Research Article

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ANTIBACTERIAL, ANTIFUNGAL AND DPPH RADICAL SCAVENGING ACTIVITY OF *ACHYRANTHES ASPERA* L.

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ABSTRACT

The plant Apamarg (*Achyranthes aspera* L.) was tested for antibacterial (*Klebsiella pneumoniae, Pseudomonas aeruginosa, Bacillus subtilis, Escherichia coli, Staphylococcus aureus*), antifungal (*Aspergillus niger* and *Fusarium oxysporum*) activities. For extraction, methanol extract was chosen. The DPPH Radical Scavenging activity was used to test the antioxidant action. The current study reveals a link between indigenous *Achyranthes aspera* L. treatment and in vitro antibacterial and antifungal responses. Ascorbic acid had the lowest DPPH scavenging activity than methanolic extracts of *Achyranthes aspera* L., which exhibited antioxidant activity. The effects of DPPH radical scavenging are dose-dependent. The stems and roots of *Achyranthes aspera* L. are high in antioxidants that could be utilized as a natural supplier of antioxidants and antimicrobial components.

Keywords: *Achyranthes aspera* L., DPPH Radical Scavenging, Antioxidant, Antimicrobial

INTRODUCTION

Plants have always been employed as ingredients in most therapeutic methods throughout the history of the indigenous system of medicine. Plants produce a wide range of chemical compounds that, although the thought of as waste and detoxification products or expressions of shunt and overflow metabolism, continue to help humanity[1]. A wide range of Indian medicinal plants has long been used in traditional medicine to treat a wide range of diseases[2]. *Achyranthes aspera* L. is an Asian, South American, and African medicinal plant. It belongs to the Amaranthaceae family and can be found throughout India[3].

Being pungent, anti-phlegmatic, anti-periodic, diuretic, purgative, and laxative, it treats oedema, dropsy, piles, boils, and skin eruptions, among other things. Plant infusions are used to treat pneumonia, and root infusions treat digestive issues. Plant inflorescence is cooked in water, sieved, and consumed orally to treat jaundice. Eye disorders are treated with a paste of roots in water[2,4]. As per Ayurveda, *Achyranthes aspera* L. is bitter, pungent, laxative, stomachic, carminative, and beneficial for the treatment of vomiting, bronchitis, heart illness, piles, itching, abdominal discomfort, ascites, dysentery, and blood disease[5,6].

It is utilized for spermicidal[7], anti-pyretic[8], abortifacient action[9], antibacterial[10] antifungal[11], wound healing[12], anti-parasitic[13], anti-helminthic[14]. According to a phytochemical study, saponin, sterols, polysaccharides, and alkaloids are among the phytochemical components discovered in the whole plant of *Achyranthes aspera* L.[15]. The study objective was to look for phytochemicals, antioxidant, anti-bacterial, and anti-fungal activities.

MATERIALS AND METHODS

Fresh *Achyranthes aspera* L. stems and roots were obtained. Then it was gently washed before being dried in the shade at room temperature, then at 60 ºC in an oven. Dried materials were equally ground to generate fine powder using a mechanical grinder. These extracts were concentrated in a Rotary evaporator at 40 °C and dried in a lyophilizer under decreased pressure. The dried extract was stored at 4 °C in an airtight container until required.

Various pharmacopoeias propose pathogens and bacteria for antibiotic testing; thus, the experimental microorganisms are typical of those pathogens and bacteria. The experiment used both Gram-positive and Gram-negative microorganisms. These organisms were kept alive using nutritional agar and Sabouraud's agar slants. Subcultures were established on fresh mediums as needed. The master cultures were subcultured once a month. The test organisms in this study included *Klebsiella penumoniae, Pseudomonas aeruginosa, Bacillus subtilis, Escherichia coli, Staphylococcus aureus*, and fungal strains of *Aspergillus niger* and *Fusarium oxysporum*. *Klebsiella penumoniae, Pseudomonas aeruginosa, Bacillus subtilis, Escherichia coli, Staphylococcus aureus, Aspergillus niger* and *Fusarium oxysporum* fungal strains were collected off the slant and put to a flask containing sterilized nutrient broth, where they were allowed to develop at 37 ºC. The morphological characteristics were examined after plating the 18-hour culture on a nutrient agar plate. The nutrient broth cultures were centrifuged, and sterile saline made a cell suspension. 0.2 millilitres of the culture suspension was added to 12 millilitres of sterile nutritional agar medium in tubes. Before being placed in sterile plates in a uniform layer, the tubes were thoroughly mixed.

