

**ANTIBACTERIAL ACTIVITY OF *CUSCUTA REFLEXA*  
(CONVOLVULACEAE) AGAINST HUMAN PATHOGENIC  
MICROBIAL STRAINS**

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Article Received on  
31 July 2015,

Revised on 20 Aug 2015,  
Accepted on 09 Sep 2015

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### ABSTRACT

*Cuscuta reflexa* is a parasitic plant. It is used as a purgative, constipation, alternative in different disorder. Antimicrobial agent are in the treatment of infections because of their selective toxicity that is they have ability to kill an microorganism without harming the cells of the host. Antibacterial activity of leaves of *Cuscuta reflexa* were studied using ethanolic extract against Gram positive bacteria like *Streptococcus mutants*, *Staphylococcus aureus* and Gram negative bacteria like *Escherichia coli*, *Klebsiella pneumonia*, *Solmonella typhi*, *Proteus mirabilis*, *Staphylococcus aureus*. Using Standard antibiotic tetracycline. Antibacterial activity was tested using a modification of the disc diffusion method. The etahanolic extract were poured into the

well of sterile nutrient agar medium using agar cup plate technique. The result were analysed by using zone of inhibitions and it was observed that Gram negative and Gram positive strains showed more antibacterial activity as compared to the Gram positive bacteria.

**KEY WORDS:** *Cuscuta reflexa*, Antimicrobial activity, Ethanolic extract, Microorganism.

### INTRODUCTION

*Cuscuta reflexa* is a rootless, leafless perennial parasitic twining herb of Convolvulaceae family, commonly known as Akashvalli (Sky Twinner), Amarbel (Immortal Twine), or Dodder in English. The plant is distributed worldwide and in India about 6 species are found. It has no chlorophyll and cannot make its own food by photosynthesis. The twining stem

develops haustoria which are root like and penetrate the host stem to draw water and nourishment. The flowers are small and white, having a perfect bell shape and a fleshy calyx, attached directly to the stem nodes. It grows on thorny or other shrubs, sometimes completely covering the bushes and trees. The Juice of *Cuscuta reflexa* Internally useful in appetizer, digestive, liver stimulant, anthelmintic and reduces intestinal motility. Externally useful in inflammation, pain, hair disorder, conjunctivitis and also used against itch and other skin diseases. The Whole plant is used as a wash for sores. Stem is Useful in bilious disorders. Fruits is Used in fever and cough. The Seed used in Cold infusion is given as a depurative and carminative in pains and stomach-aches.

## MATERIAL AND METHODS

### Plant material

*Cuscuta reflexa* plant leaves were collected from G.B. Pant University of Ag. & Technology, Pantnagar, U.K India.

### Preparation of aqueous extract

Hundred grams each of dried leaves of *C.reflexa* collected from different location of North India were macerated with 100 ml sterile distilled water in a blender for 10 min. The macerate was first filtered through double layered muslin cloth and centrifuged at 4000 rpm for 30 min. The supernatant was filtered through Whatman No.1 filter paper and heat sterilized at 120°C for 30 min. The extracts were preserved aseptically in brown bottles at 4°C until further use.

### Preparation of plant Solvent extracts

Soxhlet extraction will be the method used for plant extraction. A portion of dried leaves (100 g) of *Cuscuta reflexa* was placed in a Soxhlet apparatus. Extraction was performed with 500 ml of an appropriate solvent (Ethanol, Methanol, Chloroform) with increased polarity for 24 h at 95°C temperature not exceeding the boiling point of the solvent. The extract was filtered through a 45 µm filter paper and concentrated under vacuum. In this experiment three solvents were used: Ethanol, Chloroform and methanol. The resulting three solutions were concentrated in vacuum to dryness to give Ethanol (4 g), Chloroform extract (10 g) and methanol extract MeOHE (12 g). The stock solutions were kept at 4°C until further use.

