



***Abutilon Indicum*: Phytochemical Screening and Antimicrobial Assessment**

SWATANTR BAHADUR SINGH*, MHAVEER SINGH¹ and POOJA SAINI¹

*School of Pharmaceutical Sciences, Faculty of Pharmacy, IFTM University,
Moradabad, 244001 Uttar Pradesh, India

¹School of pharmaceutical sciences, Faculty of Pharmacy, IFTM University,
Moradabad, 244001, Uttar Pradesh, India.

*Corresponding author E-mail: swatantrsng@gmail.com

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ABSTRACT

The current scenario reflects customers' interest in natural medications, a growing interest in wellness, and a shift towards preventative healthcare. The market for herbal medicines is rapidly developing, and their potential as antibacterial agents has recently sparked widespread interest. The goal of this research is to look at the antibacterial capabilities and phytochemical composition of *Abutilon indicum*. The organic extract (ethanol, methanol, and hexane) and hydro extract of *Abutilon indicum* are evaluated against the fungus *Candida albicans* and a few bacterial species, including *Shigella dysenteriae*, *Proteus vulgaris*, and *Salmonella typhimurium*, using the Agar disc diffusion technique. The observed results demonstrated considerable antibacterial action against the microbiological pathogens tested. The plant's alcoholic extract contains antifungal and antibacterial effects. The outcomes of the study were compared with the well-known standard antimicrobial drugs such as nystatin for antifungal efficacy and streptomycin for antibacterial efficacy. The ethanol extract containing terpenoids, alkaloids, tannins, flavonoids, and steroids was found to have the strongest antibacterial impact of any of them. As a result, it has been determined that the plant's alcoholic extract contains antifungal and antibacterial characteristics.

Key words: *Abutilon indicum*, antimicrobial activity, Agar disc diffusion method, Morphology, phytochemical screening.

INTRODUCTION

The increasing interest in natural, plant-based alternatives to synthetic pharmaceuticals

has led to a resurgence of herbal medications in global health practices in recent years¹. Herbal medications are gaining significance in contemporary healthcare, driven by scientific research, traditional



knowledge, and rising consumer demand for natural alternatives². The antimicrobial potential of herbal medications is well established, and the use of plants to combat diseases is expanding as safer medication options are sought. This trend is largely attributed to the global challenge of antimicrobial resistance (AMR), the limitations of conventional antibiotics, and a renewed focus on alternative and traditional therapies. Herbal remedies have been studied for their ability to inhibit a broad spectrum of infectious pathogens, including bacteria, yeasts, and other microorganisms. Many plants contain bioactive compounds such as alkaloids, flavonoids, phenolic acids, terpenoids, and essential oils with demonstrated antibacterial properties³. These compounds may act through various mechanisms to reduce the pathogenicity of microorganisms.

Traditional medicines derived from medicinal plants are utilized by approximately 60% of the global population. Herbal formulations are often preferred due to their lower cost and lesser side effects, despite the availability of alternative treatments for various illnesses and complications⁴. The demand for pharmaceuticals and plant-based nutritional supplements has increased in recent years. Researchers across disciplines, including microbiology, chemistry, ethnopharmacology, and botany, are actively seeking phytochemicals and drugs for the organic treatment of highly contagious diseases⁵. Although 30 to 50% of current drugs are plant-derived, relatively few are employed as antimicrobials. Plants have historically been used by traditional healers to treat and prevent infectious diseases. Secondary metabolites such as terpenoids, tannins, alkaloids, flavonoids, saponins, and anthraquinones present in plants have demonstrated antibacterial properties⁶.

