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EVALUATION OF ANTIBACTERIAL ACTIVITY OF *CUSCUTA REFLEXA* ROXB.

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ABSTRACT

In the present study antibacterial activity of methanolic extract of stem of *Cuscuta reflexa* Roxb. was evaluated against Gram positive bacteria like *Bacillus subtilis, Staphylococcus aureus* and Gram negative bacteria like *Escherichia coli, Pseudomonas aeruginosa*. Extract was poured into the wells of sterile Mueller hinton agar plates and antibacterial activity was determined by agar diffusion assay. The plates were observed for the inhibition of bacterial growth that was indicated by a clear zone around the wells. The size of the zones of inhibition was measured and the antibacterial activity was expressed in terms of average diameter of the zone of inhibition in millimeters. The results showed remarkable inhibition of growth for the tested organisms. More activity was observed against Gram negative bacteria (*Pseudomonas aeruginosa*) as compared to Gram positive bacteria.

KEYWORDS: Antimicrobial Activity, *Cuscuta reflexa* Roxb., Methanolic Extract, Zone of Inhibition.

INTRODUCTION

Cuscuta reflexa belongs to the family convulvulaceae and is commonly known as Amarvela (Sanskrit), Dodder (English) and Akasbel (Hindi).[1] Other names include hell weed, devil's gut, beggar weed, strangle tare, scald weed, dodder of thyme, greater dodder, and lesser dodder. The plant is distributed throughout India, Ceylon and Malaya. It grows in a prolific manner over host plants or other support with intertwined stems, giving it a common name of Devils Hair. The plant is leafless and rootless. It has no chlorophyll and cannot make its own food by photosynthesis. Initially the starter plant would have had some roots. Within a few days of germination, the plant, which is touch sensitive, finds a host or dies. After establishing itself on a host body, it draws nutrition from the host as a stem parasite and the roots wither away. The twining stem develops Haustoria which are root like and penetrate the host stem to draw water and nourishment. The flowers are small, white, having a perfect bell shape and a fleshy calyx, attached directly to the stem nodes. *Cuscuta reflexa* has been investigated for antispasmodic, haemodynamic, bradycardia,[2] antisteroidogenic,[3] antihypertensive, muscle relaxant, cardiotonic,[4] psychopharmacological[5] and antiviral and anticonvulsant[6] activities. It has long history of ethnomedicinal use. Many chemical constituents have been isolated from *Cuscuta reflexa* such as cuscutin, amarbelin, myricetin, quercitin, coumarin and oleanolic acid.[7] The plant is bitter, acrid, and hence useful as aphrodisiac and in alterative and bolius disorder. The seeds are used as carminative and purgative. The juice of the plant used as anthelmintics, purifies the blood. Fruit decoction is used in cough and fever. The medicinal properties of the plant are attributed as purgative, used in flatulence and liver complaints and externally for itch.[8] Stem shows antimicrobial activity.[9] The stem is thread like filaments which begin to grow and attach themselves to nearby host plants. The plant lives its entire life without attachment to the ground. In the present study methanolic extract of stem of *Cuscuta reflexa* Roxb. was studied for its antibacterial activity against *Bacillus subtilis,*

Staphylococcus aureus, *Escherichia coli* and *Pseudomonas aeruginosa*..

MATERIALS & METHODS

Sample collection and processing

Stem of *Cuscuta reflexa* Roxb. was collected from Forest Research Institute, Dehradun, Uttarakhand, India, in the month of September 2015.

Bacterial strains

Bacterial strains used in this study were *Bacillus subtilis* (MTCC 441), *Staphylococcus aureus* (MTCC 3160), *Escherichia coli* (MTCC 614) *and Psudomonas aeruginosa* (MTCC 2453). All the strains were purchased from Institute of Microbial Technology, Chandigarh.

Solvent extraction

The fresh stem sample was washed under running tap water, dried at room temperature for 30 days and homogenized to a fine powder in the grinder. 250 gm powder was extracted with 80% methanol using soxhlet extractor. Extract was filtered through Whatman filter paper. Filtrate was dried and preserved at 4^{o}C in dark, air tight bottles. The yield of methanolic extract of stem of *Cuscuta reflexa* was found to be 10.9% w/w of the dry plant.

Antibacterial activity

To determine the antibacterial activity of methanolic extract of the *Cuscuta* stem, disc diffusion assay was carried out. Different concentrations of plant extracts were prepared in the order of 50µg/ml, 100µg/ml, 150µg/ml and 200µg/ml in methanol. Methanol was used as negative control. Penicillin at the concentration of 100µg/ml served as positive control. 6mm ditch was made into the Mueller hinton agar media with the help of sterile cork borer. Different concentrations of methanolic extract was loaded into the wells and the plates were incubated at 37^{o}C for 24 hours. Zone of inhibition was indicated by the clear area around the wells which showed no bacterial growth. Antibacterial activity was determined by measuring the inhibition zone diameter around the wells and reported as zoe of inhibition (in mm) ± SEM (Standard error mean).