### Test microorganism

The Bacterial strains are identified strain and procured from IMTECH, Chandigarh, India for antimicrobial susceptibility testing. The microorganisms are,

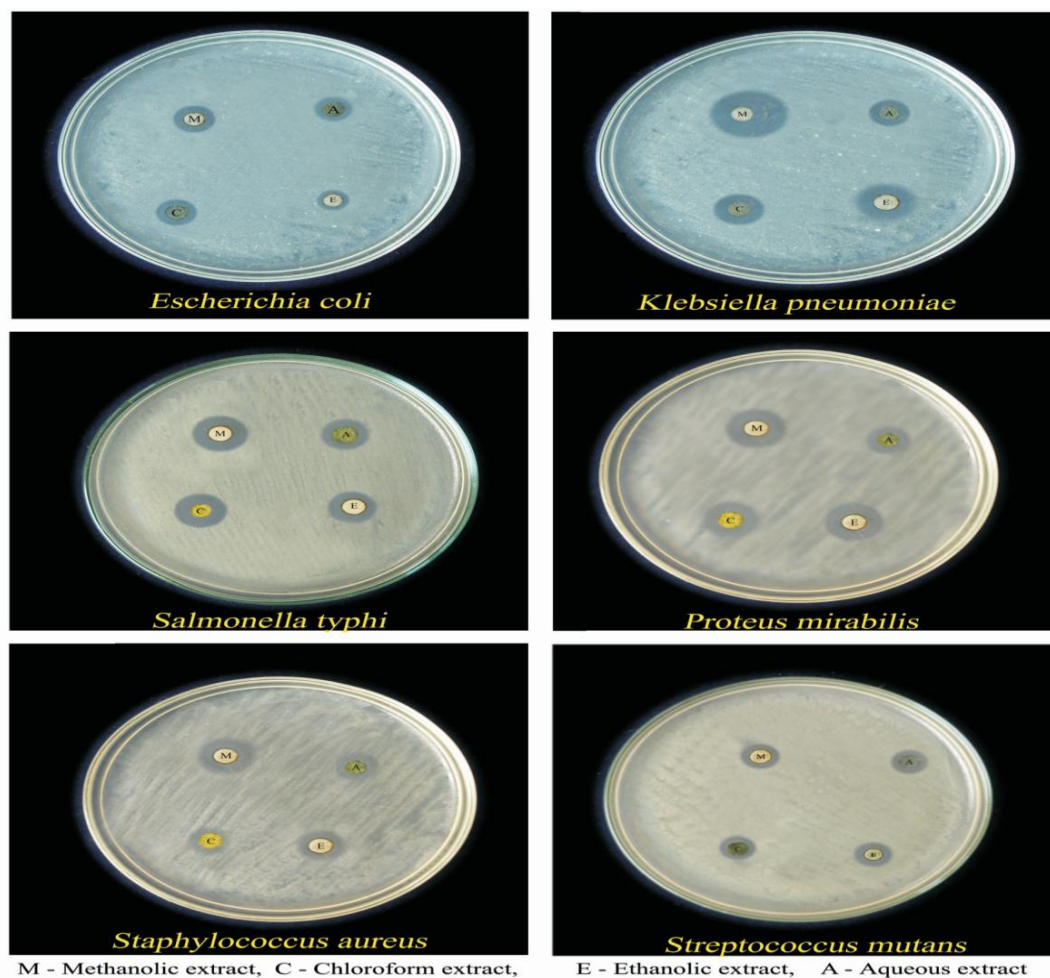
- Staphylococcus aureus* : Microbial Type Culture Collection  
(MTCC 389) IMTECH, Chandigarh, India.
- Solmonella typhi* : Microbial Type Culture Collection  
(MTCC 424) IMTECH, Chandigarh, India.
- Escherichia coli* : Microbial Type Culture Collection  
(MTCC 40) IMTECH, Chandigarh, India.
- Klebsiella pneumonia* : Microbial Type Culture Collection  
(MTCC 1924) IMTECH, Chandigarh, India.
- Proteus mirabilis* : Microbial Type Culture Collection  
(MTCC 0425) IMTECH, Chandigarh, India.
- Streptococcus mutans* : Microbial Type Culture Collection  
(MTCC 0899) IMTECH, Chandigarh, India.

### Antibacterial Activity Assay

Antibacterial activity was tested using a modification of the disc diffusion method originally described by Bauer *et al.* (1966). A loop of bacteria from the agar slant stock was cultured in nutrient broth overnight and spread with a sterile cotton swap into petriplates containing 10 ml of Nutrient Agar. Sterile Whatman No.1 filter paper discs were (Whatman Ltd., England) (6mm in diameter) impregnated with the plant extract and placed on the culture plates and incubated at 25 or 37°C, depending on the bacteria. The solvent without extracts served as negative control. After 24 h of incubation, the diameter in mm of the inhibitory or clear zones (MIC) around the disks was recorded. Standard antibiotic tetracycline 30 mg (Span Diagnostics Limited, Surat, India) was used as reference or positive control.

