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Comparative Antimicrobial Activity of Aerial Parts of *Melothria maderaspatana* of Indian and Srilankan Origin

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Abstract: Soil & Environment is among of the major factors that affects the activity of medicinal plants. In some cases there may be gross change in activity were observed. Hence an attempt has been made to check the antimicrobial activity of *Melothria madraspatana* obtained from India and Srilanka by cup plate method. The hexane, chloroform, ethyl acetate and methanol fraction from both Indian and Srilankan species were tested against there gram negative bacilli (*E.coli, Pseudomonas aeruginosa* and *Klebsiella pneumonia*) and two gram positive organism (*Streptococcus* and *Pneumonia*) and towards two fungi ie. *Candida albicans* and *Aspergillus niger* at 200µg/ml, 400µg/ml and 800µg/ml concentrations. The hexane fraction of Indian variety showed pronounced activity at 800µg/ml where as the Srilankan variety showed negligible activity against *Staphylococcus.* The chloroform and ethyl acetate fraction of both varieties showed negligible activity, where as methanol fraction of Srilankan variety showed pronounced activity against *Staphylococci aureus*. Both varieties showed negligible activity against *Klebsiella* and *E.coli*. The methanolic extract of Srilankan variety showed pronounced activity against *E.coli.* When compared to standard Ciprofloxacin. The Indian variety showed mild antibacterial activity against *Pseudomonas* whereas the Srilankan variety showed negligible activity. Both the varieties showed pronounced antibacterial activity against *Streptococc*i. The antifungal activity was found to be negligible for both the varieties. Ciprofloxacin 5µg was used as standard for anti bacterial activity and clotrimazole 100µg was used as standard for antifungal studies.
Key words: Antimicrobial activity , *Pseudomonas aeruginosa* , *Klebsiella pneumonia, Melothria madraspatana.*

INTRODUCTION

Melothria maderaspatana (Linn.) Cogn. Belongs to the family Cucurbitaceae. The plant is a tendril climber / prostate herb, used in the treatment of asthma and respiratory infections[1]. It is distributed through out the India and Srilanka. The earlier reports showed that it posses anti-inflamatory, hepatoprotective[2] and anti-rheumatic activities[3]. The tender shoots and bitter leaves were used as a gentle aperients and prescribed in vertigo and biliousness. The roots of the plant when masticated relieve toothaches. It is distributed through out the India and Srilanka.

Since the antimicrobial activity has not been reported, an attempt has been made to find the antimicrobial activity of both the Indian and Srilankan variety in both bacteria and fungi. Two gram positive bacteria (*Streptococci pyogens, Staphylococci aureus*) and three gram negative bacteria (*E.coli, Pseudomonas,* and *Klebsiella*) and two fungi *(Candida albicans, Aspergillus niger*) were used for the study.

The preliminary phytochemical screening[4] shows the presence of alkaloids, flavonoids, triterpenes and glycosides in the aerial parts of *Melothria maderaspatana.*

MATERIALS AND METHODS

COLLECTION OF PLANT MATERIAL

The aerial parts of Indian varierty of *Melothria maderaspatana* was obtained from herbal garden, Sri Ramachandra Medical College & Research Institute and authenticated in the Pharmacognosy department, SRMC & RI, porur, Chennai. A herbarium has been deposited, in the department of Pharmacognosy No: 021 Pharma. The Srilankan variety was obtained from Colombo and authenticated by Dr. Ira Thabrew, senior professor, University of kelaniya, Srilanka and a herbarium is deposited at Department of Pharmacognosy, SRMC & RI, No:022 Pharma.

PREPARATION OF PLANT EXTRACTS

The fresh plants were collected in the month of July from India and Srilanka, thoroughly dried in shade after separating aerial parts from roots for a period of 2 weeks. The dried plant material was made into coarse powder (yield 750 g). The powder was subjected to maceration for 72 hrs, followed by exhaustive maceration for 48 hrs by various solvents of increasing polarity (n-hexane, chloroform, ethyl acetate and methanol), the solvents were filtered pooled together and recovered by distillation. The extracts were dried under desiccators and percentage yields were determined. The percentage yield of n-hexane, chloroform, ethylacetate and methanol extract of Indian variety was 0.33%w/w, 0.43%w/w, 1.12%w/w, 2.07%w/w respectively. Preliminary phytochemical screening carried out in both Indian and Srilankan variety [5].

