Short Communication

Antibacterial activity of *Boenninghausenia albiflora* Reichb. (Rutaceae)

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Various organic and aqueous extracts of aerial part of *Boenninghausenia albiflora* (Rutaceae) obtained by infusion and maceration were screened for their antimicrobial activity against eight animal and plant pathogenic bacteria (*Bacillus subtilis*, *Staphylococcus aureus*, *Erwinia chrysanthemi*, *Escherichia coli*, *Proteus vulgaris*, *Xanthomonas phaseoli*, *Agrobacterium tumefaciens* and *Xanthomonas campestris*) using disc diffusion method. Out of total 32 tests performed, 27 tests showed positive antibacterial activity at 1000 μ g/ml concentration, whereas 15 instances exhibited zone of inhibition \geq 10 mm at same concentration. The activity shown by some of the extracts was found higher than ampicillin (10 mcg) and erythromycin (15 mcg), standard antibiotic used.

Key words: Boenninghausenia albiflora, Rutaceae, antibacterial, ampicillin, erythromycin.

INTRODUCTION

As a result of indiscriminate use of antimicrobial drugs in the treatment of infectious diseases, microorganisms have developed resistance to many antibiotics (Cowan, 1999). There is a need to develop alternative antimicrobial drugs. One approach is to screen local medicinal plants, which represent a rich source of novel antimicrobial agents.

Boenninghausenia albiflora belonging to the family Rutaceae and is well known for its medicinal properties in traditional system of medicine. In ethnobotanical literature, the aerial as well as the root part has been described as an antiseptic. Gaur (1999) mentioned that leaf part has been used to apply on cuts and wounds whereas root powder is being used as antiseptic. Sometimes its juice is also being given in vomiting and dysentery. Some workers also reported this plant to have flea repellent (Sood et al., 1966), as well as calcium blocking activity (Yamaha et al., 1987).

The present study was carried out to investigate the antibacterial properties of *B. albiflora* extracted by four solvents of different polarity. As no previous records on the antimicrobial activity of this plant could be found in

the literature, the present study claims some useful results.

MATERIALS AND METHODS

B. albiflora Reichb. (Rutaceae) aerial parts were collected in October, 2004 from Nainital, India and authenticated by Dr. YPS Pangtey, Department of Botany of the University. A voucher specimen was deposited in the herbarium of the Department.

Aerial part of the plant was powdered in an electric grinder. Fine powdered plant materials were subjected serially to hexane, chloroform, methanol and water. After extraction, each extract was passed through Whatman filter paper No.1. The filtrate was concentrated on a rotary evaporator under vacuum at 20 $^{\circ}{\rm C}$ and stored at 4 $^{\circ}{\rm C}$ for further use.

Microorganisms (Escherichia coli MTCC No.40 (G-ve), Bacillus subtilis MTCC No.121 (G-ve), Proteus vulgaris MTCC No.426 (G+ve), Staphylococcus aureus MTCC No. 87 (G+ve), Agrobacterium tumefaciens MTCC No. 609 (G-ve), Xanthomonas campestris MTCC No. 2286 (G-ve) were obtained from the Institute of Microbial technology, Chandigarh, India and Erwinia crysanthmi (G-ve) and Xanthomonas phaseoli (G-ve) were obtained from plant pathology department, G.B. Pant University of Agriculture, Pantnagar, India.

The disc diffusion method (Bauer et al., 1966; Cruickshank, 1968) was used to evaluate the antibacterial activity. Muller Hinton agar was prepared in the plate as the media for the test microorganisms. Sterile filter paper discs (whatman No.1, 5 mm) were impregnated with each of the extract to give a final concentration of 1000 µg/ml.

The bacteria were streaked in radial pattern on the agar plate. After inoculation plates were allowed to dry at room temperature for

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Microorganism	Inhibition zone (mm) ^a						
	Н	С	M	W	G	E	Α
Gram-positive						•	
Bacillus subtilis	12	-	16	8	12	7	7
Staphylococcus aureus	10	11	9	16	16	10	8
Gram-negative							
Erwinia crysanthmi	-	8	11	-	9	7	7
Escherichia coli	14	9	17	10	14	11	9
Proteus vulgaris	12	9	16	12	15	9	-
Agrobacterium tumefaciens	-	7	9	8	18	14	10
Xanthomonas campestris	6	7	10	9	10	8	8
Xanthomonas phaseoli	-	10	8	10	12	8	7

Table 1. Antibacterial activity of *Boenninghausenia albiflora* extracts at 1000 μg/ml.

H, C, M, W = Hexane, chloroform, methanol and water extracts, respectively.

Standards: G = Gentamycin (10 mg), E = erythromycin (15 mg), A = ampicillin (10 mg).

30 min. Gentamycin (10 mg), erythromycin (15 mg) and ampicilin (10 mg) were used as positive control. Respective solvents were used as the negative control.

Each extract was analyzed in triplicate. All the plates were incubated for 24 h at $37\,^{\circ}\text{C}$. The antibacterial activity was interpreted from the size of the diameter of zone of inhibition measured to the nearest (mm) as observed from the clear zone surrounding the disc.

RESULTS AND DISSCUSSION

At 1000 μ g/ml concentration, 27 tests out of total 32 tests (Table 1) showed positive antibacterial activity with only methanol extracts exhibiting activity against all the tested strains. *S. aureus* and *E. coli* and *P. vulgaris* were found sensitive against all the tested extracts whereas *E. crysanthmi* was found most resistant among all the tested bacterial strains. The activity shown by soe of the extracts was found higher than ampicillin (10 mcg) and erythromycin (15 mcg), standard antibiotic used.

In general antibiotics agents are rather selective in their inhibitory action, most of them being inhibitory to Grampositive organisms. Unlike Gram-positive bacteria, the lipopolysaccharide layer along with proteins and phospholipids are the major components in the outer surface of Gram-negative bacteria (Burn, 1988). The outer lipopolysaccharide layer hinders access of most compounds to the peptidoglycan layer of the cell wall. In the present investigation (Table 1), all the extract inhibited the growth of all the tested microorganisms both of Gramnegative and Gram-positive bacteria and no influence was observed on growth inhibition by Gram reaction.

Though the earlier workers have investigated *B. albiflora* for its chemical composition and reported various terpenoids, alkaloids and coumarins (Gupta et al., 1970; Sood, 1972; Ohta and Miyazati, 1958; Miyazaki and Mihashi, 1964; Nayar et al., 1973; Talapatra et al., 1973; Bhan et al., 1973; Talapatra et al., 1975; Shibata and

Noguchi, 1977), but the present investigation is the first attempt to identify its antibacterial potentiality which might be due to the selective or synergistic action of various chemicals reported previously.

Relying upon the results obtained for its inhibitory effect on various microorganisms used indicates that the different extracts of *B. albiflora* aerial part could be useful for treating diseases of plants and animal. Therefore, study on the extracts of this plant can be recommended for the preparation of effective antimicrobials. The present study on broad-spectrum antimicrobial activity of plant justifies its known uses in dysentery and antiseptic agent in traditional medicine. However an extensive study is also needed to isolate and to identify the active components of plant.

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^{- =} Not active against tested micro-organism.

^aData are the average of three experiments.

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