200 mg of powder was dissolved in 40 ml of methanol-based solvent, filtered through Whatman filters and collected in tubes after 24 hours. For evaporative drying, the solvent filtrates were kept in a water bath at 70 ^{o}C. 20 mL dimethyl sulfoxide was added to the dry tubes and centrifuged to dissolve the active components. Samples and blanks/controls were permeated in sterile 5 mm discs and aseptically put onto nutrition agar plates. After a 48-hour incubation period at 37 °C, the plates were inspected for zones of the microbe and fungal growth inhibition. The inhibitory zone was measured with a vernier calliper.

The DPPH radical is a well-known test for determining the potential of natural compounds to scavenge free radicals. This experiment measures the antioxidant chemicals' ability to scavenge the stable radical. The antioxidant activity of a methanolic extract of *Achyranthes aspera* L. was tested using the stable DPPH procedure for hydrogen contributing or radical-scavenging ability.

Various quantities of *Achyranthes aspera* L. extracts were diluted in methanol (1.25, 2.5, 3.75, and 5 g/ml), then 1 ml of each diluted extract was combined with 0.5 ml of 20 mg/l DPPH methanolic solution. The extract concentration and DPPH mixture were stored in the dark for 30 minutes at room temperature. The absorbance of the final solution was measured at 518 nm. The antiradical activity was measured using the IC50 (g/ml). Ascorbic acid was used as the control in this experiment.

RESULTS

The methanolic extract was utilised to estimate antibacterial and antifungal activity. Extracts from *Achyranthes aspera* L. were found to be effective against all five bacterial strains as well as two fungal strains. The methanolic extract was also plotted (Control, 100, 500, and 1000 µl/disc) as a positive control Vs blank, demonstrating that the extract has high antibacterial and antifungal action (Table 1). The outcomes of the DPPH radical scavenging assay of *Achyranthes aspera* L. stem and root are shown and summarized as methanolic extract compared to the control ascorbic acid in different concentrations. The methanolic extract has a higher activity than ascorbic acid. Compared to methanolic extracts of *Achyranthes aspera* L. that demonstrated antioxidant activity, ascorbic acid had the lower DPPH scavenging activity. The DPPH radical scavenging activities are dosage dependant, meaning that as the concentration increases, so does the extract inhibition of the DPPH radical (Figure 1).

Table 1: Bacterial and Fungal growth inhibition for fraction extract sample and zone assessment

Target Organism	Zone of inhibition (mm) as produced in the methanolic organic solvent extract of the plant			
	CONTROL	100	500	1000
Bacteria				
Klebsellia penumoniae	31	11	3	30
Pseudomonas aeruginosa	21	15	22	12
Bacillus subtilis	28	19	21	12
Escherichia coli	29	13	14	9
Staphylococcus aureus	27	9	16	10
Fungus				
Aspergillus niger	26	8	11	12
Fusarium oxysporum	28	11	10	13

