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Research Article

ANTIMICROBIAL ACTIVITY OF THE DRIED ROOT POWDER OF *CAPPARIS GRANDIFLORA* WALL.EX HOOK .F & THOMSON

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ABSTRACT

The present study was designed to screen the anti microbial activity of dried root powder of *Capparis grandiflora* Wall ex.Hook.f & Thomson. The coarse material of *Capparis grandiflora* roots was successively extracted with petroleum ether, chloroform and ethanol using a soxhlet apparatus and water extracted by cold maceration. *In vitro* antimicrobial activity of various extracts from the roots of *Capparis grandiflora* (Capparidaceae), growing in the adjacent regions of South India was evaluated using agar well diffusion method. The micro organisms used for antibacterial and antifungal activity were *Staphylococcus aureus* (NCIM-2079), *Bacillus subtilis* (NCIM-2063), *Bacillus pumillus* (NCIM-2752), *Escherichia coli* (NCIM-2064), *Klebsiella pneumoniae* (NCIM-2957), *Proteus vulgaris* (NCIM-2027), *Candida albicans* (NCIM 3100) and *Aspergillus niger* (MTCC-404).Gentamicin (10 mg/ml) and Clotrimazol (10 mg/ml) were used as standards. The extracts that showed antimicrobial activity were subjected to minimum inhibitory concentration assay by two-fold dilution method. All extracts, at higher concentrations showed varying degrees of inhibitory activity against all bacteria. None of the extracts showed antifungal activity.

Keywords: *Capparis grandiflora*, Antimicrobial activity, Minimum Inhibitory Concentration (MIC), Zone of inhibition.

INTRODUCTION

The emergence of pathogens resistant to antibiotics as a result of excessive use of them in clinical and veterinary applications, represent a serious problem for public health. Despite the existence of efficient antibiotics, drug resistant or multi- drug resistant strains are steadily appearing and require longer and more expensive treatments. [1]

However, there has also been a rising interest in the research for natural products from plants for the discovery of new antimicrobial and antioxidant agents in the last three decades and in recent times. [2] Most plant extracts have been shown to possess anti-microbial agents active against micro organisms in vitro. These plants contain medicinal properties which make them potent to cure or prevent diseases. [3]

Hence, the plant kingdom is being screened for newer and effective chemotherapeutic agents. Higher plants can serve both as potential antimicrobial crude drugs as well as a source of new anti-infective agents. [4] More so, many of these plants have been known to synthesize active secondary metabolites such as phenolic compound found in essential oils with established antioxidant and antimicrobial activities,[5] which indeed has formed the basis for their applications in some pharmaceuticals, alternative medicines and natural therapies .[6]

Capparis grandiflora is a climbing shrub with spreading branches found mainly in the adjacent regions of Coimbatore, Nilgiry andTiruchirappalli, South India. [7]

Communication with the traditional practitioners have revealed the use of the plant as a stomachic, diuretic , anti rheumatic, shortness of breath and anti tumour. *Capparis* species has been reported to have anthelmintic, [8] antimicrobial and anti inflammatory activities. [9] Modern phytochemical screening has shown the presence of fatty acids, [10] flavonoids and alkaloids. [11, 12] An attempt was made to evaluate the antimicrobial activity of different extracts of *C. grandiflora* roots.

MATERIALS AND METHODS

The roots of *Capparis grandiflora* Wall. ex Hook.f &Thomson (Capparidaceae), were collected from the tribal areas of Palakkad district, Kerala state, India and were authenticated by the Botanical survey of India, Combatore, Tamilnadu (BSI). A voucher specimen (no.BSI/SRC/5/23/10-11/Tech-565) is deposited in the departmental herbarium.

Preparation of Crude extract:

The roots were cleaned and shade dried. The dried roots were pulverized by a mechanical grinder and passed through a 20 mesh sieve. A powdered root material was successively extracted 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. [13]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 *Staphylococcus aureus* (NCIM-2901), *Bacillus subtilis* (NCIM-2063), *Bacillus pumillus* (NCIM-2752), *Escherichia coli* (NCIM-2256), *Klebsiella pneumoniae* (NCIM-2957), *Proteus vulgaris* (NCIM-2027), *Candida albicans* (MTCC-3018) and *Aspergillus niger* (MTCC-404). Activity of above mentioned extracts was tested separately using agar well diffusion method. The bacterial strains employed in the study were obtained from National Chemical and Industrial Micro organisms (NCIM), Pune.Anti fungal strains were obtained from MTTC.The medium was sterilized by autoclaving at 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 (10 µg/ml) was used as positive antibacterial control, whereas Clotrimazole (10 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 .[14] 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].

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

Test samples	Concentration(µg/ml	Zone of inhibition Gram positive/Gram negative/Fungi							
		S.a	Bs	Bp	Ec	Kp	Pv	Ca	An
Petroleum ether extract	18.5	11	--	13	--	15	--	--	- -
Ethanol extract	14.5	16	11	9	8	12	7	--	---
Chloroform extract	14.0	8	12	8	7	11	8	---	---
Water extract	14.0	10	12	8	14	11	9	---	---
Gentamycin	10.0	21	22	23	21	20	21	nt	nt
Clotrimazole	10.0	Nt	nt	nt	nt	nt	nt	17	19

*Values are mean of three assays,- no activity; Nt: not tested; Sa: *Staphylococcus aureus* ; Bs:*Bacillus subtilis* ; Bp: *Bacillus pumillus*; Ec: *Escherichia coli;* Kp: *Klebsiella pneumoniae* ; Pv: *Proteus vulgaris*; Ca:*Candida albicans*; An:*Aspergillus niger* .

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

Test samples	Minimum inhibitory concentrations (mg/ml)					
	Gram pos itive			Gram negative		
	Sa	Bs	Bp	E.c	Kp	Pv
Ethanol extract	11.75	12.25	13.0	11.75	13.75	11.5
Chloroform extract	14.0	13.5	11.0	13.5	11.0	12.5
Water extract	13.5	12.0	14.5	12.75	14.5	11.75
Petroleum ether extract	14.25	nt	15.5	nt	12.5	nt

Values are mean of three assays; NT: not tested; SA: *Staphylococcus aureus*; Bs: *Bacillus subtilis*; Bp: *Bacillus pumillus*; Ec: *Escherichia coli;* KP: *Klebsiella pneumoniae*; PV: *Proteus vulgaris.*

RESULTS AND DISCUSSION

Phytochemical constituents present in the plant extract included tannins, saponins, sesquiterpenes, alkaloids, and phlobatannins. Results of the antimicrobial activity of the plant extracts are shown in [Table 1]. The result shows that Petroleum ether, Chloroform, ethanol and water extracts of *C.grandiflora* roots showed inhibitory activity against all bacterial strains at various concentrations. None of the extracts showed antifungal activity. Minimum inhibitory activity for the water extract was found to be (12.0-`14.5 µg/ml), ethanol extract (11.75-13.75 µg/ml), petroleum ether extract(12.5-15.5 µg/ml) and chloroform extract(11.0-14.0 µg/ml)against the bacteria tested[Table 2].The results reveal that extracts of *C.grandiflora* roots were effective against both gram positive and gram-negative bacteria. Isolation, purification and characterisation of the phyto chemicals responsible for the aforementioned activity are in progress. Further work on the profile and nature of chemical constituents of *C.grandiflora* roots will provide more information on the bioactive principles responsible for their pharmacological properties.

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