OPEN ACCESS

Scholars International Journal of Traditional and Complementary Medicine

Abbreviated Key Title: Sch Int J Tradit Complement Med
ISSN 2616-8634 (Print) | ISSN 2617-3891 (Online)
Scholars Middle East Publishers, Dubai, United Arab Emirates
Journal homepage: https://saudijournals.com

Original Research Article

Deciphering of Antioxidant and Antimicrobial Potential of *Achyranthens aspera*

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DOI: https://doi.org/10.36348/sijtcm.2026.v09i08.002 | **Received:** 16.06.2026 | **Accepted:** 10.08.2026 | **Published:** 14.08.2026

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Abstract

Background: One of the most significant plants in the Indian medical systems of Unani, Ayurveda, and Sidha is *Achyranthes aspera* (Amaranthaceae). It is found in many tropical nations, especially India, where this therapeutic herb is a common weed. Many medicinal plants serve as antibiotic substitutes and are regarded as novel resources for the production of medicines in the treatment of antibiotic-resistant bacteria. ***Aim:*** This study aimed to assess the antibacterial and antioxidant activity of several A. aspera leaf and stem extracts. ***Methodology:*** In present investigation the antioxidant and antimicrobial activities of the stem and leaf extracts of Achyranthes *aspera* had been studied by the DPPH radical-scavenging assay and the disc-diffusion method, respectively**.** ***Result:*** The percentage inhibition of aqueous leaf and stem extract demonstrated a significant effect when compared to the standard, according to the investigation's findings. The findings show that leaf aqueous extracts have the highest antioxidant activity, followed by stem aqueous extracts, while ethanolic extracts have the lowest. The results of the antimicrobial investigation show that ethanolic and aqueous extracts of the leaves and stem of Achyranthens aspera Linn have antibacterial activity against particular microorganisms. It has been discovered that ASL aqueous extract is more active than ethanolic extracts. ***Conclusion:*** While the antimicrobial study demonstrated strong inhibitory effects against particular pathogenic bacteria, the antioxidant activity evaluated by the DPPH test demonstrated significant free radical scavenging capabilities of the selected extracts. the presence of flavonoids, tannins, terpenoids, and alkaloids, which are known to be effective at scavenging radicals and preventing microbial growth.

Keywords: Achyranthens aspera**,** Antioxidant, Antimicrobial & DPPH.

INTRODUCTION

The medicinal plant Achyranthes aspera, also referred to as "Prickly Chaff Flower," is deeply ingrained in many traditional treatment systems around the world and has considerable ethnopharmacological significance [1]. The plant's cultural value and therapeutic possibilities are highlighted by its historical use in traditional medicine systems to treat wounds, inflammation, gastrointestinal issues, respiratory conditions, and discomfort. Alkaloids, flavonoids, saponins, and triterpenoids are among the bioactive substances that researchers have studied over the years and linked to their different pharmacological effects. Achyranthes aspera's pharmacological actions include immunomodulatory, analgesic, antibacterial, anti-inflammatory, and antioxidant qualities. The plant is a good option for medication development and the creation of standardized herbal formulations because of these varied actions, which have attracted the attention of pharmaceutical researchers and medical professionals. Despite its extensive historical use and growing scientific interest, the safety and toxicity of Achyranthes aspera require further investigation [2-3]. Global health is seriously threatened by the significant rise in antibiotic resistance in microorganisms. In particular, streptococcus pneumoniae contaminated by methicillin-safe Penicillin-safe MRSA Staphylococcus aureus Safe for vancomycin Enterococcus and Mycobacterium tuberculosis are the most prevalent since many of these organisms are resistant to a few types of well-known antimicrobials. This situation is driving researchers to look for novel antibacterial medications that block important bacterial targets without being impacted by mechanisms of resistance to currently available chemotherapeutic agents. [4]. The major characteristics of antimicrobial medication are the kinds of microorganisms it targets, such as viruses and bacteria. Based on their distinct chemical makeup, antimicrobial compounds are separated into two categories. Antibiotic medications and metal and metal oxide nanoparticles are

