IN VITRO ANTIMICROBIAL ACTIVITY OF CUSCUTA REFLEXA ROXB.
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ABSTRACT
Antimicrobial agents are effective in the treatment of infections because of their selective toxicity that is they have the ability to injure or kill an invading microorganism without harming the cells of the host. Cuscuta reflexa Roxb. (Convolvulaceae) is parasitic plant. It is used as anthelmintic, carminative, purgative, constipation, aphrodisiac, alterative in different disorder. Ethanolic extract of Cuscuta reflexa Roxb. were prepared by soxhlet extraction method. The qualitative and quantitative, macroscopic, microscopic, phytochemical, physicochemical, analysis of plant has been studied. Antimicrobial activity of stems of cuscuta reflexa Roxb were studied using ethanolic extract against Gram positive bacteria like Bacillus subtilis, staphylococcus aureus and Gram negative bacteria like Escherichia coli, Pseudomonas aeruginosa as well as on some fungal strains like Penicillium citrium, Aspergillus niger using standard (Penicillin). Sterile nutrient agar plates were prepared by using Pour plate & Spread plate method. The ethanolic extracts were poured into the wells of sterile nutrient agar medium using agar cup plate method. The results were analysed by using zone of inhibitions and it was observed that Gram negative and Fungal strains showed more antimicrobial activity as compared to the Gram positive bacteria.

KEY WORDS: Cuscuta reflexa Roxb., ethanolic extract, antimicrobial activity.

INTRODUCTION
Cuscuta reflexa Roxb Belongs to the family convolvulaceae. It is growing throughout India. The plant mainly abundant in Bengal plains, Western ghat, Ceylon, Satara region (M. S.), Himachal Pradesh. It is leafless green yellowish and thread like twinning herb. It has no root under the ground but only grown as parasitic twine on other plants hence it is known as akaswel (sky twinner) or amarbel (Immortal twine).In English it is known as Dodder. The plant is bitter, acidic, and hence useful in aphrodisiacs, alterative and the bolius disorder. The seeds used as carminative, purgative. The juice of the plant used as anthelminitics, purify the blood. Fruit decoction is used in cough and fever. Stem is useful in constipation, flatulence, liver complaints. Stem shows antimicrobial activity.

MATERIALS AND METHODS
Plant Collection and Authentication
Fresh stem of Cuscuta reflexa Roxb. was collected from pune municipal corporation region shivaji nagar, pune (M.S.), India in month of October 2009. Its taxonomic identities and authenticated by Agharkar Research Institute, Agharkar Road, Pune. The plant were wash under the running tap water, air dried and then homogenized to fine powder and store in air tight bottle.

Preparation of Extracts
The 500g of the powder were successively extracted with ethanol using soxhlet extractor. The ethanolic extract was vacuum dried in a rotary vacuum evaporator. The dark green extract obtained was kept in refrigerator for further use. Extractive yield of ethanolic extract of stem were found to be 12.3% w/w of dry plant.

Test Microorganisms
The bacterial strains are identified strains and procured from National Chemical Laboratory (NCL) Pune, (M.S.) India for antimicrobial susceptibility testing. The microorganisms are Bacillus subtilis (NCIM 2063), Staphylococcus aureus (NCIM 2079), Escherichia coli (NCIM 2065), Pseudomonas aeruginosa (NCIM 2109), Penicillium citrium (NCIM 765), Aspergillus niger (NCIM 1196)
All strains were maintained, preserved on Muller-Hinton agar slant throughout the antimicrobial study.

Antimicrobial Assay
In vitro antimicrobial activity of stem extract of Cuscuta reflexa Roxb. was studied by agar cup plate technique. The sterilized media (nutrient agar media for bacteria and sabroaud agar for fungal strain) were poured into the
Petri plates. After medium was solidified, ditch was made into the petri plate with the help of sterile cork borer (6mm). The different concentrations of Ethanolic extract 50,100,150,200(µg/ml) was made using DMSO solvent, which was loaded into the respective wells and incubated at 37°C for 24 hrs for bacteria and 25°C-30°C for 72 hrs for fungi. Penicillin was used as a positive control. The experiment was performed under aseptic condition. Inhibition of microbial growth determined by measuring diameter of inhibition as shown in Table 1.

**RESULTS AND DISCUSSION**

Antimicrobial activity of *Cuscuta reflexa* Roxb. against Gram positive bacteria (*Bacillus subtilis* and *Staphylococcus aureus*), Gram negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*), Fungi (*Penicillin citrium* and *Aspergillus niger*), was done and results summarized in tabular form in table 1 and Figures.

**CONCLUSION**

Ethanolic extract of stem of *Cuscuta reflexa* Roxb. showed the presence of alkaloids, carbohydrates, some glycosides, flavonoids, tannins, phenolic compounds, steroids. Flavonoid glycosides show antimicrobial activity. At the end it is concluded that plant *Cuscuta reflexa* Roxb. possesses antimicrobial activity. The extract was used for antimicrobial activity. In this Fungi and Gram negative bacteria shows more antimicrobial activity than Gram positive bacteria.

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**REFERENCES**


| Table 1: Anti microbial Activity of *Cuscuta reflexa* Roxb. |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| **Species**     | **Concentration of Extract In(µg/ml)** | **Zone of inhibition in mm** |
| **50**          | **100**         | **150**         | **200**         | **Std(100)**(Penicillin) |
| *S.Aureus*      | 7.1             | 7.5             | 8.6             | 9.5             | 14.1            |
| *Bacillus Subtilis* | 7.6             | 8.6             | 9.2             | 10.1            | 14.9            |
| *E.Coli*        | 8.2             | 8.4             | 8.9             | 9.7             | 16.5            |
| *P.Aeruginosa*  | 9.3             | 9.6             | 9.7             | 10.4            | 13.2            |
| *P.Citrium*     | 9.4             | 9.6             | 9.9             | 10.7            | 16.7            |
| *A.Niger*       | 9.6             | 9.9             | 10.4            | 11.2            | 15.6            |
Anti-Microbial Activity

Fig. 2 Anti-bacterial activity of Ethanolic extract of Cuscuta reflexa Roxb. Against *S. Aureus*

Fig. 3 Anti-bacterial activity of Ethanolic extract of Cuscuta reflexa Roxb. Against *B. Subtilis*

Fig. 4 Anti-bacterial activity of Ethanolic extract of Cuscuta reflexa Roxb. Against *E. Coli*

Fig. 5 Anti-bacterial activity of Ethanolic extract of Cuscuta reflexa Roxb. Against *P. Aeruginosa*

Fig. 6 Anti-fungal activity of Ethanolic extract of Cuscuta reflexa Roxb. Against *P. Citrium*

Fig. 7 Anti-fungal activity of Ethanolic extract of Cuscuta reflexa Roxb. Against *A. Niger*

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