### RESULT AND DISCUSSION

In this research Antimicrobial activity of *Cuscuta reflexa* were compared with control drug Tetracyclin against Gram positive bacteria like *Streptococcus mutans*, *Staphylococcus aureus* and Gram negative bacteria like *Escherichia coli*, *Klebsiella pneumonia*, *Solmonella typhi*, *Proteus mirabilis*, *Staphylococcus aureus* was done and results summarized in tabular form in table 1 and Graphs (1) Figure (1).



**Fig- 1: ANTIBACTERIAL ACTIVITY OF *Cuscuta reflexa*- Pantnagar sample Against Six types of Bacteria.**

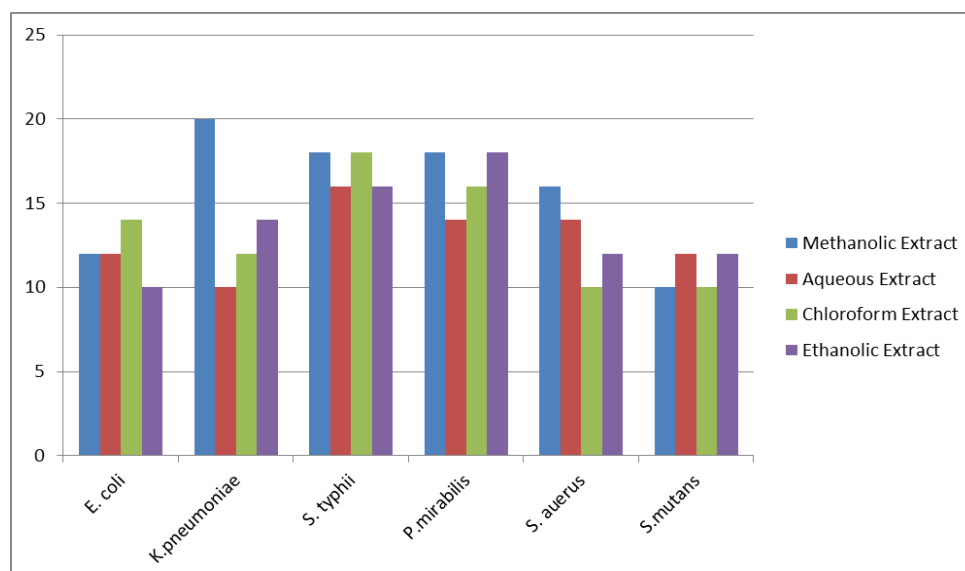
**Table-1. Antibacterial activity of different solvent extracts of *C. reflex* Pantnagar Sample against human pathogenic bacteria.**

Solvent	Zone of inhibition (mm)					
	<i>E. coli</i>	<i>K.pneumoniae</i>	<i>S. typhi</i>	<i>P.mirabilis</i>	<i>S. auerus</i>	<i>S.mutans</i>
Methanolic Extract	12	20	18	18	16	10
Aqueous Extract	12	10	16	14	14	12
Chloroform Extract	14	12	18	16	10	10
Ethanolic Extract	10	14	16	18	12	12

Controls - Inhibition zone against the bacteria by sterile distilled water =0 mm.

Inhibition zone against the bacteria by different solvent = 0 mm.

\*More effective than standard antibiotic (Tetracycline).



**Graph- 1. Graphical representation of Antibacterial activity of different solvent extracts of *C. reflexa* Pantnagar Sample against human pathogenic bacteria.**

## CONCLUSION

The results of this study indicate that the antibacterial activity of ethanolic extract of *Cuscuta reflexa* is most active. The two Gram-positive, four Gram-negative bacterial strains were used. According to the results in the table 1 and Graphs (1) Figures.(1), only methanolic, chloroform, ethanolic and aqueous extracts of *Cuscuta* samples showed antibacterial activity against Gram positive bacteria like *Streptococcus mutans*, *Staphylococcus aureus* and Gram negative bacteria like *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella typhi*, *Proteus mirabilis*, *Staphylococcus aureus*.

