



ANTIBACTERIAL ACTIVITY OF *ALSTONIA SCHOLARIS*: AN *IN VITRO* STUDY

Jai Bahadur Singh Kachhawa¹, Neha Sharma¹, Swati Tyagi¹, Radhey Shyam Gupta², Krishna Kumar Sharma^{1*}

¹ Molecular Developmental Biology Laboratory, Department of Zoology, M.D.S. University, Ajmer-305 009, Rajasthan, India.

² Center for Advanced Studies, Department of Zoology, University of Rajasthan, Jaipur-302 004, Rajasthan, India.

*Corresponding author's E-mail: kksmds@gmail.com

Accepted on: 23-10-2011; Finalized on: 20-01-2012.

ABSTRACT

Alstonia scholaris a potent therapeutic plant was used in the present study to evaluate the antibacterial activity against Gram-positive bacteria i.e. *Bacillus coagulans* and gram negative bacteria i.e. *Escherichia coli*. Ciprofloxacin was used as standard drug, and methanol extract of *Alstonia scholaris* bark at different concentrations were used for the experimental evaluations, where inhibition zones were calculated after disc diffusion assay for their antibacterial activity. Calculations of inhibition zones proved that *Alstonia scholaris* is a persuasive inhibitor against both the bacteria.

Keywords: *Alstonia scholaris*, ciprofloxacin, methanol extract, *Bacillus coagulans*, *Escherichia coli*.

INTRODUCTION

Finding healing powers in plant is an ancient idea. The increasing interest on the traditional ethno medicine may lead to the discovery of novel therapeutic agents. Many of the plant species have been documented pharmacologically and clinically in the world which are endowed with phytochemicals with marked activity on human pathogenic bacteria. The present study was carried out on the phytochemical and antibacterial activity of bark of *Alstonia scholaris*.

MATERIALS AND METHODS

Plant materials

Alstonia scholaris is belonging to family Apocynaceae, commonly known as Blackboard tree, Indian devil tree and White cheesewood in English; Saptparna in Hindi and Vishamachhada in Sanskrit. It is an evergreen, tropical tree native to the Indian subcontinent. It is a well known remedy for the treatment of various types of disorders in the ayurvedic, homoeopathic and folklore system of medicine in India.^{1,2}

Different parts of this plant are used in traditional medicines.³ The bark is used for its tonic bitter and astringent properties; it is particularly useful for chronic diarrhoea and dysentery, indigestion and typhoid. It acts as galactagogue, stomachic, laxative and liver tonic. Jahan *et al* demonstrates the chemopreventive potential of *Alstonia scholaris* bark extract in DMBA-induced skin tumorigenesis in Swiss albino mice.⁴ Hadi and Bremner tested *A. scholaris* for antimalarial properties. *A. scholaris* showed significant luteolytic and anti-implantational effect in rats.⁵ *A. scholaris* showed molluscicidal and anti-cholinestase activity against freshwater snail *Lymnaea acuminata*.⁶

Moreover, a lot of work has been done on its phytochemical investigations. Chatterjee *et al.* studied

the alkaloids in the leaves of *A. scholaris*. A new indole alkaloid, alstonamine and a sibiricine type indole alkaloid, rhazimanine, also have been isolated from the leaves of *A. scholaris*.⁷ Qualitative tests of *Alstonia scholaris* methanol extract done by Khyade and Vaikos suggested that plant is rich in iridoids, alkaloids, coumarins, flavonoids, phlobatannin, reducing sugars, simple phenolics, saponins and tannins.⁸ Banerji and Banerji reported that a large number of alkaloids, steroids and triterpenoids are present in *A. scholaris*.⁹

Preparation of plant extracts

Bark of *A. scholaris* were collected from the campus of University of Rajasthan, Jaipur and a specimen sample was identified and submitted to the Department of Botany, University of Rajasthan, Jaipur. Barks of the plant were washed with tap water and shade dried for about 15 days. These dried barks were then powdered and extracted with 100% methanol solvent. The filtrates were evaporated under reduced pressure to get a thick residue. This residue treated as experimental drug for the present study.

Test microorganism

The antimicrobial activity was individually tested against Gram-positive bacteria i.e. *Bacillus coagulans* and Gram-negative bacteria i.e. *Escherichia coli*. Both test strains were maintained on nutrient agar (Hi-media Laboratory Pvt. Ltd., Mumbai, India) and were sub-cultured every two weeks. The bacteria *B. coagulans* was obtained from Sporlac tablets and *E. coli* was procured from Institute of Microbial Technology, Chandigarh, India.

Antibacterial activity assay

The disc diffusion method was adopted to test the antibacterial activity where Ciprofloxacin was used as a standard drug to compare the results of experimental plant.



Paper disc diffusion method

The disc diffusion method was used to determine the growth inhibition of bacteria by the plant extracts. Discs containing different concentration (200, 100, 50 and 25 mg/ml) of dissolved plant extract and prepared by using sterile Whatman filter paper No. 1 (6 mm in diameter). The discs were dried at 50°C. Overnight cultures of each of bacterial isolates was diluted with sterile normal saline to give inoculum size of 10⁶ cfu/ml. Nutrient agar medium was prepared, sterilized, cooled and poured in to sterile petri dishes to a depth of 4 mm about 25 ml/plate to solidify. Pure cultures of the test organism were used to inoculate the petri dishes. This was done by spreading the inoculum on the surface of the prepared nutrient agar plate using sterile cotton swabs which have been dipped in the diluted suspension of the organism. The discs were then aseptically placed evenly on the surface of the inoculation and gently pressed down to ensure contact using a pair of forceps. The plates were finally incubated at 37°C for 18-24hrs. The plates were examined after 24 hrs for clear zone of inhibition. All measurements were taken in mm.