The shrub *Abutilon indicum* Linn. belongs to the *Malvaceae* family and can be found in India, Africa, Australia, and other arid regions of the world⁷. The herb's leaves, roots, spores, and seed oil are all widely used to cure a variety of ailments in ancient medical systems such as Ayurveda and Siddha. The roots might be utilized to treat hemorrhagic uterine discharge. The leaves can be used to treat a number of inflammations, including toothaches, lumbago, and piles⁸. *Abutilon indicum*, is also known as Atibala in Sanskrit, Kanghi in Hindi, and Country

Mallow in English. *Abutilon indicum* a soft, hairy evergreen shrub, grows to a height of about three meters. The evergreen shrubberies are 1.9-2.5 cm long, cordiform, oblong, acicular, notched, and rarely subtrilobate. The cylindrical, white, stellate, hairy petiole measures 1.5-1.70 cm long. The golden blooms have peduncles that meet above the center^{9,10}. Seeds are 3-5 mm in size and have a bean shape with obovate, tubercled, or stelliform black or brown hairs. The branches are 3.8-7.5 cm long, the petals are 9 mm long, the shafts are commonly 2.5-5.0 mm long, and the axillary is single and jointed together on top¹¹.

The current study aims to determine the initial phytochemical components and antibacterial efficacy of various concentrations of *Abutilon indicum* shrub and branches. This study includes the evaluation of potency of plant's extract towards microorganisms, by agar disc diffusion method. Collection, authentication, isolation and characterization of phytoconstituents are done to discover the responsible group for the potentiality.

EXPERIMENTAL

Chemicals and Instruments

Soxhlet apparatus, rotatory evaporator, and autoclave are the major instruments used for the experimentation. Ethanol, methanol, hexane, hydrochloric acid, sulphuric acid, NaCl, ferric chloride, and lead acetate solution are the core chemicals, obtained from sigma Aldrich. Conventional drugs streptomycin and nystatin were purchased from the local market. Experimental glassware was of analytical grade and sterilized before experimentation. All the experimental work was carried out in the laboratory of Department of pharmacognosy, IFTM University, Moradabad, India.

Collection and authentication

The plant was obtained from the nursery of Bareilly, Uttar Pradesh. The plant's sample sent to the Ministry of Environment, Forests & Climate Change, Botanical Survey of India, Central Regional Centre, Allahabad-211002, Uttar Pradesh, India for the verification. The plant was confirmed and authenticated by the Botanical Survey of India (BSI), Central Regional Centre in Allahabad, Uttar Pradesh

(Uttar Pradesh) India under authentication number स.भा.व.स./म.क्षे.के./प्रषा./2023-24/792.

Extraction and isolation

Various organic and inorganic solvents were utilized to prepare the extracts of the plant. The continuous hot extraction process (soxhlet extraction) was employed for the extraction. The plants were ground and allowed to air dry. Using a Soxhlet apparatus, the powdered drug was refluxed in 500 mL of methanol for 24 hours. Whatmann filter paper, No. 1, was used to filter the extract. After that, the filtrate was dried by evaporating the solvent using a rotatory evaporator. Extracts of ethanol, methanol, hexane, and DW (distilled water) are produced and kept safe¹². The dried extract was kept in sterile, labeled vials with a lid at 20°C. Active cultures were prepared by cultivating microorganisms in tubes of Muller-Hinton (MH)/ Potato dextrose agar (PDA) for microbes and Sabouraud dextrose Agar (SDA) broth for yeast. Stock cultures of microbes are kept at 4°C.

Phytochemical screening

Phytochemical screening was performed by following standard procedures. Prepared extracts were exposed to the chemical test for the detection of tannin, flavonoids, anthraquinones, saponin, cardiac glycoside, alkaloids, steroids, and terpenoids²⁷. All the tests were performed in the departmental laboratory.

Antimicrobial assay

The cultures of the bacterial colonies of *S. typhimurium*, *S. dysenteria*, *P. vulagaris*, and *C. albicans* were kept on nutrient agar media and maintained at 4°C. After forming colonies, they were cultivated from the School of Biotechnology, IFTM University, Moradabad^{13, 14}.

