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Research Article

ANTI-BACTERIAL PROPERTIES OF PROSOPIS **CINERARIA** (L.) DRUCE

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Abstract

In the present study, the antibacterial efficacy of ethanol and aqueous extracts of leaf of Prosopis cineraria was examined against four bacterial species (Bacillus subtilis, Staphylococcus aureus, Klebsiella pneumoniae and Proteus mirabilis) using agar well diffusion method. Results showed that both the plant extracts exhibited a dosedependent inhibition of microorganisms. Ethanol extracts showed higher degree of antibacterial activity than aqueous extracts. The phytochemical test confirmed the presence of bioactive secondary metabolites like alkaloids, tannins, flavonoids, saponins, steroids, terpenoids and glycosides. The plant has high value of secondary metabolite which in turn is responsible for its antioxidant and antibacterial activity.

Key words: Prosopis cineraria, agar well diffusion assay, antibacterial activity, ethanol and aqueous extracts.

INTRODUCTION

In recent years, the indiscriminate use of synthetic drugs against microbial pathogens has resulted in mutation of strains making them insensitive to these chemical agents leading to the global hazard of drug resistance. The scientists of the 21st century are generally reviving our traditional knowledge and are screening various parts of plants scientifically, in search of newer lead compounds having antimicrobial efficacy¹. It is high time the hidden wonders of plant molecules were revived with the modern tools of target-based screening to develop newer advanced generation of antimicrobials with novel modes of action.

Prosopis cineraria is a flowering tree belonging to the family Fabaceae and contains approximately forty four species². It is native to Western and South Asia, including Afghanistan, Iran, India and Pakistan. Its common names include Khejri (India), Jand and Kandi (Pakistan). It is the state tree of Rajasthan (India) and provincial tree of the Sindh province of Pakistan³. Different parts of the plant are useful for the treatment of many diseases like skin diseases, piles, worms, vertigo and dyspnoea⁴, protection from miscarriage, eye diseases, snake bite, asthma, bronchitis, dysentery, leucoderma, leprosy and muscle tremors⁵.

MATERIALS AND METHODS **Plant Collection and Extraction**

The leaves of Prosopis cineraria were collected from Thindal, Erode district and were authenticated and deposited at the PG and Research Department of Botany, Vellalar College for Women, Erode (Tamil Nadu), India. Fresh leaves were collected and airdried at room temperature and then homogenized to obtain coarse powder. The powder leaf was extracted ⁶ with the solvent ethanol by hot extraction using soxhlet apparatus, collected and stored in a vial for further analysis.

Phytochemical Screening

Preliminary Phytochemical analysis of leaf was undertaken and tested for various antibacterial phytoconstitutents^{7,8}.

Antibacterial Assay **Agar Well Diffusion Method**

The antibacterial activity of ethanolic and aqueous leaf extracts of Prosopis cineraria was evaluated by well diffusion method⁹. The inoculation of microorganisms was prepared from bacterial culture. About 20ml of Muller Hinton agar medium was poured in a sterilized petridish and allowed to solidify. One drop of bacterial strain was spread over the medium by a rod. Wells of 5mm in diameter and about 2cm apart were punctured in the culture medium using sterile cork borers. Varying concentrations of the ethanol and aqueous leaf extracts (25, 50, 75 and 100μ g/ml) was added to the wells separately and the plates were incubated at 37°C for 24h. Streptomycin (50 μ g/ml) the standard antibiotic was used as positive control. Antibacterial activities were assessed by measuring inhibition zone in diameters and the results were given in relative magnitude of inhibition (RMI).

Microorganisms Tested

The test microorganisms used in the present study were Gram positive:

Bacillus subtilis, Staphylococcus aureus Gram negative: Proteus mirabilis, Klebsiella pneumoniae.

Bacterial Strains used in this study were purchased from Kovai Medical College Hospital (KMCH), Coimbatore. Bacteria were cultured overnight at 37⁰C in Muller and Hilton Broth (MHB) 72hours in Potato Dextrose Broth and used as inoculum.

RESULTS AND DISCUSSION

The basic theme and fundamental need of this plant oriented project was to explore the hidden medicinal potential of the plants. These secondary metabolites comprises of alkaloids, flavonoids, glycosides, tannins, saponins, steroids and triterpenoids as shown

in the Table 1. The result of the antibacterial activities of the ethanol and aqueous leaf extracts of P. cineraria compared with that of the standard antibiotic are shown in Tables 2 and 3. The ethanol extract showed maximum activity against Staphylococcus aureus and Klebsiella pneumoniae at 100µg/ml concentration which was comparable with the standard antibiotic drug at 50µg/ml concentration (Table 2). On the other hand, the aqueous extract showed maximum inhibitory effect against Proteus mirabilis followed by Klebsiella pneumoniae at 100µg/ml concentration. Earlier the phytochemical and antimicrobial properties were reported by researchers from dried unripe pods¹⁰, seeds¹¹ stem, leaf and bark¹², roots¹³, leaves and fruits¹⁴ of Prosopis cineraria.

