Antibacterial and Antifungal Activities of *Sesbania aegyptiaca* Pers. Leaves

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Accepted 30 December 2005
Revised 24 March 2005
Received 23 November 2004

The crude extracts (petroleum ether, benzene, chloroform, ethyl acetate, methanol and water) of *Sesbania aegyptiaca* leaves exhibited varying degrees of antibacterial activity against seven Gram positive and Gram negative bacterial strains out of nine tested and antifungal activity against two fungal strains out of four tested.

Sesbania aegyptiaca Pers. (Fabaceae) is a shrub. The leaves of the plant are used in the treatment of wounds and skin ailments. Juice of the leaves is credited with anthelmintic properties and poultice of the leaves is claimed to promote suppuration of boils, abscesses and abnormalities of inflammatory rheumatic swellings[1]. Leaves are purgative, demulcent; useful in hydrocele and in all types of inflammations[2]. A water-soluble galactomannan, saponin, and kaempferol trisaccharide were isolated from the plant[3,4,5]. The present investigation was undertaken to evaluate antibacterial and antifungal activities of leaf extracts of *S. aegyptiaca.*

Leaves of the plant were collected from Coimbatore district and identified by Dr. Gopalan, Scientist, Botanical Survey of India (Southern Circle), Government of India, Coimbatore. The leaves were shade dried and powdered. They were extracted successively in soxhlet apparatus with petroleum ether, benzene, chloroform, ethyl acetate, methanol and hot water. The extracts were condensed to dryness by rotary flash evaporator (Buchi type). For antibacterial and antifungal activity 10, 5, 2.5 mg/ml concentrations were made from the crude extracts. The microbial strains, *Escherichia coli*, MTCC 443, *Staphylococcus aureus* MTCC 737, *Salmonella typhi* 734 and *Pseudomonas aeruginosa* MTCC 741 were procured from Institute of Microbial Technology (IMTECH), Chandigarh. Other microbial strains mentioned here *Bacillus subtilis, Streptococcus pyogenes, Klebsiela pneumoniae, Pseudomomas fluorescence* and *Salmonella enteritidis* were clinical isolates obtained from the Department of Microbiology of our college. The fungal strains *Mucor* sp., *Aspergillus niger, Aspergillus fumigatus* and *Alternaria alternata* were also obtained from the department of Microbiology. Antibacterial and antifungal activities of the plant extracts were studied by disc diffusion method and determination of Minimum Inhibitory Concentration (MIC) by broth dilution methods[6]. Bacterial concentration of $1x10^8$ CFU/ml was used for antibacterial activity and fungal suspension of $1x10^6$ CFU/ml for antifungal activity. Filter paper discs of 6 mm (Whatman No. 1) were used. The discs were used in duplicate for each concentration. The controls employed in the antibacterial and antifungal activities were streptomycin and clotrimazole, respectively.

The petroleum ether, benzene, chloroform, ethyl acetate, methanol and water extracts of *S. aegyptiaca* were tested against 3 g positive and 6 g negative bacteria, and four fungal strains. The extracts were dissolved in dimethyl sulfoxide (DMSO) which was used as vehicle control. The results are reported in Table I and II. All the extracts at 10, 5, 2.5 mg/ml concentrations exhibited appreciable antimicrobial activity against the tested bacterial and fungal strains. The extracts were found to be more active against *P. aeruginosa* (MIC=0.125-1 mg/ml). None of them was active against *E. coli* and *K. pneumoniae*. The extracts showed significant activity against *A. alternata* and *Mucor* sp. and not active against *A. niger* and *A. fumigatus*.

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ACKNOWLEDGEMENTS

The authors express their sincere gratitude to the

TABLE 1: ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY OF DIFFERENT EXTRACTS OF *SESBANIA AEGYPTIA*LEAVES

Microorganisms	Zone of inhibition including disc diameter (mm)													
	PE				BE				CE				Str.	Clot.
	a	b	c	MIC	a	b	c	MIC	a	b	c	MIC		
B. subtilis	20	16	9	1	18	16	13	1	19	16	12	1	22	NT
S. aureus	13	10	9	2	16	14	10	1	16	10	9	1	16	NT
S. pyogenes	22	16	10	0.5	19	16	13	1	15	11	9	2	20	NT
E. coli	–	–	–	ND	–	–	–	ND	–	–	–	ND	21	NT
K. pneumoniae	–	–	–	ND	–	–	–	ND	–	–	–	ND	15	NT
P. aeruginosa	16	10	9	1	22	20	18	0.125	21	16	11	0.5	17	NT
P. fluorescence	–	–	–	ND	15	13	9	1	13	11	0	4	16	NT
S. enteritidis	15	10	9	2	14	11	10	4	12	10	9	4	14	NT
S. typhi	14	11	9	2	20	16	11	1	14	11	10	2	20	NT
Mucor sp.	20	17	12	ND	21	18	16	ND	21	19	15	ND	NT	21
A. fumigatus	–	–	–	ND	–	–	–	ND	–	–	–	ND	NT	14
A. niger	–	–	–	ND	–	–	–	ND	–	–	–	ND	NT	15
A. alternata	19	15	13	ND	27	21	18	ND	19	15	12	ND	NT	22