## ACKNOWLEDGEMENT

The authors thankful to Dr. Anju Pal Dept. of Horticulture, G.B.Pant University of Agriculture. And Technology, Pantnagar, U.K, India and the Ex. Dean Dr. D.P Mishra and Director Dr. S.K Garg Department of Science and Humanities G.B. Pant University of Ag. & Technology, Pantnagar, U.K. And also thanks to Dr. Krishan Pal Dept. of Biotechnology, Shri Venkateshwara University, Gajraula, and U.P.India for providing necessary laboratory requirement, facilities to carry out this work and useful discussion and suggestion.

## REFERENCES

1. Ben Hsouna A, Trigui M, Ben Mansour R, Mezghani Jarraya R, Damak M, Jaoua S: Chemical composition, cytotoxicity effect and antimicrobial activity of *ceratonia siliqua*

- essential oil with preservative effects against *Listeria* inoculated in minced beef meat. *Int J Food Microbiol*, 2011; 148: 66-72.
2. Gulluce M, Sahin F, Sokmen M, Ozer H, Daferera D, Sokmen A, Polissiou M, Adiguzel A, Ozkan H: Antimicrobial and antioxidant properties of the essential oils and methanol extract from *menthe longifolia* L. ssp. *Longifolia*. *Food Chem*, 2007; 103: 1449-1456.
  3. Guven K, Yucel E, etintaj F: Antimicrobial activities of fruits of *crataegus* and *pyrus* species. *Pharm Biol*, 2006; 44: 79-83.
  4. Hussain AI, Anwar F, Shahid M, Ashraf M, Przybylski R: Chemical composition, antioxidant and antimicrobial activities of essential oil of spearmint (*mentha spicata* L.) from Pakistan. *Journal Essential Oil Research*, 2010; 22: 78-84.
  5. Sartoratto A, Machado ALM, Delarmelina C, Figueira GM, Duarte MCT, Rehder VLG: Composition and antimicrobial activity of essential oils from aromatic plants used in Brazil. *Braz J Microbiol*, 2004; 35: 275-280.
  6. Shojaee-Aliabadi S, Hosseini H, nMohammadifar MA, Mohammadi A, Ghasemlou M, Mahdi Ojagh S, Marzieh Hosseini S, Khaksar R: Characterization of antioxidant-antimicrobial Carrageenan films containing *satureja hortensis* essential oil. *Int J Biol Macromol*, 2012; In Press.
  7. Iwu, W.M., A.R. Duncan and C.O. Okunji, New antimicrobials of plant origin In Janick J. (ed.). *Perspectives on new crops and new uses*. ASHS Press, Alexandria, VA., 1999; 457-462.
  8. Prabuseenivasan, S., M. Jayakumar and S. Ignacimuthu, *In vitro* antibacterial activity of some plant essential oils. *BMC Complementary and Alternative Medicine*, 2006; 6(1): 39.
  9. Yi, Z., Y. Yu, Y. Liang and B. Zeng, *In vitro* antioxidant and antimicrobial activities of the extract of *Pericarpium Citri reticulatae* of a new Citrus cultivar and its main flavonoids. *LWT*, 2008; 41: 597-603.
  10. Cristani, M., M. d'Arrigo, G. Mandalari, F. Castelli, M.G. Sarpietro and D. Micieli,
  11. Interaction of four monoterpenes contained in essential oils with model membranes: Implications for their antibacterial activity. *J. Agric. Food Chem.*, 2007; 55(15): 6300-6308.
  12. Baratta, M.T., H.J.D. Dorman, S.G. Deans, D.M. Biondi and G. Ruberto, Chemical
  13. composition, antimicrobial and antioxidative activity of laurel, sage, rosemary, oregano and coriander essential oils. *J. Essent. Oil Res.*, 1998; 10: 618-627.
  14. Pal DK, Mandal M, SenthilKumar GP, Padhiari A: Antibacterial activity of *Cuscuta reflexa* stem and *Corchorus olitorius* seed. *Fitoterapia*, 2006; 77: 589-591.

15. Inderjit, K.G. Mukerji, (Eds.) Allelochemicals: Biological Control of Plant Pathogens and Diseases Series: Disease management of Fruits and Vegetables, 2006; 2: 214.
16. Inamdar Faiyyaz B, Oswal Rajesh, Chorage Trushal V, Garje Kapil : In vitro Antimicrobial activity of *Cuscuta reflexa* Roxb, 2011; 214-216.