RESULT & DISCUSSION

Antibacterial activity of C*uscuta reflexa Roxb.* stem extract against Gram positive bacteria (*Bacillus subtilis* and *Staphylococcus aureus*) and Gram negative bacteria (*Escherichia coli and Pseudomonas aeruginosa*) was studied and the results are summarized in table 1, figure 1 to 4 and Graph 1. The present study was designed to evaluate the antibacterial activity of methanolic extract of stem of Cuscuta reflexa Roxb. Antibacterial activity of Cuscuta reflexa has also been confirmed in other research work.[9,10] When tested with disc diffusion method, Cuscuta reflexa Roxb. showed significant activity against all the four tested bacterial strains i.e., Bacillus subtilis, Staphylococcus aureus, Escherichia coli and Pseudomonas aeruginosa. More activity was observed against Gram negative bacteria as compared to Gram positive bacteria..

CONCLUSION

Methanolic extract of stem of *Cuscuta reflexa* Roxb. was used for the study. By the present study it was concluded that *Cuscuta reflexa* Roxb. possesses good antibacterial activity.

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Table 1: Antibacterial activity of methanolic extract of stem of *Cuscuta reflexa* Roxb.

Bacterial strains	**Zone of inhibition (mm±standard error mean)**				
	50 µg/ml	**100 µg/ml**	**150 µg/ml**	**200 µg/ml**	**Std (100µg/ml)(Penicillin)**
Bacillus subtilis	6.5±0.00	8.2±0.77	9.1±0.90	9.8±1.05	4.90±0.09
Staphylococcus aureus	7.1±0.07	8.8±0.79	9.5±1.15	10.5±0.99	7.39±0.35
Escherichia coli	6.7±0.01	8.4±0.55	9.6±1.02	10.2±1.86	8.69±0.40
Pseudomonas aeruginosa	7.9±0.82	9.7±1.04	11.2±1.58	11.9±1.97	10.90±1.50

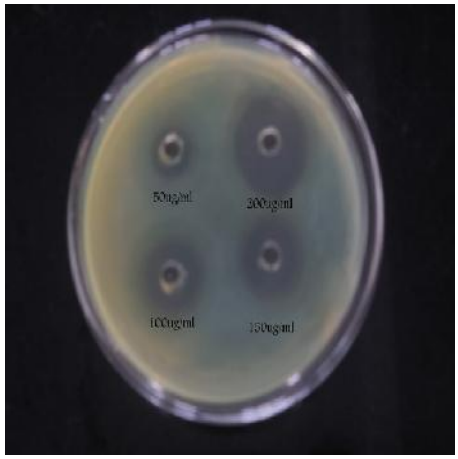

Figure 1: Antibacterial activity of methanolic extract of *C. reflexa* Roxb. stem *a*gainst *B. subtilis*

Figure 2: Antibacterial activity of methanolic extract of *C. reflexa* Roxb. stem against *S. Aureus*

Figure 3: Antibacterial activity of methanolic extract of *C. reflexa* Roxb. stem against *E. coli*

Figure 4: Antibacterial activity of methanolic extract of *C. reflexa* Roxb. stem against *P. aeruginosa*

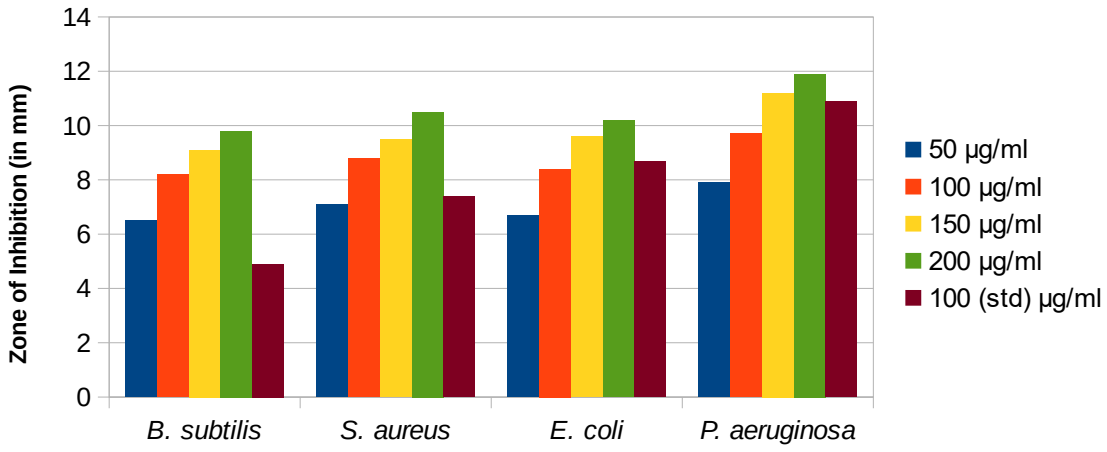

Figure 5: Antibacterial activity of methanolic extract of stem of *Cuscuta reflexa* Roxb.

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