examples of synthetic antimicrobial agents, often known as chemical antimicrobial agents. Herbal antibacterial agents make up the second category. As antimicrobial agents, antibiotics and other substances greatly aid in the disruption of germs' cell walls. Herbal plants are the source of the other category we should think about. By acting as free radical scavengers, these organic compounds can successfully stop the generation of reactive oxygen species (ROS) [5]. Antioxidants are a family of naturally occurring chemicals that can either prevent or lessen the physiological system's oxidative stress. Because the body uses oxygen often, free radicals are continuously produced. These free radicals cause cell damage in the body and are linked to a number of health issues, including cancer, diabetes, heart disease, and macular degeneration. As excellent free radical scavengers, antioxidants aid in both avoiding and mending the harm that these radicals do to cells. Antioxidants are abundantly produced naturally by both plants and mammals [6]. It is essential to promote the use of herbal treatments and increase research on medicinal plants. Microbial infections are a major cause of sickness in people of all ages and places.

Experimental Work

Collection & Extraction

Freshly collected plant material of Achyranthes aspera consisting of leaves was subjected to shade drying to remove inherent moisture content without affecting thermolabile phytoconstituents. After complete drying, plant material was pulverized using mechanical grinder to obtain coarse powder. The powdered medication underwent sequential solvent extraction via a Soxhlet system, employing solvents in ascending order of polarity: petroleum ether, chloroform, ethanol, and distilled water. Successive extraction method allows systematic separation of phytoconstituents based on polarity differences.

Antioxidant Activity (DPPH Radical Scavenging Method)

Different concentrations of extract were prepared and mixed with DPPH solution. The combination was incubated for a designated duration under darkness. Absorbance was quantified with a UV–Visible spectrophotometer at a wavelength of 517 nm [7]. The percentage inhibition of free radicals was determined using the formula:

% inhibition = (Control absorbance – Sample absorbance / Control absorbance) × 100

Antimicrobial Activity (Agar Well Diffusion Method)

The antimicrobial efficacy of plant extracts was assessed utilizing the agar well diffusion technique. Nutrient agar plates were infected with chosen microbial strains. Wells were prepared in agar plate and different concentrations of plant extract were added into wells. Plates were incubated at suitable temperature for specified time period. After incubation, zone of inhibition around well was measured in millimeters.

Preparation of microorganisms for experiment

Micro-organism strains used for antimicrobial studies were obtained from a medical college. Organisms used for the experiments were subcultured in nutrient growth, blood agar media, MacConkey agar, and nutrient agar. For antibiotic sensitivity testing, Muller-Hinton agar was used.

Preparation and application of disks for experiment

Various concentrations of the extracts (10-60 µg/ml) were generated by reconstituting with DMSO. The streaking plate method was employed to inoculate test microorganisms onto Mueller-Hinton agar medium. Following the streaking process, the autoclaved filter paper discs (5 mm in diameter) infused with the extracts were positioned on plates utilizing flame-sterilized forceps. The antibacterial assay plates were incubated at 37°C for 24 hours. Amoxicillin/Cefotaxime (60µg/ml) was utilized as the positive control, while DMSO served as the negative control [8-9].

Observation of results

Zone inhibition's presence or absence was noted as the experiment's outcome. The existence of an inhibitory zone indicated the absence of bacterial growth, which was noted as a good result; the absence of an inhibition zone was noted as a negative result. The experiment was conducted three times during the day to ensure accurate findings. After deducting the disk diameter, or 5 mm, the inhibitory zone diameters are expressed in millimeters.

Statistical analysis

All values were statistically examined using one-way analysis of variance (ANOVA) followed by Dunnett's test. Comparisons between the control and drug-treated groups were deemed significant (*P<0.01). All values are shown as mean ± SEM.

