ANTIBACTERIAL ACTIVITY OF THE EXTRACT FROM THE AERIAL PARTS OF HOLOPTELEA *INTEGRIFOLIA* ROXB.

[1]BENJAMIN JEYA RATHNA KUMAR P AND [2]GHATHIKUMAR S

[1]PG and Research Department of Plant Biology and Biotechnology,
[2]Centre for Biodiversity and Biotechnology,
St. Xavier's College (Autonomous), Palayamkottai-627 002, Tamil Nadu, India.
Email:benjamin02kumar@rediffmail.com[1], ghanthi@gmail.com[2]

Summary

The search for biologically active compounds from natural sources has always been of great interest to researchers looking for new sources of drugs useful in infectious diseases. The aromatic and medicinal plants represent enormous reservoir of potential microbiocidal compounds that are alternative to synthetic microbicides. In the present investigation the leaf and stem powder extracts of four different solvents and water extract of *Holoptelea integrifolia* were tested against ten different randomly selected bacteria by disc diffusion method. It was found that the methanol and benzene leaf extract was strongly effective against all the chosen bacteria.

Key words: *Antibacterial study, Holoptelea integrifolia*

Introduction

Naturally occurring compounds in higher plants that possessed antimicrobial activity they contained some active substances in their extracts which have an inhibitory action towards bacteria and fungi. The use of plant extracts as antimicrobial substance has gained popularity in recent years which in mainly due to the presence of alkaloids and steroids present in the plant extracts. The *Holoptelea integrifolia* belongs to family Ulmaceae. It is commonly known as Indian Elm Tree. The plant has several medicinal properties. The decoction of the bark is externally used in rheumatism [1] intestinal tumors [7] and oxytocic in pregnant ladies [10]. Decoction of the leaves is used to regulate fat metabolism [11] treat ringworm eczema and cutaneous diseases [8]. Paste of the stem bark is externally applied to treat the inflammation of lymph glands, common fever [9] and ringworm and scabies. Stem bark acts as an anti-inflammatory agent specifically for eyes [6]. Bark and leaf paste of the plant are applied externally on the white patches or leucoderma [4].

Materials and Methods

The specimen plant for the study was collected from Singikulam hills near Palayamkottai. The taxonomic features collected from the species have been confirmed with the 'Flora of Presidency of Madras [3] and the 'Flora of the Tamilnadu Carnatic[5]. It was subjected to the antibacterial studies.

Extraction of antibacterial compounds

The plant parts were dried in shade and ground well to make fine powder. About 30 gm of powdered plant material was extracted successively with 200 ml of Petroleum ether, Benzene, Chloroform, Methanol and Water by Soxlet apparatus method. This sequence of solvents allows for leaching all compounds based on their polarity. The individual fractions were collected and concentrated to obtain crude extracts. The above solvents were diluted and the final concentrations of 5-10 mg/ml of solvents were used for bacterial bio-assay.

Microorganisms

The different bacterial strains such as *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Streptococcus pyogenes, Salmonella typhii, Serratia marcescens, Klebsiella pneumoniae, Enterobacter aerogenes, Proteus vulgaris* and *Bacillus subtilis* are collected from the Department of Microbiology, Sri. Paramakalyani College of Arts and Science, Azhwarkurichi. The medium used for antibacterial tests was Muller Hinton (MH) agar (Himedia Laboratories Pvt. Ltd, India). Each organism was maintained in a respective culture medium and was recovered for testing by sub culturing on a fresh media.

Assay of antibacterial activity

Antibacterial activity was assayed by filter paper disc diffusion method[2]. Whatman No. 1 filter paper of 5 mm diameter was used. These discs were sterilized before use. Each sterile disc incorporated individually with 500 µl of different plant extracts and dried. 0.5 ml of the dilute microbial culture was spread on sterile Muller Hinton Agar plates. The dried discs were placed on the seeded plates and gently pressed down to assure contact with the medium. Streptomycin 10 mg/ml was used as positive control and respective solvents which were used to dissolve the crude extracts served as negative control. The plates were incubated at room temperature for 24 hrs. After the incubation period the diameter of the inhibition zone around the discs were measured and recorded. Three replicates for each concentration were maintained.

