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Antibacterial Screening of *Achyranthes aspera* Linn. Root Extracts

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ABSTRACT

Achyranthes aspera Linn., a medicinal plant, belongs to the amaranthaceae family. In Ayurvedic system of medicine, the parts of this plant are used to treat various type of infectious diseases. Air shade dried and pulverized root material was extracted by using ethyl acetate, acetone and ethanol solvents at room temperature for twenty four hours. The solvents were collected under reduced pressure to get crude mass. The crude mass of all extracts were studied against Gram-positive and Gram-negative strains such as *Bacillus subtilis*, *Staphylococcus aureus*, *Salmonella typhimurium*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella abony* by disc diffusion method. Among these extracts, ethanol extract exhibit significant antibacterial activity against most of the bacterial strains while ethyl acetate and acetone extracts demonstrated less activity. The results obtained in the present study reveals, the potency order of the such extracts, can be employed in the treatment of diseases caused by tested pathogenic bacterial strains.

Keywords: Antibacterial activity, *Achyranthes aspera* Linn., Root extracts, Disc diffusion method.

INTRODUCTION

According to the World Health Organization more than 80 % of the world's population relies on traditional herbal medicine for their primary health care[1]. A wide range of medicinal plant part extracts are used as raw drugs and they possess varied medicinal properties. Microorganisms have developed resistance to many antibiotics and this has created vast clinical inconvenience in the treatment of infectious diseases[2]. The increase in microorganisms resistance to antibiotics, the use of antimicrobial drugs forced scientists to search for new antimicrobial substances from various sources including medicinal plants[3]. The trend of using natural products has increased and the active plant extracts are frequently used for new drug discoveries and the presence of antimicrobial substances[4].

Achyranthes aspera belongs to the family amaranthaceae is one of the important medicinal plant. It is used as traditional medicaments in the treatment of fever, especially malaria fever, dysentery, asthma, hypertension and diabetes[5,6]. The chloroform and ethanol root extracts of the *A. aspera* are reported to have anti-implantation and abortifacient activity[7,8]. The ethanol extract of the root possesses spermicidal activity[9]. The literature survey

indicates that aqueous and methanolic extracts of the whole plant have hypoglycaemic effect[10]. Roots are used as astringents to wounds, in abdominal tumor and stomach pains [11]. The stem shows abortifacient activity in the rat[12]. Leaf extracts were reported to possess thyroidstimulating and antiperoxidative properties [13].

The present work is carried out to evaluate the efficiency of ethanol, acetone and ethyl acetate root extracts of this plant for antibacterial activity.

MATERIALS AND METHODS

The plant material was collected from the Purander district of Pune, Maharashtra, India. It was authenticated at Botanical survey of India, Pune, Maharashtra, India.

Air shade dried and pulverized roots material (10gm) was soaked in ethanol, acetone and ethyl acetate solvents (70 ml) at room temperature for 24 hours. The solvents were collected under reduced pressure to obtain crude roots extracts. Antibacterial study was carried out against six bacterial strains (*Bacillus subtilis*, *Staphylococcus aureus*, *Salmonella typhimurium*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella abony*).

The paper disc diffusion method was employed. Samples of each extracts (50 mg) were dissolved in respective solvent (1 ml). Sterile Whatman filter paper discs (5 mm diameter) were impregnated with 10 µL of these solvent extracts (500µg/disc). The nutrient agar medium was prepared and transferred to the sterile petridishes in such a way to keep a uniform depth of approximately 4mm in sterile area. The 100µl of test organism cultured in nutrient broth media was spread with a sterile spreader on the surface of solid nutrient agar media. The sterile discs were impregnated with different extracts and placed on agar plates. The plates were incubated at 37 ± 0.1 °C for 24 hours.

After incubation, plates developed zones of inhibition which was measured (Table 1). All test were performed under sterile conditions. Ampicillin (1mg/disc) was employed as positive control.

RESULTS AND DISCUSSION

The ethanol, acetone and ethyl acetate root extracts display varying degree of antibacterial activities against the tested bacterial strains. The ethanol root extract is found to be the most effective against *Salmonella typhimurium*, *Escherichia coli*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Salmonella abony* strains. Acetone extract indicate the activity against *Salmonella typhimurium*, *Escherichia coli*, *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* strains. Ethyl acetate extract has noticed activity against *Salmonella typhimurium*, *Escherichia coli*, *Bacillus subtilis*, *Staphylococcus aureus* strains. The results are reported (Table-1)

Table 1: Zone of inhibition of ethanol, acetone and ethyl acetate root extracts

Test bacteria	Zone of inhibition(mm) ^a			Ampicillin
	Ethanol root extract	Acetone root extract	Ethyl acetate root extract	
	500µg/disc			1mg/disc
<i>Salmonella typhimurium</i>	17	14	13	16
<i>Escherichia coli</i>	14	11	9	11
<i>Bacillus subtilis</i>	13	10	10	12
<i>Staphylococcus aureus</i>	-	11	12	14
<i>Pseudomonas aeruginosa</i>	8	9	-	10
<i>Salmonella abony</i>	8	-	-	8

a. Zone of inhibition including the diameter of Whatman filter paper disc (5 mm)

This study indicates surprising results for all the tested extracts against most of the tested bacterial strains. The ethanol extract is found to be highly potent therefore, the MIC of this extract is determined.

The antibacterial assay of the ethanol extract against *Salmonella typhimurium* at different concentrations (400 µg, 300 µg, 200 µg, 100 µg, 50 µg, 40µg/disc) are tested by disc diffusion method and presented (Table 2).

Table 2. Zones of inhibition of ethanol root extract against *salmonella typhimurium* at different concentrations

Test bacteria	Zone of inhibition(mm) ^a						Ampicillin 1mg/disc
	Ethanol root extract						
	400 µg/disc	300 µg/disc	200 µg/disc	100 µg/disc	50 µg/disc	40 µg/disc	
<i>Salmonella typhimurium</i>	16	14	13	11	7	-	16

Zone of inhibition including the diameter of Whatman filter paper disc (5 mm)

The antibacterial activity of ethanol extract against *salmonella typhimurium* shows significant reduction in bacterial growth in terms of zone of inhibition. The zone of inhibition decreases on decreasing the concentration of extract. This indicates the concentration dependent activity. The MIC (Minimum Inhibitory concentration) is the lowest concentration of the compound at which the tested bacteria does not demonstrate visible growth at 40 µg/disc. The result reveals that the MIC of ethanol extract of *A.aspera* against *salmonella typhimurium* is 50 µg/disc.

CONCLUSION

The results obtained in the present study suggest that the ethanol, acetone and ethyl acetate root extracts of *A. aspera* reveals a significant scope to develop a novel broad spectrum of antibacterial drug formulation. These extracts can be used for development of a new alternative medicine system.

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