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In Vitro Evaluation of Antibacterial Activity of Ethanolic and Aqueous leaf Extracts of *Sesbania Grandiflora (Linn)* Against Clinical Pathogens.

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Abstract

The aim of the present study is to evaluate the antibacterial activity of ethanolic and aqueous leaf extract of *Sesbania grandiflora* Linn on selected bacterial species. *Sesbania grandiflora* (*Linn*) *is* a medicinal herb belongs to the family Leguminosae .The various parts of the plant like roots, bark, leaves, flower and fruit are known to possess different pharmacological properties. The leaf extract of *Sesbania* possess good anticonvulsant and anxiolytic activity. The leaves of *Sesbania* is also found to possess hepatoprotective and potent anti oxidant and anti urolithiatic activity .The Ethanolic and Aqueous extract of leaves were screened for the antimicrobial activity by agar well diffusion method. The test organisms used were *Staphylococcus aureus*, *E.coli, Klebsiella pneumoniae, Psuedomonas aeruginosa* and *Bacillus subtilus*. The results obtained from the study showed that the ethanolic extract has significant antibacterial activity on *Staphylococcus aureus* and *E.coli* compared with standards.

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Anti-Bacterial activity, *Sesbania grandiflora*, Clinical pathogens, Agar well diffusion technique, Zone of Inhibition.

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Introduction

Ayurvedic medicine, also called Ayurveda, originated in India several thousand years ago.

The term "Ayurveda" combines the Sanskrit words *ayur* (life) and *Veda* (science or knowledge) thus; Ayurveda means "the science of life. *Sesbania grandiflora* (also known as agati, syn. *Aeschynomene grandiflora*) or hummingbird tree / scarlet wisteria is a small tree in the genus *Sesbania*. *Sesbania grandiflora* (Linn) belonging to family *Leguminosae* contains plenty of sterols, saponins and tannins which are responsible for its various pharmacological properties and has been widely used in Ayurveda. *Sesbania grandiflora* is a loosely branching tree up to 15 m tall. Its leaves are pinnately compound up to 30 cm long with 20-50 leaflets in pairs, dimensions 12-44 x 5-15 mm, oblong to elliptical in shape. Flowers are large, white, yellowish, rose pink or red with a calyx 15-22 mm long. The standard has dimensions up to 10.5 x 6 cm. Pods are long (20-60 cm) and thin (6-9 mm) with broad sutures containing 15-50 seeds. It is believed to have originated either in India or Southeast Asia and grows primarily in hot and humid areas of the world. Plant pacifies vitiated vata, rheumatism, arthritis and scabies. Agathi is used as an ayurvedic remedy for treating fever, sinus, bruises, and venereal diseases etc[1].It is one of the richest natural sources of vitamin A.This tree occurs also in India to the Mascarene Islands, through Malaya to tropical Australia, and is planted in other tropical countries [2-7]. The active ingredients of sesbania are leucocyanidin and cyanidin present in seeds, oleanolic acid and its methyl ester and kaemferol-3-rutinoside which are present in flower. The bark contains tannins and gum. Saponin is originated from the seeds.Sesbanimide is derived from seeds. The various parts of *sesbania* are used as medicine for many diseases and disorders .[8]In Folk Medicine it is resorted to be aperient, diuretic, emetic, emmenagogue, febrifuge, laxative, and tonic.[9] Agati is a folk remedy for bruises, catarrh, dysentery, eyes, fevers, headaches, smallpox, sores, sore throat, and stomatitis.[10] Agati is also used in the treatment of anemia, bronchitis, headache, nasal, inflammation, leprosy, gout and rheumatism[11,12,13]

The Bark of *Sesbania* possess potent Anti ulcerogenic activity.[14] *Sesbania* is a potent antidote for tobacco and smoking-related diseases. [15]The flower of *Sesbania* is shown to possess Anti-cancer, chemo preventive [16]Anti- microbial[17], Analgesic, Anti pyretic activity[18]. The leaf extract of *Sesbania* possess good anticonvulsant and Anxiolytic activity[19] due to the presence of triterpene. The leaves of *Sesbania* is also found to possess Hepatoprotective and potent antioxidant and Anti urolithiatic activity [20]against calcium oxalate-type stones. Sesbania gum is available locally in large quantities and has been explored by some studies as pharmaceutical excipients. *Sesbania grandiflora* (Agati) has also been studied for formulation and evaluation of ophthalmic dosage form for the improvement of dim vision [21]. Hence in our study we tested the effect of Aqueous and Ethanolic leaf extract of *Sesbania grandiflora* against few clinical pathogens to treat infectious disease.

