

## Antibacterial Activity of Medicinal Plants against Pathogens from Extracts of *Achyranthes Aspera*

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### Abstract

In this present study various extracts of petroleum ether, methanol, ethanol, ethyl acetate and chloroform of medicinal plant *Achyranthes aspera* was evaluated for their antibacterial activities against multi drug resistance organisms such as *Pseudomonas aeruginosa*, *Proteus mirabilis* and *Enterococcus faecalis*. The organic extracts of both the leaf and stem parts of the plants at a concentration of 5 mg/ml and their activities were measured by estimating zones of inhibition as produced by antibiotic sensitivity method on Mueller-Hinton agar. The corresponding inhibitory zone was measured in Table 1. The results of this research support the use for further analysis in the treatment of infectious diseases such as urinary tract an infection caused by bacteria and has significant scope for antibacterial research.

**Keywords:** *Achyranthes aspera*; Antibacterial

### Introduction

Knowledge of herbs using from generation to generation for hundreds of years as part of traditional. It used for medicine because of easily available and inexpensive. Phytochemicals useful as drugs in terms of broad spectrum activity for many microorganisms that leads for treating microbial diseases. *Achyranthes aspera* is an important medicinal plant and all parts are used in medicines, seeds and roots are the most. The present research shows an updated information for pharmacological and anti-bacterial properties [1]. Nature has been an excellent source of phytochemical agents in the field of clinical microbiology [2]. Phytochemicals contain a wide range of components that is effective for bacterial diseases such as urinary tract infections. The use of plant extract for medical treatments is enjoying great popularity since 1990s. These phytochemical drugs are useful when people misuse of traditional antibiotics that cause drug resistance [3].

The antimicrobial activities of plant extracts consist many phytochemical components, including flavonoids and alkaloid compounds [4]. These extract having greater minimal antibacterial activity than actual individual components [5]. In this work, we have selected *Achyranthes aspera* (family *Amaranthaceae*; local name, apang). *A. aspera* is distributed throughout the tropical and subtropical regions, including the Indian sub-continent, Africa, Australia and America [6]. *Achyranthes aspera* commonly used in Bangladesh as herbal medicine. This study was conducted to address their antimicrobial activities against some pathogenic bacteria causing pneumonia, wound infections and other diseases, when extracted their leaf and stem parts in different organic solvents.

### Materials and Methods

#### Bacteria and growth conditions

Five bacterial species were employed as test organisms which include *Pseudomonas aeruginosa*, *Proteus mirabilis*, and *Enterococcus faecalis*. The bacteria were grown in Mueller-Hinton Agar (MH). Inocula were prepared by adding an overnight culture of the organism in MH broth to obtain an OD<sub>600</sub> 0.1. The cells were incubated to grow until they obtain the McFarland standard 0.5 (approximately 105-108 CFU/ml).

#### Test plants and their extraction

*Achyranthes aspera* was collected from the V-fort St. Lucia for the study. The leaves and stem parts of the plant *Achyranthes aspera* were separated, washed with sterile water, desiccated at room temperature and then made to powder. 10 g of the crushed powder is diluted with 45 ml of chloroform in a 250 ml beaker and was kept at 30°C for 10 h. The solution was filtered and dried by vacuum dryer at 45°C for 24 h to get the chloroform extract. After chloroform extraction, a part of the solid residue was dried at 40°C overnight to remove residual chloroform. The solid powder was resuspended in 40 ml ethyl acetate and kept at 25°C for 12 h. Ethyl acetate extract was recovered following the same procedure as stated for chloroform extract. Similarly, Petroleum ether, methanol and ethanol extracts were prepared by applying the same procedure. Finally, the extracted substance was resuspended in the organic solvents at a concentration of 100 mg/ml prior it was tested for the antimicrobial activity.

#### Determination of antibacterial activity

Sterile discs (Oxoid) were soaked separately with 50 µl of each of the organic extract prepared in chloroform, ethyl acetate, methanol and ethanol solvents, at a concentration of 100 mg/ml and then dried. These discs were placed on Mueller-Hinton agar plates, previously swabbed with the target bacterial isolate at a concentration of 105-108 CFU/ml. In one disc, the respective organic solvent was added as negative control to determine possible inhibitory activity of the solvent. This preparation was incubated for 24 h at 37°C. Antibacterial activity was determined in diameter/mm of the clear inhibitory zone formed around the antibiotic discs. The MIC of the extract was determined by

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Kirby Bauer methods in Mueller-Hinton broth (Merck) according to NCCLS [7]. Antibiotic Inhibitory test was performed and incubated at 37°C for 24 h. The minimum lowest concentration that inhibits the bacteria (no growth on plate) was recorded as Minimum Inhibitory Concentrations (MIC).

