

# ANTIBACTERIAL ACTIVITY OF *HYGROPHILA SPINOSA* T. ANDERS LEAVES – A COMPARATIVE STUDY

Arjun Patra<sup>1\*</sup>, S. Jha<sup>2</sup>, P.N. Murthy<sup>3</sup>, S. Satpathy<sup>3</sup>

<sup>1</sup>College of Pharmacy, IFTM, Moradabad - 244 001, UP, India

<sup>2</sup>Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra, Ranchi – 835 215, Jharkhand, India

<sup>3</sup>Royal College of Pharmacy and Health Sciences, Berhampur - 760 002, Orissa, India

\*E-mail: arjun.patra@rediffmail.com; Mob: 09761459749

**ABSTRACT :** *Hygrophila spinosa* (Acanthaceae) is a potent medicinal plant in the Indian systems of medicine. Traditionally the leaves are used as/in diuretic, jaundice, antibacterial, dropsy, rheumatism, anasaraca, diseases of urinogenital tract, leucor, sweet, sour, bitter, tonic, oleaginous, aphrodisiac, hypnotic, diarrhea, dysentery, urinary calculi, urinary discharge, anti-inflammatory, joint pain, biliousness, eye disease, ascites, abdominal troubles, anemia, anuria, gleets, cough, demulcent, stomachic, lumbago, arthritis, gastric disorder, leucorrhoea. In the present study the hydroalcoholic extract of the leaves of *H. spinosa* collected during different time periods were studied for their antibacterial activity by disc-diffusion method against different gram positive (*Staphylococcus aureus* & *Bacillus subtilis*) and gram negative (*Escherchia coli* & *Pseudomonas aeruginosa*) bacteria. It was observed that the leaves collected during mid-September to October end showed significant activity compared to the extracts of leaves collected at some other time.

**KEYWORDS:** *Hygrophila spinosa*, disc-diffusion method, antibacterial activity, minimum inhibitory concentration.

## INTRODUCTION

*Hygrophila spinosa* T. Anders (Acanthaceae) is described in Ayurvedic literature as Ikshura, Ikshugandha and Kokilasha “having eyes like Kokila or Indian cuckoo”, common in moist places on the banks of tanks, ditches, paddy fields etc.; widely distributed throughout India from Himalayas to Ceylon, Srilanka, Burma, Malaysia and Nepal <sup>1, 2, 3</sup>. Traditionally the leaves are used as/in diuretic, jaundice, dropsy, rheumatism, anasaraca, diseases of urinogenital tract, leucor, sweet, sour, bitter, tonic, oleaginous, aphrodisiac, hypnotic, diarrhea, dysentery, urinary calculi, urinary discharge, anti-inflammatory, joint pain, biliousness, eye disease, ascites, abdominal troubles, anaemia, anuria, gleet, cough, demulcent, stomachic, lumbago, arthritis, gastric disorder, leucorrhoea <sup>1, 2, 4, 5, 6</sup>. Leaf contains proteins, nitrogen, polyphenols <sup>7</sup>, minerals as Ca, Mg, K, Fe, Cu, Zn, Mn, Co & Cr <sup>8</sup>, glycosides, reducing sugars <sup>9</sup>, acacetin, proanthocyanins, and phenolic acid <sup>6</sup>. Present study deals with the antibacterial activity of the leaves collected at different times.

## EXPERIMENTAL

### COLLECTION AND AUTHENTICATION OF PLANT MATERIAL

The leaves of *Hygrophila spinosa* were collected from the campus of Royal College of Pharmacy and Health Sciences, Berhampur and authenticated through Regional Research Laboratory, Bhubaneswar and a voucher specimen has been preserved for further reference.

### PREPARATION OF EXTRACTS

The leaves were collected during different times like august to mid September, mid September to October end and November to mid December. The leaves were dried under shade, coarsely powdered and the hydroalcoholic extracts were prepared separately by maceration.

### ANTIBACTERIAL ACTIVITY

The antibacterial activity was evaluated by disc-diffusion method <sup>10, 11</sup>. The bacterial strains used were *Escherchia*

*coli* (NCIM NO. 2341), *Staphylococcus aureus* (NCIM NO. 2654), *Bacillus subtilis* (NCIM NO. 2195) and *Pseudomonas aeruginosa* (NCIM NO. 2914). Nutrient agar media was taken in a pre-sterilized Petri-dish and the microorganisms were grown. The extracts were dissolved separately in distilled water and used in the concentration of 100µg/disc in triplicate, placed in petri dishes and incubated at 37°C for 24 hrs. The diameters of zone of inhibition (mm) were recorded and compared with standard drug ofloxacin. The minimum inhibitory concentration (MIC) of the different extracts was also determined according to standard method<sup>12</sup>.