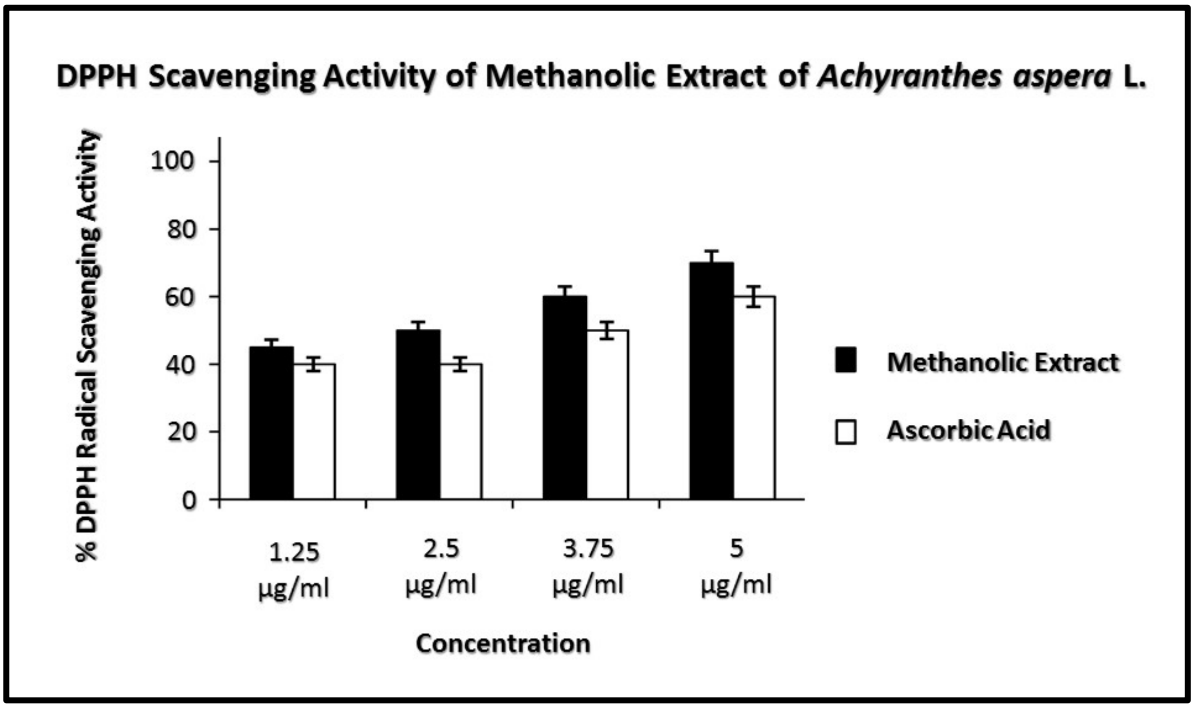

Figure 1: The potential of *Achyranthes aspera* L. to scavenge DPPH radicals. The Mean and Standard Deviation are used to calculate all values

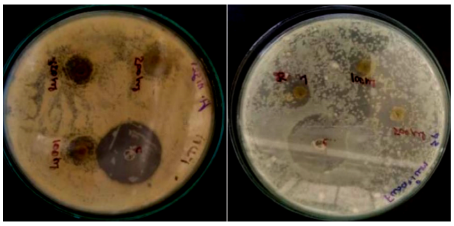

Figure 2: An antifungal exhibit of Methanolic Extract of *Achyranthes aspera* L.

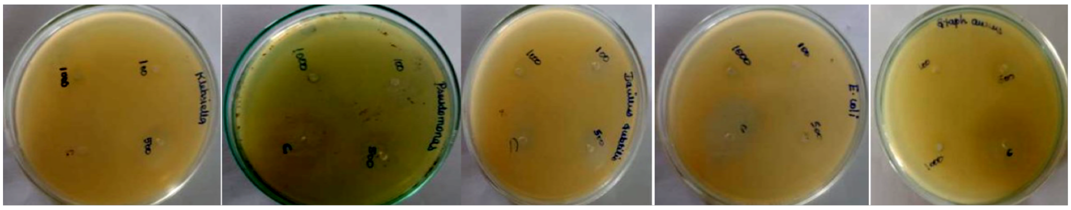

Figure 3: An antimicrobial exhibit of Methanolic Extract of *Achyranthes aspera* L.

