16 mm at a concentration of 16.5 µg/ml, whereas chloroform, ethanol and water extracts showed inhibitory activity against all six bacterial strains at concentrations of 13.5, 14.0 and 14.0 µg/ml, respectively (Table 1).

Minimum inhibitory concentration of active extracts is shown in Table 2. The lowest MIC values were observed for ethanol extract (11.25-14 µg/ml), chloroform extract (11.5-13.5 µg/ml), water extract (12.5-14 µg/ml) and petroleum ether extract (12.5-16.5 µg/ml) against the bacteria tested (Table 2). The results reveal that extracts of *C. zeylanica* roots were effective against both Gram-positive and Gram-negative bacteria; some of these bacteria are associated with conditions like infections of the skin and urinary tract. Preliminary phytochemical screening of the extracts showed the presence of alkaloids, flavonoids, terpenoids, sterols, tannins and carbohydrates. Isolation, purification and characterization of the phytochemicals responsible for the aforementioned activity are in progress. Further work on the profile and nature of chemical constituents of *C. zeylanica* roots will provide more information on the

Table 1: Antimicrobial activity of *Capparis zeylanica* root extracts

Test samples	Concentration (µg/ml)	Zone of inhibition (mm)*							
		Gram-positive/Gram-negative fungi							
		S.a.	*B.s.*	*B.p.*	*E.c.*	*K.p.*	*P.v.*	*C.a.*	*A.n.*
Petroleum ether extract	16.5	10	-	13	-	16	-	-	-
Ethanol extract	13.5	14	11	10	7	12	8	-	-
Chloroform extract	14.0	9	12	9	13	12	8	-	-
Water extract	14.0	10	11	8	15	11	9	-	-
Gentamicin	10.0	23	22	21	23	20	21	nt	nt
Clotrimazole	10.0	nt	nt	nt	nt	nt	nt	16	18

*Values are the means of three assays, -: no activity; nt: not tested, *S.a. - Staphylococcus aureus* (NCIM-2901); *B.s. - Bacillus subtilis* (NCIM-2063); *B.p. - Bacillus pumillus* (NCIM-2752); *E.c. - Escherichia coli* (NCIM-2256); *K.p. - Klebsiella pneumoniae* (NCIM-2957); *P.v. - Proteus vulgaris* (NCIM-2027); *C.a. - Candida albicans* (MTCC-3018); *A.n. - Aspergillus niger* (MTCC-404)

Table 2: Minimum inhibitory concentrations of *Capparis zeylanica* root extracts

Test sample(s)	Minimum inhibitory concentrations (mg/ml)*					
	Gram-positive/Gram-negative					
	S.a.	*B.s.*	*B.p.*	*E.c.*	*K.p.*	*P.v.*
Ethanol extract	12.5	11.25	13.25	11.75	14.0	12.25
Chloroform extract	13.5	11.75	11.5	13.25	11.5	12.25
Water extract	12.75	13.25	14.0	12.5	14.0	13.75
Petroleum ether extract	16.25	nt	16.50	nt	12.5	nt

*Values are the means of three assays. nt - not tested. *S.a. - Staphylococcus aureus* (NCIM-2901); *B.s. - Bacillus subtilis* (NCIM-2063); *B.p. - Bacillus pumillus* (NCIM-2752); *E.c. - Escherichia coli* (NCIM-2256); *K.p. - Klebsiella pneumoniae* (NCIM-2957); *P.v. - Proteus vulgaris* (NCIM-2027)

bioactive principles responsible for their pharmacological properties.

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