Antibacterial potentiality¹⁵ of the extracts was identified by measuring the width of the zone of inhibition (ZOI). Standard procedure, agar disc diffusion method was employed to perform the activity¹⁶⁻¹⁸. MH agar plates having an inoculum dimension of 106 colony-forming units (CFU) was prepared and covered with 4.0 mm discs saturated with each extract in a concentration of 100mg/mL. Similarly, antibiotic discs (6.0 mm in diameter) Streptomycin (20 µg/mL) for microbes, and Nystatin (20 µg/mL) for fungus were also employed as a positive control¹⁹. Each plate also contained a blank disc by placing the solvent control alone in the middle. Every plate was incubated for 18 hours at 37°C for microbes and 48 hours at 28°C for fungus. Ethanolic extract elicited captivating findings among the four extracts, but the aqueous extract elicited no reaction²⁰.

RESULTS AND DISCUSSION

The plant's methanolic extract had a maximum ZOI of 30.4 mm against *Salmonella typhimurium*, but the ethanolic extract had a

Table 1: Phytochemical investigation of Secondary Metabolites of Ethanolic extract

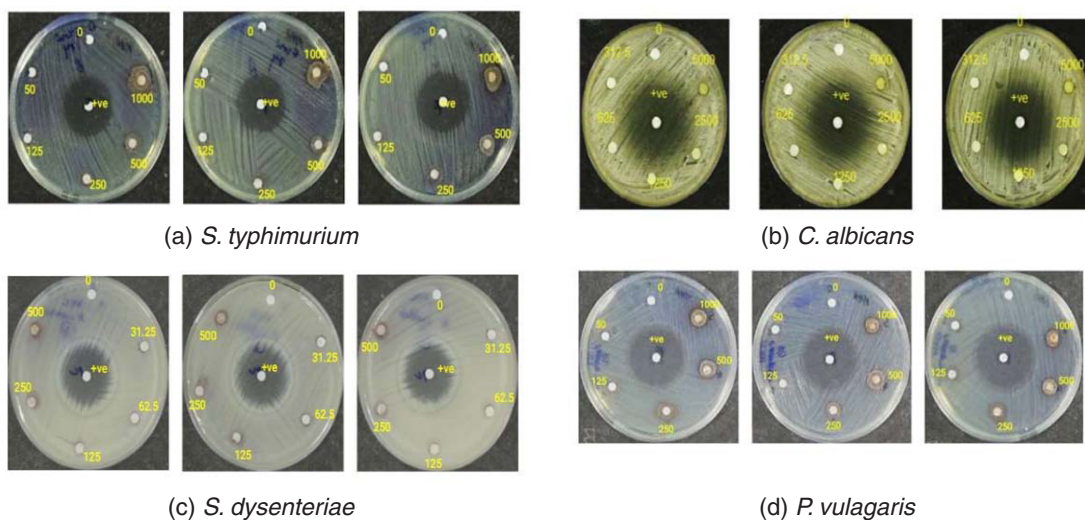
S. No.	Secondary metabolites	Test	Parts of the plant		
			Leaf	Stem	Root
1	Tannins	Braymer test	Negative	negative	positive
2	Flavonoids	Shinoda test	Positive	negative	positive
3	Anthraquinones	KOH test	Negative	negative	negative
4	Saponins	Frothing test	Positive	positive	negative
5	Cardiac glycosides	Killer-Killiani test	Positive	negative	negative
6	Alkaloids	Dragendorff's test	Positive	positive	negative
7	Steroids	Liebermann Burchard test	Positive	negative	positive
		Steroids test	Positive	negative	negative
8	Terpenoids	Liebermann Burchard test	Positive	positive	negative
		Salkowski test	Positive	positive	negative

Positive= present, negative = absent

Table 2: Result of Antimicrobial activity of *Abutilon indicum*.

Solvent extracts	Concentration mL	ZOI (in mm) $\pm$ SD*			
		<i>S. typhimurium</i>	<i>S. dysenteriae</i>	<i>P. vulagaris</i>	<i>C. albicans</i>
Methanol	20	30.4 $\pm$ 0.5	27 $\pm$ 0.9	-	21 $\pm$ 0.6
Ethanol	20	19 $\pm$ 0.5	35.8 $\pm$ 0.1	26.4 $\pm$ 0.4	15.9 $\pm$ 0.7
Aqueous	20	33.5 $\pm$ 0.9	21 $\pm$ 0.7	19.3 $\pm$ 0.4	7.9 $\pm$ 0.8
Hexane	20	21.5 $\pm$ 0.4	5.8 $\pm$ 0.6	-	15.2 $\pm$ 0.4
Streptomycin	20	30 $\pm$ 0.8	35.5 $\pm$ 0.8	27 $\pm$ 0.5	-
Nystatin	20	-	-	37.2 $\pm$ 0.1	17 $\pm$ 1.8