PE, BE, CE indicate petroleum ether extract, benzene extract and chloroform extract respectively. a, b, c indicate 10, 5, 2.5 mg/ml concentrations respectively. MIC denotes minimum inhibitory concentration. ND denotes not determined. NT denotes not tested. "-" indicates no activity. Str. and Clot. indicate streptomycin and clotrimazole respectively at 500 µg/ml

TABLE 2: ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY OF DIFFERENT EXTRACTS OF *SESBANIA AEGYPTIACA* LEAVES

Microorganisms	Zone of inhibition including disc diameter (mm)													
	EE				ME				WE				Str	Clot
	a	b	c	MIC	a	b	c	MIC	a	b	c	MIC		
B. subtilis	20	18	12	1	18	15	12	2	15	12	19	4	22	NT
S. aureus	16	14	11	1	14	11	10	2	16	12	10	2	16	NT
S. pyogenes	20	17	15	1	19	17	10	1	16	13	10	2	20	NT
E. coli	–	–	–	ND	–	–	–	ND	–	–	–	ND	21	NT
K. pneumoniae	–	–	–	ND	–	–	–	ND	–	–	–	ND	15	NT
P. aeruginosa	20	18	13	0.5	21	17	12	0.5	18	15	14	1	17	NT
P. fluorescence	12	10	9	4	13	10	9	4	–	–	–	ND	16	NT
S. enteritidis	12	9	8	4	14	11	9	2	14	11	9	2	14	NT
S. typhi	15	13	10	1	20	17	13	0.5	19	14	11	1	20	NT
Mucor sp.	25	21	18	ND	18	16	14	ND	22	20	16	ND	NT	21
A. fumigatus	–	–	–	ND	–	–	–	ND	–	–	–	ND	NT	14
A. niger	–	–	–	ND	–	–	–	ND	–	–	–	ND	NT	15
A. alternata	22	20	18	ND	21	18	15	ND	20	19	17	ND	NT	22

EE, ME, WE indicate ethyl acetate extract, methanol extract and water extract respectively. a, b, c indicate 10, 5, 2.5 mg/ml concentrations respectively. MIC denotes minimum inhibitory concentration. ND denotes not determined. NT denotes not tested. "-"indicates no activity. Str and Clot indicate streptomycin and clotrimazole respectively at 500 µg/ml

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TLC-Colourimetric Estimation of Free and Combined Forms of Chrysophanol, Emodin and Physcione in some *Cassia* species

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Accepted 30 December 2005
Revised 22 March 2005
Received 15 May 2004

Estimation of emodin, chrysophanol and physcione in free and combined form in some *Cassia* species namely *Cassia auriculata, Cassia fistula, Cassia javanica, Cassia roxburghii, Cassia siamea,* in the plant parts like bark, flower, leaf, pericarp, seed and stem (wood) has been carried out. The TLC-colorimetric method was employed for the estimation of the same. The total amount of free and combined forms of emodin, chrysophanol and physcione were found in maximum amount in *C. siamea* (1.01%) followed by *C. javanica* (0.80%), *C. fistula* (0.68%), *C. auriculata* (0.53%) and *C. roxburghii* (0.37%). The standards, chrysophanol, emodin and physcione were developed from Indian Rhubarb (*Rheum emodi*) by TLC-spectroscopic techniques.

The genus *Cassia* belongs to family Leguminosae and subfamily Caesalpiniaceae[1], out of which 23 species are found in India[2]. All the species of *Cassia* contain anthraquinone glycosides as major chemical constituent and traces of tannins and flavonoids[3]. They are identified by Borntrager's test[4]. They are useful in the treatment of constipation and they also have antispasmodic and antiinflammatory action[5].

Chrysophanol is 1,8-dihydroxy-3-methyl anthraquinone. It occurs in free and combined states in cascara, senna, and rhubarb. Emodin is 1,3,8-trihydroxy-6-methyl anthraquinone. It occurs in free and combined states in rhubarb, cascara rumex. Physcione is 1,8-dihydroxy-3-methoxy-6-methyl anthraquinone. Other anthraquinones present in free and combined states are aloe emodin and rhein[6].

Till date the qualitative estimation of anthraquinone glycosides on 23 *Cassia* species has been carried out but no systematic work has been undertaken to find out the exact amount of anthraquinone derivatives in each plant part of *Cassia* species. Therefore, the present work is carried out to see the quantitative distribution of emodin, chrysophanol and physcione within these five *Cassia* species.

The plant parts like bark, flower, leaf, pericarp, seed and stem-wood of *C. auriculata, C. fistula, C. javanica, C. roxburghii* and *C. siamea* have been collected when each and every part of the species got matured. The plant parts were then air dried, powdered and used for estimation.

A TLC-colourimetric method has been employed to estimate the same. The free anthraquinone derivatives produced pink red or violet colour with aqueous ammonia

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