## RESULT AND DISCUSSION

The minimum inhibitory concentration of the different extracts was shown in figure. 1. It was observed that all the three extracts showed antibacterial activity. The extract of the leaves collected during mid-September to October end showed significant antibacterial activity in comparison to the other two extracts, where the activity are almost similar (Table. 1). Further, the hydroalcoholic extract was found to contain alkaloids, glycosides, tannins, flavonoids, reducing sugars, proteins, sterols etc. through preliminary photochemical screening. The antibacterial activity may be due to one/more group of above phytoconstituent(s) and the plant may contain higher amount of antibacterial constituents during mid-September to October end.

**Table 1: \*Observation on Determination of Zone of Inhibition of Different Extracts of *H. spinosa***

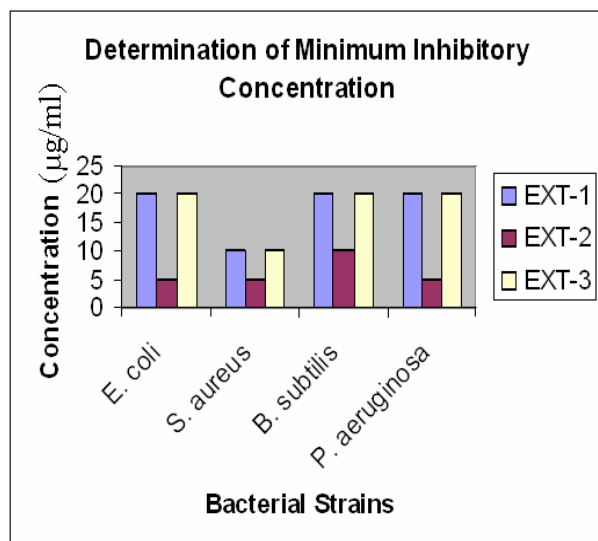
Test Organism	Different Extracts (100µg/disc)			Ofloxacin (5µg/disc)
	EXT-1	EXT-2	EXT-3	
<i>E. coli</i> (NCIM 2341)	10	16	9	23
<i>S. aureus</i> (NCIM 2654)	13	16	12	25
<i>B. subtilis</i> (NCIM 2195)	8	14	7	23
<i>P. aeruginosa</i> (NCIM 2914)	9	15	10	26

\*Average of three readings

EXT-1 = Leaves collected during august to mid-September.

EXT-2 = Leaves collected during mid-September to October end.

EXT-3 = Leaves collected during November to mid-December.



\*Average of three readings

EXT-1 = Leaves collected during august to mid-September.

EXT-2 = Leaves collected during mid-September to October end.

EXT-3 = Leaves collected during November to mid-December.

**Figure 1: \*Determination of Minimum Inhibitory Concentration of Different Extracts of *H. spinosa***

## REFERENCES

1. Nadkarni A.K., Indian Materia Medica, Vol. I, Popular Prakashan, Mumbai, 2007, 668-669.
2. Kirtikar K.R. and Basu B.D., Indian Medicinal plants, Vol. III, International Book Distributors, 2005, 1863-1865.
3. Chopra R.N., Nayar S.L. and Chopra I.C., Glossary of Indian Medicinal Plants, NISCAIR, CSIR, New Delhi, 2006, 29.
4. Atal C.K. and Kapur B.M., Cultivation and Utilization of Medicinal Plants, Regional Research Laboratory, CSIR, Jammu-Tawi, 1982, 548-549.
5. Sharma P.C., Yelne M.B. and Dennis T.J., Database on Medicinal Plants Used in Ayurveda. Vol. 4, Central Council for Research in Ayurveda and Siddha, New Delhi, 2002, 320-331.
6. Daniel M., Medicinal Plants: Chemistry and Properties, Science Publishers, Texas, 2005, 193.
7. Dewanji A., Chanda S., Si L., Barik S. and Maiti, S., Extractability and nutritional value of leaf protein from tropical aquatic plants, Plant food Human Nutr. 1997, 50, 349-357.
8. Kar A., Choudhary B.K. and Bandyopadhyay N.B., Important mineral contents and medicinal properties of *M. oleifera* and *H. spinosa*, Sachitra Ayurveda 1998, 50(7), 543-549.
9. Samy R.P., Antimicrobial activity of some medicinal plants from India, Fitoterapia 2005, 76, 697-699.
10. Mali R.G., Hundiware J.C. and Sonawane R.S., Evaluation of *Capparis decidua* for Anthelmintic and Antimicrobial Activities, Ind. J. Nat. Prod. 2004, 20(4), 10-13.
11. Anonymous, Indian Pharmacopoeia, Volume II, The Controller of Publications, Delhi, 1996, 100-106.
12. Walker T.S., Microbiology, W.B. Saunders Company, Philadelphia, Pennsylvania, 1998, 80.

\*\*\*\*\*