RESULT AND DISCUSSION

The ethnopharmacological relevance of *Achyranthens aspera* stems from its diverse traditional use. It has been used for generations to treat a variety of illnesses, including wounds, gastrointestinal issues, respiratory diseases, inflammation, and discomfort. Researchers are investigating its possible uses in contemporary medicine due to the wide range of its pharmacological actions, which include anti-inflammatory, antioxidant, antibacterial, and analgesic effects. Given these factors, it is imperative to progress the development of natural antibacterial agents. The native plant Achyranthens aspera L. was chosen for this investigation. The anti-microbial activity of PEE, CE, EE, and AE of ASL = Achyranthens aspera Linn was investigated. Achyranthens aspera Linn. leaves and ASS. Stem: When compared to other studied extracts and conventional medications, both aqueous and ethanolic extracts shown notable activity. The findings of this study demonstrate the antibacterial activity of ethanolic and aqueous extracts of Achyranthens aspera Linn's

leaves and stem against specific microorganisms. ASL aqueous extract is found to be more active than ethanolic extracts. At a concentration of 60µg/ml, extracts were shown to be less active than the standard. The results further illustrate the extracts' tremendous potential with further separation and purification (table 1 & Fig 2). DPPH techniques were used to measure the antioxidant activity of PEE, CE, EE, and AE in the leaves and stem of Achyranthens aspera L. Table 2 displays the results, demonstrating that AEBPL exhibited the highest inhibition when compared to other extracts. The results also show that the leaf aqueous extracts have the highest antioxidant activity, followed by the stem aqueous extract, while the ethanolic extracts have the lowest. The DPPH test, which measures antioxidant activity, revealed that the selected extracts had a significant capacity to scavenge free radicals. Phytochemical levels and antioxidant capacity are positively correlated, as seen by the increased antioxidant activity of extracts with higher phenolic and flavonoid concentrations. Significant inhibitory effects against particular hazardous bacteria were demonstrated by the antimicrobial investigation. The presence of flavonoids, tannins, terpenoids, and alkaloids—all of which are known to impede microbial growth—may be responsible for the observed antibacterial effects.