DISCUSSION

Medicinal herbs have been used for centuries. Our predecessors used plants to heal and ameliorate the biological system's ailments. Herbal remedies are once again favoured for many purposes, including their widespread procurement, lack of adverse reactions, low cost of healthcare, and efficacy. Herbal extracts have a therapeutic effect due to the synergistic action of their many ingredients acting on a single or several target locations[16].

Plants with phenolic or polyphenolic antioxidant properties are found in the largest phytochemical class. Biological activity has been demonstrated for phenols, benzoquinones, phenolic acids, polypropylene, flavonoids, isoflavonoids, phenylpropanoids, phenolic quinines, lignins, melanins, tannins, and other phenolic compounds[17]. These phenolic compounds are produced by plants and can be used as natural antioxidants. Because of its long history of use in the treatment of several illnesses, as well as its clinical relevance, *Achyranthes aspera* L. was chosen for the study. In this study, we looked at the antioxidant potential of *Achyranthes aspera* L. methanolic leaf extract as an antibiotic and antifungal.

The current study found that *Achyranthes aspera* L. has antimicrobial properties against *Klebsiella penumoniae, Pseudomonas aeruginosa, Bacillus subtilis, Escherichia coli, Staphylococcus aureus, Aspergillus niger,* and *Fusarium oxysporum*, which are pathogenic organisms involved in the aetiology of a wide range of diseases (Table 1, Figure 2 & 3). It's worth noting that *Achyranthes aspera* L. root and stem extract may be used to fight multidrug-resistant microorganisms. The antimicrobial activities of *Achyranthes aspera* L. were studied, and the majority of them were found to be effective towards infestations.

The presence of flavonoids has antimicrobial pathways that include suppression of nucleic acid biosynthesis, cytoplasmic membrane functionality, and mitochondrial biogenesis [18]. Phenolic molecules have also been shown to have antimicrobial properties. The action involves changing the porosity of the cellular membranes, which could cause oxidative phosphorylation to be uncoupled, active transport to be inhibited, and pool metabolites to be lost due to membrane damage[19]. The occurrence of hydroxyl groups in phenolic compounds may affect their antimicrobial potency by anchoring to binding sites of enzymes, forming hydrogen bonds with enzymes, and altering their metabolic activity; additionally, phenolic molecular structures' lipid solubility and stearic barrier may influence their antimicrobial properties[20].

Oxidative cell damage causes a variety of physiological and neurological disorders. The formation of free radicals causes oxidative damage during the oxidation reaction[21,22]. Antioxidant belligerence is a defence mechanism that protects the body from oxidative stress. Oxidative stress to the cell happens when the number of available radicals is exceptionally high to overcome the antioxidant defence mechanism[23]. The antioxidant potential of the methanol extract was calculated and reported as a percentage suppression of DPPH radical scavenging activity. The DPPH radical scavenging activity increased when the dose was raised (Figure 2). More research is needed to identify and describe the active components discovered in *Achyranthes aspera* L. extracts. Components like these are virtually undoubtedly valuable for the development of pharmaceuticals[24].

CONCLUSION

For its longstanding tradition of use in managing bacterial and fungal illnesses, the herb Achyranthes aspera was selected for this research. The present study suggests a relationship between traditional *Achyranthes aspera* L. therapy and antibacterial and antifungal findings in vitro. The significance of ethnobotanical investigations in recognizing plants as biologically active biochemical resources is supported through these observations. Flavonoids and other phenolic chemicals are toxic to various bacteria and fungi, including *Staphylococcus aureus, Pseudomonas aeruginosa, Bacillus subtilis, Escherichia coli, Aspergillus niger,* and *Fusarium oxysporum*. Based on earlier facts, this exploratory inquiry could identify novel antimicrobial drugs for the speedy and comprehensive management of infectious diseases. Based on the outcomes of this study, we conclude that *Achyranthes aspera* L. stem and root can be used as a credible generator of naturally derived antioxidants.

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