ANTIBACTERIAL ACTIVITY

The anti-bacterial studies were carried out aspetically under in-vitro conditions by "Cup plate method"[6]. The authentic bacterial cultures were inoculated in nutrient broth over night and used. Strains of *Streptococci* were cultured in blood agar media and used. The sterile nutrient agar media at 40-50°C was transferred aseptically to sterile Petri plates and allowed to solidify. The bacterial cultures were then inoculated by swabbing techniques. Bores of 8mm diameter were made on the bacterial seeded agar. The various fractions of the plant extracts were dissolved in DMSO, so as to contain 200, 400 and 800 µg/ml of the drug. 100 µl of each drug solution were added to the respective cups for each organism, along with the solvent control DMSO& standard ciprofloxacin (5µg/ml). The bacteria seeded agar plates were aseptically transferred to incubator and incubated at 37°C for 18-24 hrs of incubation and compared with standard antibiotic, Ciprofloxacin. The extract which shown a zone of inhibition above 12mm were considered for minimum inhibitory concentration by double dilution method. The observations were tabulated in Table-1.

ANTI-FUNGAL ACTIVITY

The anti-fungal studies were carried out on Sabourauds Dextrose Agar (SDA) of Hi-media laboratories.

Inoculation of fungi

The fully grown fungal mat from the fresh cultures of fungi, *Candida albicans* and *Aspergillus niger* were used for inoculation. A loop full of fungi from the SDA slant was inoculated into 100µl of sterilized SDA medium and shaken well. It was then poured into sterile Petri plates and allowed to solidify. With the help of sterile cork bores, 8mm diameter wells were cut out on the fungi seeded SDA medium. 100µl of various fractions of the plant extracts in different concentrations were aspetically added, along with the standard drug Clotrimazole (100 µg/ml) and solvent control DMSO with the help of finn pipettes. The plates were maintained at room temperature for a period of 48 hrs. The diameter of zone of inhibition was measured at the end of 48 hours [7].

RESULTS AND DISCUSSION

The hexane fraction of Indian variety showed pronounced activity at 800 µg/ml where as the Srilankan variety showed negligible activity against *Staphylococci aureus*. Both the varieties showed negligible activity against *Klebsiella*. The methanolic extract of Srilankan variety showed pronounced activity against *E.coli* than Indian variety and the values are comparable with that of standard Ciprofloxacin. The Indian variety showed mild antibacterial activity against *Pseudomonas* where as the Srilankan variety showed negligible activity. The antifungal activity was found to be negligible for both the varieties. The phyto-constituents like triterpens, flavonoids, and alkaloids may be responsible for antimicrobial activity[8].

The hexane, ethyl acetate and methanol fraction of both the varieties were equally potent against *streptococci*, whereas the chloroform extract of Srilankan variety showed well pronounced antibacterial action than Indian variety.

CONCLUSION

It is concluded that methanolic extract of the Srilankan variety was found to be effective, and hexane extract of Indian variety showed moderate antibacterial activity.

Table 1: Effect of *melothrio maderaspatana* of varying concentration on *staphylococcus*

Indian variety					Srilankan variety			
Concentration µg/ml	H	C	EA	M	H	C	EA	M
200	19	9	-	-	-	-	-	12
400	21	9	-	-	-	-	-	18
800	22	10	9	-	-	-	-	22
Ciprofloxacin Standard	34	34	34	34	34	34	34	34
Solvent	-	-	-	-	-	-	-	-

Table 2: Effect *of melothrio maderaspatana* of varying concentration on *klebsiella pneumoniae*

Indian variety					Srilankan variety			
Concentration µg/ml	H	C	EA	M	H	C	EA	M
200	-	-	9	-	-	-	-	-
400	-	-	10	-	-	-	-	-
800	9	-	10	-	-	-	-	-
Ciprofloxacin Standard	18	18	18	18	18	18	18	18
Solvent	-	-	-	-	-	-	-	-

Table 3: Effect *of melothrio maderaspatana* of varying concentration on *streptococci*

Indian variety					Srilankan variety			
Concentration µg/ml	H	C	EA	M	H	C	EA	M
200	12	10	10	10	10	14	-	10
400	14	12	10	16	12	16	-	14
800	16	14	11	18	14	26	14	16
Ciprofloxacin Standard	32	32	32	32	32	32	32	32
Solvent	-	-	-	-	-	-	-	-

Table 4: Effect of *melothrio maderaspatana* of varying concentration on *E-coli*

Indian variety					Srilankan variety			
Concentration µg/ml	H	C	EA	M	H	C	EA	M
200	-	9	-	-	-	-	-	22
400	-	9	-	-	-	-	-	24
800	-	9	-	-	-	-	-	26
Ciprofloxacin Standard	23	23	23	23	23	23	23	23
Solvent	-	-	-	-	-	-	-	-

Table 5: Effect of *melothrio maderaspatana* of varying concentration on *pseudomonas aerugenosa*

Indian variety					Srilankan variety			
Concentration µg/ml	H	C	EA	M	H	C	EA	M
200	11	12	12	10	-	-	-	-
400	13	12	13	12	-	-	-	-
800	13	13	14	12	-	-	-	-
Ciprofloxacin Standard	21	21	21	21	21	21	21	21
Solvent	-							

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