Observations and Results

Leaf

The petroleum ether leaf extracts showed the antibacterial activity against *Escherichia coli, Pseudomonas aeruginosa, staphylococcus aureus, Salmonella typhii, Serratia marcescens, Proteus vulgaris* and *Bacillus subtilis.* The chloroform leaf extracts showed the antibacterial activity against *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Salmonella typhii, Serratia marcescens, Klebsiella pnuemoniae, Proteus vulgaris* and *Bacillus subtilis.* The benzene leaf extracts showed the antibacterial activities against all the testing bacteria. The methanol leaf extracts showed the antibacterial activities against all, except *Escherichia coli* and *Bacillus subtilis.* The distilled water leaf extracts showed the antibacterial activity against *Salmonella typhii, Serratia marcescens* and *Proteus vulgaris.*

The maximum inhibitory zone reported against *Streptococcus pyrogenes* (17mm) of methanol leaf extract and *Serraria marcescens* (17 mm) of benzene leaf extract. The minimum inhibitory zone reported against *Streptococcus pyrogenes* (5 mm) of benzene leaf extract, *Salmonella typhii* (5 mm) of distilled water leaf extracts and *Enterobacter aerogenes* (5 mm) of benzene leaf extract. Control (Streptomycin) produced inhibitory zone against all the chosen bacteria. The results are shown in Table 1.

Stem

The petroleum ether stem extracts showed the antibacterial activity against only *Salmonella typhii.* The chloroform and benzene stem extracts showed the antibacterial activity against *Streptococcus pyrogenes* and *Salmonella typhii*. In the methanol stem extracts showed the antibacterial activity against *Salmonella typhii, Proteus vulgaris* and *Bacillus subtilis.* The distilled water stem extracts showed did not have any antibacterial activity against any testing bacteria.

The maximum inhibitory zone reported against *Serratia marcescens* (6 mm) of methanol stem extract. The minimum inhibitory zone (3mm) reported the petroleum ether and chloroform stem extracts against *Salmonella typhii* and methanol stem extracts against *Proteus vulgaris* and *Bacillus subtilis.* Control (Streptomycin) produced inhibitory zone against all the chosen bacteria. The results are shown in Table 1.

TABLE 1: **ANTIBACTERIAL ACTIVITY OF** *HOLOPTELEA INTEGRIFOLIA* ROXB.

Sl. No	Name of the bacteria	Diameter of inhibition zone (mm)										
		Petroleum ether		Benzene		Chloro form		Methanol		Distilled water		Control
		Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	
1	*Escherichia coli*	6	-	10	-	11	-	-	-	-	-	9
2	*Pseudomonas aeruginosa*	9	-	15	-	12	-	19	-	-	-	8
3	*Staphylococcus aureus*	6	-	9	-	10	-	12	-	-	-	3
4	*Streptococcus pyrogenes*	-	-	-	5	5	-	17	-	-	-	16
5	*Salmonella typhii*	6	3	9	3	10	6	10	6	5	-	13
6	*Serratia marcescens*	12	-	14	-	17	-	12	-	8	-	7
7	*Klebsiella pnuemoniae*	-	-	6	-	8	-	8	-	-	-	8
8	*Enterobacter aerogenes*	-	-	-	-	5	-	12	-	-	-	5
9	*Proteus vulgaris*	10	-	12	-	13	3	15	3	7	-	15
10	*Bacillus subtilis*	6	-	11		9	3	-	3	-	-	6

References

1. Bajpai, A, Ojha, J.K. and Sant, H.R. Medico botany of the Varanasi District. Int J Pharmacogy, 1995; 33:172-176.
2. Bauer, A.W., Kirby, W.M., Sherris, J.C. and Turk, M.. Antibiotic susceptibility by a standardized single disk method. Amer.J.Clin.Pathol,1966; 45: 493 – 496.
3. Gamble, J.S.,. Flora of the Presidency of Madras. Volume 3, 1928; 1324 - 1325.
4. Maheswari, J. K. and Harcish Singh. Herbal remedies of Bhoxas of Nainital district, UP. Aryavaidyan, 1990; 4:30-34.
5. Matthew, K.M..Flora of the Tamilnadu Carnatic. 1981; 242-243.
6. Mudgal, V. and Pal, D.C.. Medicinal plants used by tribals of Mayurbhanj (Orissa). Bull Bot SurvIndia,1980; 22:59-62.
7. Sabnis, S.D. and Bedi, S.J.. Ethnobotanical studies in Dadra- Nagar Haveli and Daman. Indian J For,1983; 6: 65-69.
8. Sharma, M.P, Ahmad, J, Hussain, A. and Khan, S. Folklore medicinal plants of Mewat (Gurgaon District), Haryana, India. Int J Pharmacog, 1992; 30: 135-137.
9. Singh, V.K. and Ali, Z.A.. Folk medicines in primary health care: common plants used for the treatment of fevers in India. Fitoterapia, 1994;65: 68-74.
10. Tiwari, V.J and Padhye, M.D.. Ethnobotanical study of Gond tribe of Chandrapur and Gadchiroli districts of Maharashtra State, India. Fitoterapia, 1993; 64: 58-61.
11. Trivedi, V.P. and Mann, A.S.. Vegetable drugs regulating fat metabolism in Caraka (Lekhaniya Dravyas). Q J Crude Drug Res,1972;12: 1988.