Divulgence of Investigation

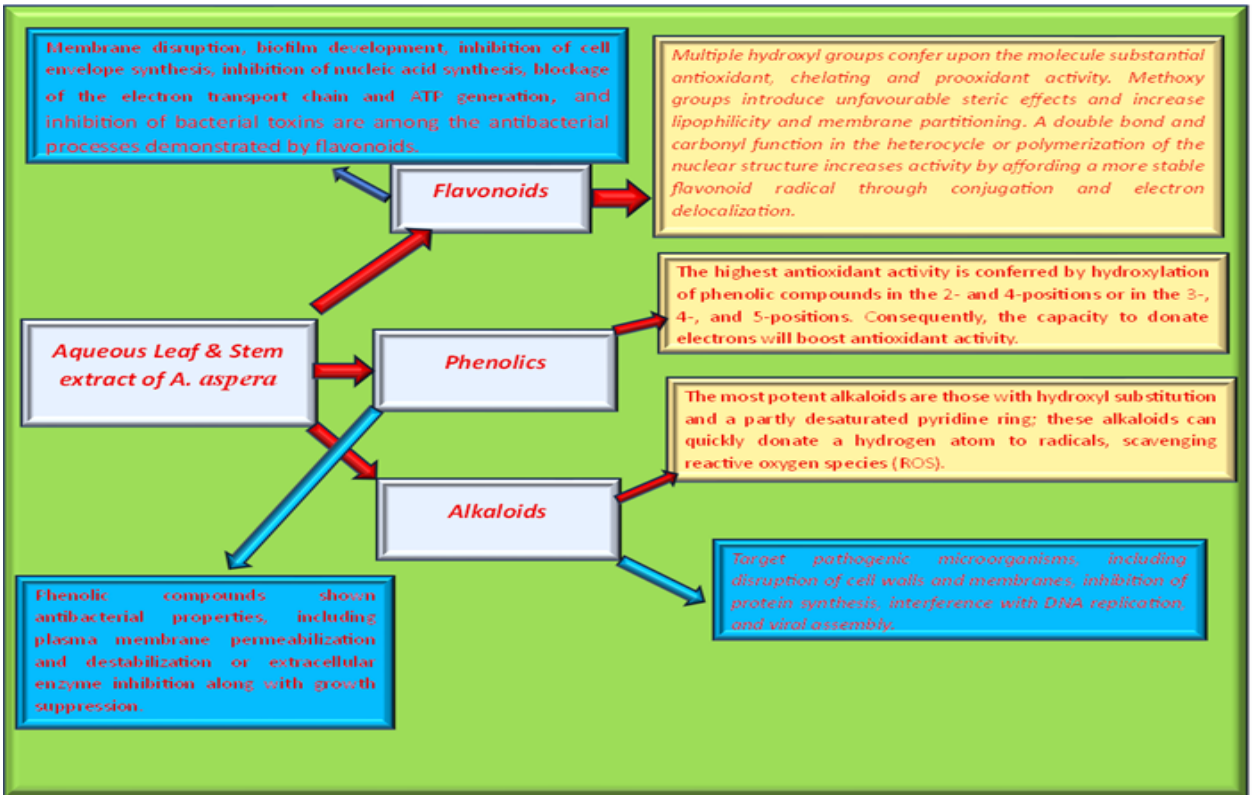

Table 1: Achyranthens aspera L. extract's antimicrobial properties

S. No	Treatment	Conc.	Inhibition Zone (mm)			
		(µg/ml)	EC	PA	SA	EF
1.	Control	-	-	-	-	-
2.	PEEASL	60	-	-	-	-
3.	CEASL	60	4.33±0.33	-	-	-
4.	EEASL	60	17.22±0.16	14.30±0.33	19.01±0.16	12.62±0.16
5.	AEASL	60	18.29±0.16	17.33±0.33	21.5±0.28	15.16±0.16
6.	PPEASS	60	-	-	-	-
7.	CEASS	60	-	16.5±0.288	14.16±0.16	-
8.	EEAAS	60	8.33±0.16	10.16±0.28	7.33±0.16	6.5±0.50
9.	AEAAS	60	10.29±0.13	12.23±0.10	9.24±0.11	8.3±0.33
10.	SD	60	22.5±0.76 (a)	24.16±0.72 (a)	25.16±0.72 (b)	19.5±0.28 (b)

Values are expressed as Mean (X) ± SEM, n=3 **Abbr**.: EC= *Escherichia coli,* PA = *Pseudomonas aeruginosa,* EF =*Enterococcus faecalis,* SA= *Staphylococcus aureus,* EC (G-), PA (G-), SA (G+), EF(G+), C: Control (DMSO), SD=Standard (a = Cefitaxime, b= Amoxycillin)