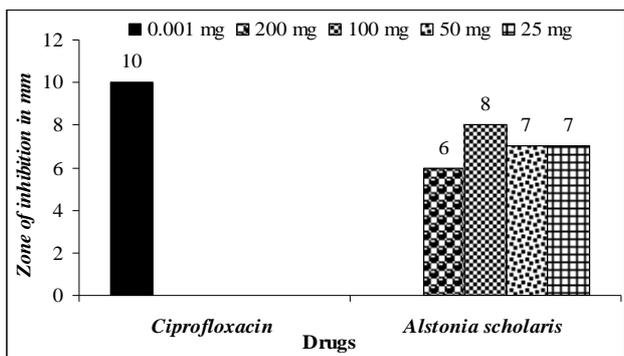


Figure 1: Antibacterial activity of different concentrations of methanol extract of *Alstonia scholaris* against *B. coagulans*

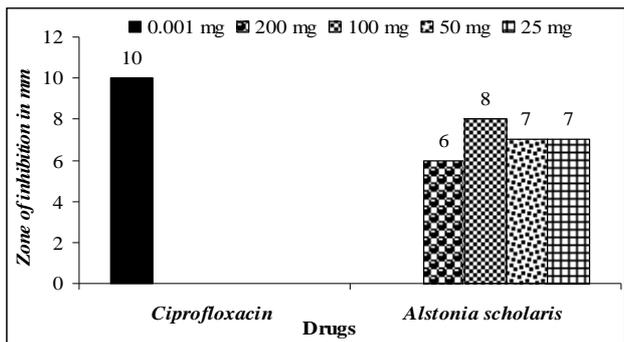


Figure 2: Antibacterial activity of different concentrations of methanol extract of *Alstonia scholaris* against *Escherichia coli*

RESULTS

Results obtained for the antibacterial test performed on different solvent extract of *Alstonia scholaris* are presented in Figure 1 and 2. Among the various extracts

tested methanol extract showed broader spectrum of antibacterial activity being active to both gram positive and gram negative organisms. 100mg/ml concentration showed maximum antibacterial activity against *Bacillus coagulans* and *Escherichia coli* with MIC of 8mm. 25mg and 50mg concentration also showed antibacterial activity against the test bacteria with a MIC of 7mm.

DISCUSSION

A Lot of work had been done by various scientist on the phytochemical properties of *A. scholaris*^{7,10} Banerji and Banerji⁹ have reported that various alkaloids, flavanoids, tannins and saponins are present in the leaves of *Alstonia scholaris*, which could be responsible for its bactericidal effects. The difference in the observed activity of various concentration of extract (25mg, 50mg, 100mg) may be due to varying degree of activity of compounds found in the bark of the experimental plant.

CONCLUSION

In the current investigation the methanol extract of the *A. scholaris* bark was found to be active on test bacteria. Demonstration of antibacterial activity of *A. scholaris* against the test bacteria is a possible indication of newer antibacterial agents.

Acknowledgement: Author Dr. Jai Bahadur Singh Kachhawa, acknowledges the financial assistance from UGC, New Delhi for Dr. D.S. Kothari Postdoc Research Fellowship.

REFERENCES

- Nadkarni AK, Indian Materia Medica, Popular Prakashan, Bombay, 1976.
- Joshi SG, Medicinal plants, Oxford and IBH publishing Co. Pvt. Ltd., New Delhi, 2000.
- Chopra RN, Chopra IC, Handa KL, Kapur LD, Chopra's indigenous drugs of India. Dhur, U.N. & Sons Pvt Ltd Calcutta, 2nd ed., 1958, pp 26.
- Jahan S, Chaudhary R, Goyal PK, Anticancer activity of an Indian medicinal plant, *Alstonia scholaris*, on skin carcinogenesis in mice, Integrative Cancer Therapy, Vol-8(3), 2009, 273-279.
- Choudhary DN, Singh JN, Singh BP, Effect of some medicinal plants on fertility of albino rats, Indian Journal of Pharmacology, Vol-23, 1991, 253-257.
- Singh SK, Singh A, Molluscicidal and anti-cholinestarse activity of *Alstonia scholaris* plant against freshwater snail *Lymnaea acuminata*, Pakistan Journal of Biological Sciences, Vol-6(16), 2003, 1442-1446.
- Rahman A, Alvi KA, Indole Alkaloids from *Alstonia scholaris*, Phytochemistry, Vol-26(7), 1987, 2139.
- Khyade MS, Vaikos NP, Phytochemical and antibacterial properties of leaves of *Alstonia scholaris* R. Br., African Journal of Biotechnology, Vol-8(22), 2009, 6434-6436.
- Banerji A, Banerji J, Isolation of pseudoakummigine from the leaves of *Alstonia scholaris* R. Br., Indian Journal of Chemistry, Vol-15, 1977, 390-391.
- Chatterjee A, Mukherjee B, Ray AB, Das B, The alkaloid of the leaves of *Alstonia scholaris* R. Br., Tetrahedron Letters, Vol-41, 1965, 3633-3637.