## Results

The results of the antimicrobial determinations for all the organic extracts of the leaf and stem parts of *Achyranthes aspera* against the three bacterial species are investigated in a disc-diffusion assay.

Given example in Figures 1 and 2 illustrates a representative plate showing the antibacterial activity of ethanolic and methanolic extracts

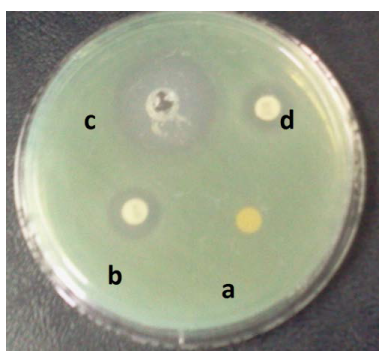


Figure 1: Antibacterial activity *Pseudomonas*.

- a. Negative Control
- b. Positive controls, Imipenem
- c. Methanol extracts
- d. Petroleum ether extracts

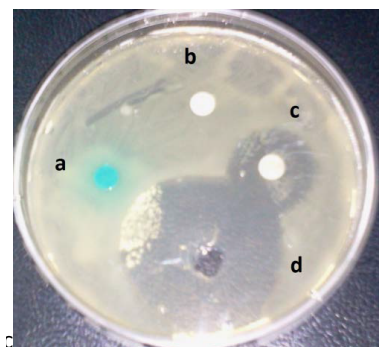


Figure 2: Antibacterial activity *Proteus*.

- a. Negative Control
- b. Resistance to Polymyxin
- c. Positive control Ampicillin
- d. Ethanol extract

from *A. aspera* that produced zones of inhibition against *pseudomonas* and *Proteus*.

Inhibition of bacterial growth was observed for fraction extracted samples and zone was measured (Table 1).

## Discussion

Phytochemicals is a fundamental requirement and pharmaceutical drug products. According to WHO, phytochemicals need to be standardized with safety guideline before releasing into market [8]. In this study, *Achyranthes aspera* was used which are easily available in St. Lucia and other tropical countries. The results revealed that extracts of *A. aspera* exhibited significant antibacterial activity against *Pseudomonas aeruginosa*, *Proteus mirabilis* and *Enterococcus faecalis*; Extracts from leaf part of *A. aspera* appeared to be more potent than that of the stem part, and the metabolic extract produced consistent level of inhibition of bacterial growth when these results compared with the leaves of *Senna alata* which was also demonstrated significant antibacterial and antifungal activities with methanol extraction [9]. It is interesting to note that the leaf extract of *A. aspera* could be used against multi drug resistance organisms.

Among the five solvent extracts of *A. aspera* studied for the antibacterial activity, most of them acted against pathogens. The outcome of the study encourages taking up screening the preliminary phytochemical analysis of leaf extract. The results were presented in Table 1. Further work is warranted to isolate and characterize the active principles available in the extracts of *A. aspera*. It is quite sure that such components could be useful in developing drugs [10].

## Conclusion

The discovery of a potent remedy from plant origin will be a great advancement in bacterial infection therapies. The results of present investigation clearly indicate that the plant has phytochemicals properties against different number of bacteria. This antibacterial study of the plant extracts demonstrated that folk medicine can be as effective as modern medicine to combat pathogenic microorganisms. The millenarian use of these plants in folk medicine suggests that they represent an economic and safe alternative to treat infectious diseases. These plants could serve as useful sources for new antimicrobial agents. Further work is needed to isolate the active principle from the plant extracts to carry out pharmaceutical studies for the treatment of infectious diseases such as urinary tract infections caused by bacteria and has significant scope for antibacterial research [11].

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Medicinal plant	Target organism	Zone of inhibition (mm) as produced in different organic solvent extracts of the plants				
		Petroleum ether	Methanol	Ethanol	Ethyl acetate	Chloroform
<i>A. aspera</i> (leaf)	<i>Pseudomonas aeruginosa</i>	13	17	12	11	13
	<i>Proteus mirabilis and</i>	22	15	11	13	18
	<i>Enterococcus faecalis</i>	21	9	8	6	7
<i>A. aspera</i> (stem)	<i>Pseudomonas aeruginosa</i>	15	7	18	20	22
	<i>Proteus mirabilis and</i>	11	9	15	17	18
	<i>Enterococcus faecalis</i>	-	-	-	-	-

Table 1: Inhibition of bacterial growth was observed for fraction extracted samples and zone was measured.

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