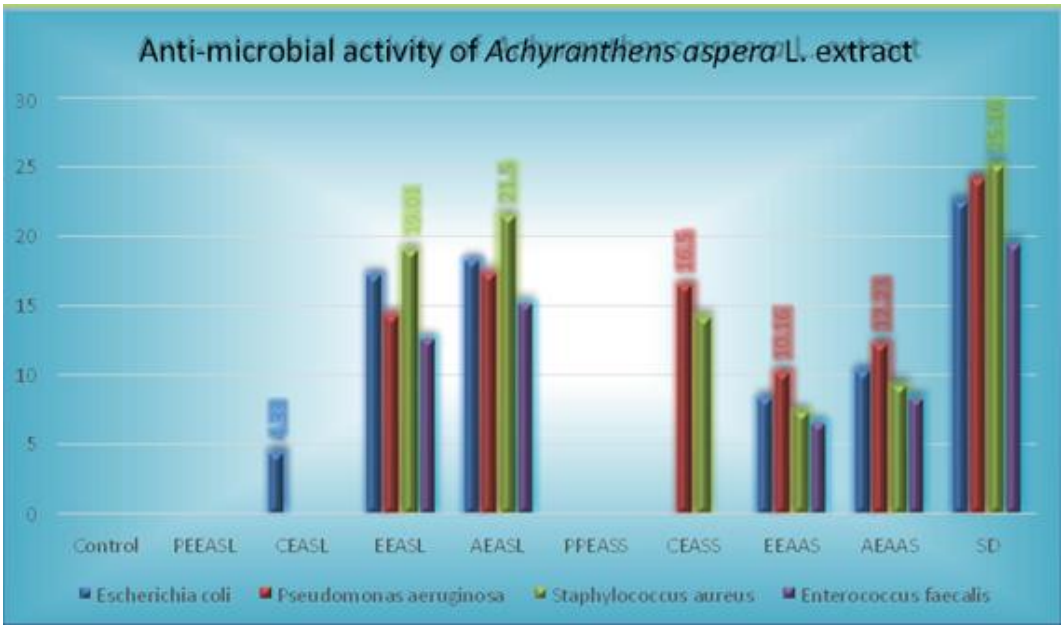

Figure 1: Anti-microbial activity of *Achyranthens aspera* L. extract

Table 2: % Inhibition of *Achyranthens aspera* L. extract using DPPH method

S/No.	Extract (100 µg/ml)	% Inhibition
1.	PEEASL	43.12
2.	CEASL	48.19
3.	EEASL	50.22
4.	AEASL	57.12
5.	PPEASS	16.02
6.	CEASS	14.18
7.	EEAAS	17.17
8.	AEAAS	20.21
9.	Ascorbic Acid	78.43

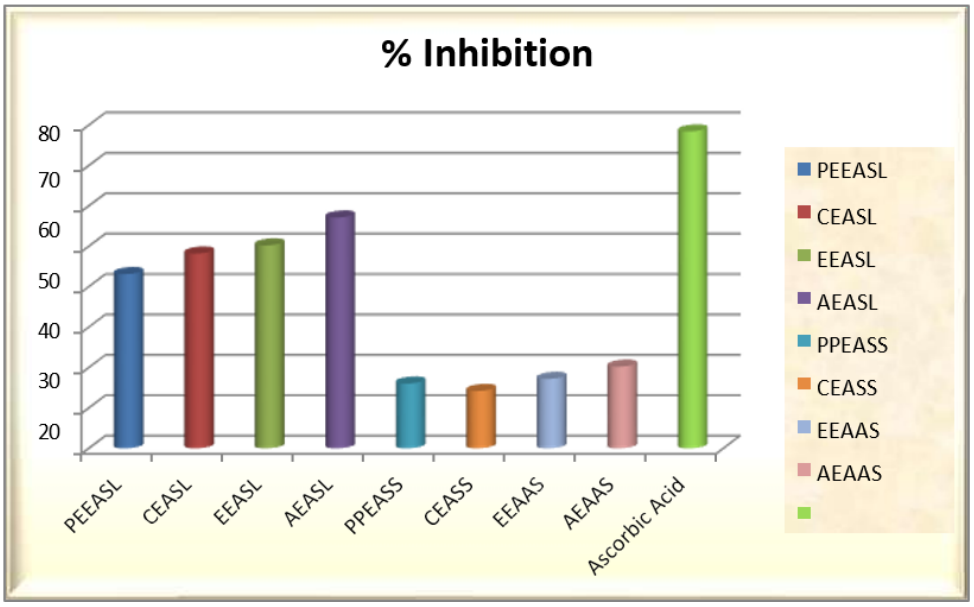

Figure 2: % Inhibition of *Achyranthens aspera* L. extract using DPPH method

CONCLUSION

The plant's traditional medicinal claims were scientifically supported by the extracts' strong antibacterial and antioxidant properties. The potential of the plant's extracts as natural remedies for microbial illnesses has been investigated.

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