CONCLUSION

Plants are the biggest source of medicine in future. The present study exhibited the antibacterial effect of ethanolic and aqueous extracts of *Prosopis cineraria* against *Bacillus subtilis*, *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Proteus mirabilis*. The obtained activity may be due to the presence of flavonoids and tannins (presence is confirmed by the preliminary phytochemical studies). The inhibitory effect of the extracts justified the medicinal use of *P.cineraria*. The plant extracts thus provides safe, easy, effective and practical solutions to every day ailments leaving behind no toxins and creating a clean, pleasant atmosphere. Further studies are under progress to characterize the active principles.

S. No.	Phytoconstituents	Ethanolic extract	Water extract
1.	Alkaloids (Dragendorff's test)	+	+
2.	Flavonoids (Alkaline reagent test)	+	+
3.	Tannins (Ferric Chloride test)	+	+
4.	Saponins (Froth test)	-	+
5.	Steroids (Salkowski's test)	+	-
6.	Triterpenoids (Libermann-Burchard's test)	+	+
7.	Glycosides (Keller-Kiliani test)	+	-

 Table 1

 PHYTOCHEMICAL SCREENING OF LEAF OF PROSOPIS CINERARIA

Table 2

ACTIVITY OF ETHANOLIC EXTRACT OF LEAF OF *P.CINERARIA* (RMI-cm²)

	Name of Bacteria	Concentration of leaf extract (µg/ml)														
S. No.		AB			25			50			75			100		
		A ₁	\mathbf{A}_2	RMI	A ₁	A_2	RMI	A ₁	\mathbf{A}_2	RMI	A ₁	\mathbf{A}_2	RMI	A ₁	A2	RMI
1.	Bacillus subtilis	0.5	1.5	3.0	0.5	0.5	1.0	0.5	0.9	1.8	0.5	1.2	2.2	0.5	1.4	2.6
2.	Proteus mirabilis	0.5	1.6	3.2	0.5	0.7	1.4	0.5	0.8	1.6	0.5	1.2	2.4	0.5	1.6	2.8
3.	Klebsiella pneumoniae	0.5	1.8	3.6	0.5	1.1	2.2	0.5	1.3	2.6	0.5	1.5	3.0	0.5	1.7	3.4
4.	Staphylococcus aureus	0.5	1.8	3.6	0.5	1.2	2.4	0.5	1.4	2.8	0.5	1.6	3.2	0.5	1.7	3.4
$A_1 =$	= Area of well in cm^2 RMI = A_2/A_1															

 A_2 = Area of zone of inhibition in cm² (including area of well)

AB = Antibiotic (Streptomycin -50 μ g/ml)

	Table 3	
ANTIBACTERIA	L ACTIVITY OF AQUEOUS EXTRACT OF LEAF OF <i>P.CINERARIA</i> ((RMI-cm ²)

	Name of Bacteria	Concentration of leaf extract (µg/ml)														
S.No.		AB			25			50			75			100		
		\mathbf{A}_{1}	\mathbf{A}_2	RMI	A ₁	A2	RMI									
1.	Bacillus subtilis	0.5	1.5	3.0	0.5	0.7	1.4	0.5	0.8	1.6	0.5	1.2	2.4	0.5	1.4	2.8
2.	Proteus mirabilis	0.5	1.7	3.4	0.5	0.9	1.8	0.5	1.1	2.2	0.5	1.4	2.8	0.5	1.6	3.2
3.	Klebsiella pneumoniae	0.5	1.6	3.2	0.5	0.6	1.2	0.5	0.8	1.6	0.5	1.0	2.0	0.5	1.5	3.0
4.	Staphylococcus aureus	0.5	1.5	3.0	0.5	0.7	1.4	0.5	0.9	1.8	0.5	1.2	2.4	0.5	1.4	2.8

 A_1 = Area of well in cm²

 A_2 = Area of zone of inhibition in cm² (including area of well)

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 $\mathbf{RMI} = \mathbf{A}_2 / \mathbf{A}_1$

AB = Antibiotic (Streptomycin -50 μ g/ml)

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