*Standard deviation

**Fig.1. Zone of inhibition (in mm) by Agar disc diffusion method (a) *S. typhimurium* (b) *C. albicans* (c) *S. dysenteriae* (d) *P. vulagaris***

maximum ZOI of 35.8 mm against the bacteria *Shigella dysenteriae*. The plant's hexane extract had a ZOI of 21.5 mm against the bacterium *Salmonella typhimurium*. As a result, phytochemical screening of secondary metabolites was carried out using ethanolic extract²¹, which exhibited the highest inhibition of any extract tested.

Table 1 displays the findings of the phytochemical screening conducted on the extracts from different portions of *Abutilon indicum* to determine if they contained tannin, flavonoid, alkaloid, anthraquinone, saponin, volatile oils, phlobatannin, cardiac glycosides, steroids and terpenoids. According to the results of the initial phytochemical screening investigation^{22,23}, *Abutilon*

indicum leaves have modest levels of tannins and steroids, as well as trace quantities of cardiac glycosides, terpenoid, alkaloids, flavonoids, and saponins,. Steroids, tannins, and flavonoids are present in trace levels in *Abutilon indicum* roots. Moderate levels of flavonoids and trace quantities of steroids and saponins are found in the blooms²⁴⁻²⁶. Terpenoids, alkaloids, and saponins are present in trace concentrations in the stem. The entire plant was discovered to be devoid of anthraquinones. Because Gram-negative bacteria have a number of distinctive characteristics, such as a cell wall that renders them resistant to several types of antibiotics, treating Gram-negative bacterial infections can be challenging. Broad-spectrum antibiotics, such beta-lactams and carbapenems, have traditionally

been used to treat infections. Researchers are now turning to natural resources, such as medicinal plants, because even these medications are no longer effective against some microorganisms. It is necessary to develop new, safe and herbal medications to treat Gram-negative bacterial infections. This study aids in the development of natural strategies to counteract some gram-negative bacteria's resistance to drugs.

CONCLUSION

The study assessed the phytochemical profile of the plant by conducting several qualitative tests for various phytochemicals. The different extracts were prepared using a Soxhlet continuous hot extractor with hexane, ethanol, methanol, and water as solvents. All extracts were evaluated for antimicrobial efficacy against *S. typhimurium*, *S. dysenteriae*, *P. vulgaris*, and *C. albicans*. Both Gram-positive and Gram-negative bacterial and fungal strains were tested using the Agar disc diffusion method. The zone of inhibition indicated that the alcoholic extract exhibited the highest activity against *Shigella dysenteriae*, while the hexane extract showed the lowest efficacy. Regarding antifungal activity, the methanol extract demonstrated the greatest effectiveness, whereas the aqueous extract displayed the weakest activity. Consequently, the plant may be further developed and investigated as a natural and potent antimicrobial agent by testing against additional Gram-positive and Gram-negative species, as our study found significant activity against *Shigella dysenteriae* compared

with streptomycin for antibacterial and nystatin for antifungal assessment.

List of abbreviation

ZOI	Zone of inhibition
Mm	Milimeter
KOH	Potassium hydroxide
%	Percent
S. No.	Serial Number
°C	Degree Celsius
mL	Mili litre
Cm	Centimetre
<i>S. typhimurium</i>	<i>Salmonella typhimurium</i>
<i>P. vulgaris</i>	<i>Proteus vulgaris</i>
<i>S. dysenteriae</i>	<i>Shigella dysenteriae</i>
<i>C. albicans</i>	<i>Candida albicans</i>

Data Availability Statement

Not applicable.

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Author receives no source of funds.

Conflict of interest

Author declares no conflict of interest.

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