MATERIALS AND METHODS

Plant material

The Ethanolic and Aqueous extracts of *Sesbania grandiflora* were obtained from Green Chem Herbal Extract & Formulations, Bangalore.

Test microorganisms

Bacterial strains used were *E.coli [GNB], Klebsiella pneumonia [GNB], Pseudomonas aeruginosa [GNB], Staphylococcus aureus, [*Gram positive cocci] and *Bacillus subtilus*. The organisms were obtained from department of Microbiology, Saveetha Dental College and maintained in nutrient agar slope at 4°C.

METHODOLOGY:

The extracts were prepared in the following concentrations in sterile water. 50mg /ml, 100mg/ml and 200mg /ml, so that 50µl of extract of different concentrations delivers 2.5mg, 5mg and 10 mg respectively.

Table 1: Pharmacological activity of various parts of *Sesbania grandiflora(Linn.)*

Scientific Name	Common Name	Family	Parts used	Pharmacological activity.
Sesbania grandiflora	Agati Sennabean Drummond Sesbania	Leguminosae	Bark Leaf Flower	Anti ulcerogenic activity. Anticonvulsant Anxiolytic,Hepatoprotective, antioxidant and Antiurolithiatic activity. Anti cancer, Anti microbial Analgesic and Anti pyretic activity.

Assay for the Antibacterial Activity Using Agar Well Diffusion Method

The screening of antibacterial activity of plant extracts was carried out using the agar well diffusion method. The bacterial strains were inoculated into tubes of Brain heart infusion agar and incubated at 37°C overnight. Each of the cultures were then adjusted to 0.5 Mc Farland turbidity standard.[22-25] Lawn culture of the test organisms were made on the Brain heart infusion agar [BHI-Hi media M211] plates using sterile cotton swab and the plates were dried for 15 minutes. A sterile cork borer was then used to make wells (6mm diameter) for different concentrations of the extracts on each of the plates containing cultures of the different bacterial strains. 50µl of the varying concentrations (2.5mg, 5mg and 10 mg) of the extracts were introduced into the wells with the help of micropipette and were then incubated in upright position at 37°C for 24 h. After 24 hrs, antibacterial activity was determined by measurement of diameter of zones of inhibition (mm). Standard antibiotic discs of Amoxicillin (30mcg/disc) and Penicillin G (30mcg/disc) were used as positive control. All the tests were done in triplicate to minimize the test error.

RESULT AND DISCUSSION

The antibacterial activity of the extracts (Ethanolic and Aqueous) at different concentrations was screened by agar well diffusion technique and the zone of inhibition was measured in mm diameter. The results are given in the table 1.

The ethanolic extract was more effective against *Staphylococcus aureus* and *E.coli* with a zone of inhibition of 22 mm and 20mm diameter (at conc. 10 mg.) respectively and was least effective against *Klebsiella pneumonia*, *Pseudomonas aeruginosa* and *Bacillus subtilus* with zone of inhibition of 10 mm,9 mm,and11 mm(at conc. 10mg.) respectively.

The aqueous extract also showed activity against *Staphylococcus aureus* and *E.coli* with a zone of inhibition of 18 mm and 16mm diameter (at conc. 10 mg). With other tested bacterial strains, the aqueous extract was not found to be effective.

Table 2: Anti bacterial activity of Aqueous and Ethanolic Leaf extracts of*Sesbania grandiflora* Linn

Extract	Conc [mg]	Zone of inhibition [in mm diameter]				
		B1	B2	B3	B4	B5
Ethanolic	2.5	10	-	-	12	7
	5	13	8	-	18	9
	10	20	10	9	22	11
Aqueous						
	2.5	8	-	-	10	-
	5	10	-	-	14	-
	10	16	-	-	18	7
Ciprofloxacin	30 mcg/ disc	24	21	22	23	20
Amoxycillin	30 mcg/ disc	25	23	20	21	18

- No significant zone of inhibition

B1-*Escherichia coli*, B2- *Klebsiella pneumoniae*, B3-*Pseudomonas aeruginosa* B4- *Staphylococcus aureus,* B5-*Bacillus subtilus*

CONCLUSION

The use of herbs in folk medicine suggests that they represent an economic and safe alternative to treat infectious diseases. It is clear from the results that, the leaf extracts of *Sesbania grandiflora* acts as a good source of antimicrobial agent against *Staphylococcus aure*us and *E.coli*. The anti-bacterial activities could be enhanced if active components are purified and adequate dosage is determined for proper administration.

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