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International Journal of Pharmaceutical & Biological Archives 2012; 3(2):391-393

ORIGINAL RESEARCH ARTICLE

Comparative Antibacterial study of Barleria prionitis Linn. Leaf Extracts

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Received 08 Jan 2012; Revised 01 Apr 2012; Accepted 08 Apr 2012

ABSTRACT

The purpose of this investigation was to find out the comparative antibacterial activity of ethanolic extracts, chloroform extract, petroleum ether extracts and column fraction of *Barleria prionotis* Linn. on various bacterial species. The different extracts of the plant were first prepared on basis of various concentration levels and then extensively applied on selected bacterial culture media for determination of minimum inhibitory concentration (mg/mL) and these extracts exhibited significant antibacterial activity. This comparative study taking with selected extracts has been demonstrated for the first time.

Key words: *Barleria prionitis*; antibacterial activity; Comparative study; Minimum Inhibitory Concentration (MIC).

INTRODUCTION

In recent time, traditional medicines have been brought a huge potential in herbal medicine with enormous therapeutic potential to heal many infectious diseases without associated with the side effects unlike synthetic drugs. Bacterial infections are one of the prominent causes of health problems, physical disabilities mortalities around the world. Symptoms and complications associated with bacterial infections such as fever, chills, headache, nausea, vomiting and organ failures affect patient's life severely. Medicinal plants are a rich source of antimicrobial agents and provide a safer and cost effective way of treating the bacterial infections. Barleria prionitis Linn. (Acanthaceae) is widely distributed throughout India, Sri Lanka, Africa and tropical Asia. The crude extract of this plant is commonly used in folk medicine to treat whooping cough. The plant extract has also shown its potential applications as diaphoretic and expectorant. The plant has also shown anti-respiratory syncytial virus, anti-arthritic, anti-inflammatory and antifertility activities. In Ayurveda the leaves and the tender branches are used for treatment of toothache, strengthening of gums, whooping cough and premature ejaculation. Whole-plant extracts of porcupine flower contain iridoid glycosides, barlerin, and verbascoside, which have shown potent activity against respiratory syncytial virus *in vitro* and may account for the plant's use in treating fever and several respiratory diseases in herbal medicine¹. Plants and plant products have been used extensively throughout history to treat medical problems. Extracts of many plants are highly efficient against parasitic as well as microbial infection. It is estimated that around 70,000 plant species, from lichens to tall trees, have been used at one time to other for medicinal purposes²⁻⁷. In this study, the various solvent extracts of *Barleria prionitis* plant were prepared and successfully determined their minimum inhibitory concentration on different bacterial species.

MATERIALS AND METHODS

The plant has been collected from the forest of Ichharia village, Bankura, West Bengal, India and has been authenticated by B.S.I., Shibpur, Howrah, West Bengal, India.

Preparation of Extract

The leaves were carefully washed under running tap water followed by sterile distilled water. These were air dried at room temperature (45°C) for five days and pulverized to a fine powder using a sterilized mixer grinder and stored in air-tight bottles. Then the leaf powder extracted with petroleum ether (40°-60°), chloroform and

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maceration was done with 70% ethanol. The column fraction (70% ethanol extract) was prepared by column chromatography. After filtration of total extracts, the extracts were evaporated to dryness in vacuum. The various media viz. solid media, liquid media and peptone water were prepared to conduct the study [8].

Determination of Minimum Inhibitory Concentration

Gram positive and gram negative bacteria were grown in peptone water for 18 h; this gave an optimum growth of the test bacteria. The petroleum ether (40°-60°C) extract, chloroform extract, 70% ethanol extract and column fraction separately dissolved in dimethyl sulfoxide (DMSO) sterilized by filtration by using sintered glass filter (G-5) and stored at 4°C [9].

Petroleum ether extract was added to molten nutrient agar in the following concentration (mg/mL): 3.33; 6.66; 16.66; 33.33. Chloroform extract and column fraction was then added to the nutrient agar in following concentration (mg/mL): 5; 10; 25; 50. Ethanol (70%) extract added to 9 ml molten nutrient agar in the following concentration (mg/mL): 10; 20; 50; 100 and poured into sterile petridish. The pH of the media was maintained at 7.2-7.4. The inoculums consisted of an over night grown broth culture of a bacterium diluted in such a manner

that a 2mm (internal diameter) loop full of that culture contain 10⁵ colony forming unit (CFU). These were then spot inoculated at the sterilized laminar air flow on nutrient agar plates containing increasing amount of a compound, inoculated at 37 ⁰C up to 72h for determination of the minimum inhibitory concentration (MIC) ^[9].

RESULTS

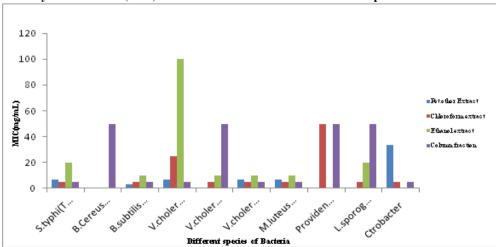
Different strain of isolated gram positive bacteria and gram negative bacteria were used for this study. After performing the antibacterial activity, it has been noticed that minimum inhibitory concentration (MIC) of the different Barleria prionitis Linn. (Acanthaceae) leaf extract was found to be 5 mg/mL for both Chloroform extract and Column fraction in Salmonella typhi, 50 mg/mL for Column fraction in Bacilus cereus, 3.33 mg/mL for Petroleum ether extract in *Bacilus* subtilis, 5 mg/mL for Column fraction in Vibrio cholerae (DN - 6), 5 mg/mL for Chloroform extract in Vibrio cholerae (793), 5 mg/mL for Chloroform extract and Column fraction in Vibrio cholerae (813), 5 mg/mL for Chloroform extract and Column fraction in Micrococcus luteus, 50 mg/mL for Chloroform extract and Column fraction in Providencia, 5 mg/mL for Chloroform extract in Lactobacillus sporogenus, 5 mg/mL for Chloroform extract and Column fraction Citrobacter shown in (Table 1).

Table 1: Comparative study of MIC among the all extract on different bacterial species

Name of Strains	Pet. Ether extract (mg/mL)	Chloroform extract (mg/mL)	Ethanol extract (mg/mL)	Column fraction (mg/mL)
Salmonella typhi	6.66	5	20	5
Bacilus cereus	N.A.	N.A.	N.A.	50
Bacilus subtilis	3.33	5	10	5
<i>Vibrio cholerae</i> (DN – 6)	6.66	25	100	5
Vibrio cholerae (793)	N.A	5	10	50
Vibrio cholerae (813)	6.66	5	10	5
Micrococcus luteus	6.66	5	10	5
Providencia	N.A.	50	N.A.	50
Lactobacillus sporogenus	N.A.	5	20	50
Citrobacter	33.33	5	N.A.	5

 $\overline{N.A.} = Not Active$

Fig 1: Minimum inhibitory concentration (MIC) of different extracts on various bacterial species



DISCUSSION

Antimicrobial activity in terms of Minimum inhibitory concentration (MIC) for chloroform extract (5 mg/mL) and Column fraction (5 mg/mL) were found to inhibit the growth of bacteria against highest number of selected bacterial species in comparison to others where as petroleum ether extracts of the plant showed least MIC (3.33 mg/mL) against the *Bacilus subtilis* shown in (**Fig 1**). The highest activity of chloroform extracts is supposed to have better permeability of chloroform across the cell membrane of bacterial species among the others.

CONCLUSION

Above results conclude that the bacterial activity were frequently inhibited by chloroform extract and Column fraction in a concentration in 5 mg/mL as MIC (**Fig 1**) and further study may be performed taking the chloroform extract and Column fraction of the plant *Barleria prionotis* Linn. as traditional medicine is still beneficial than synthetic drugs due devoid of side effects in herbal medicine.

ACKNOWLEDGMENT

The authors are thankful to the Principal and Management of Bengal College of Pharmaceutical sciences and research, Durgapur, West Bengal, India for their kind permission and help in conducting this research work.

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