International Journal of Fauna and Biological Studies
Available online at www.faunajournal.com

E-ISSN 2347-2677
P-ISSN 2394-0522
Impact Factor (RJIF): 5.69
https://www.faunajournal.com
IJFBS 2025; 12(5): 132-134
Received: 24-08-2025
Accepted: 27-09-2025

**Dr. Ujjwala Shivaji Deshmukh**
Government Vidarbha Institute of Science and Humanities (Autonomous), Amravati, Maharashtra, India

**Sharvari Shivaji Deshmukh**
Research Student, Department of Zoology, Government Vidarbha Institute of Science and Humanities (Autonomous), Amravati, Maharashtra, India

**Corresponding Author:**
**Dr. Ujjwala Shivaji Deshmukh**
Government Vidarbha Institute of Science and Humanities (Autonomous), Amravati, Maharashtra, India

# Investigation of antibacterial potential in solvent-based leaf extracts of *Alstonia scholaris*

**Ujjwala Shivaji Deshmukh and Sharvari Shivaji Deshmukh**

## Abstract
*Alstonia scholaris*, a plant known for its medicinal properties, was investigated for its antibacterial activity against *Staphylococcus aureus* using leaf extracts prepared in methanol, acetone, ethanol, chloroform and distilled water. The antibacterial potential of each extract was evaluated *in vitro* through the agar well diffusion method. After 24 hours of incubation at 37 ± 1 °C, zones of inhibition were recorded, with the aqueous extract showing the highest activity (15 mm), followed by methanolic (14 mm), acetonic (13 mm), ethanolic (10 mm) and chloroform (6 mm) extracts. These results were compared to the inhibition zones produced by standard antibiotics Vancomycin Hydrochloride (28 mm) and Maczone (20 mm). Although the plant extracts demonstrated comparatively lower antibacterial activity than the commercial antibiotics, the aqueous and methanolic extracts exhibited notable inhibitory effects, indicating the presence of bioactive constituents. These findings warrant further phytochemical and mechanistic studies to explore the therapeutic potential of *Alstonia scholaris* as a source of novel antibacterial agents.

**Keywords:** *Alstonia scholaris*, leaf extract, solvents, *Staphylococcus aureus.* Methanol, Acetone, Ethanol, Chloroform, distilled water

## Introduction
The use of plants in traditional medicine is a time-honored practice, with many species historically utilized for their therapeutic properties. Numerous plant-derived compounds have since been pharmacologically validated for their efficacy, particularly for antimicrobial applications (Kachhawa *et al*., 2012) [4]. Numerous plant species have been pharmacologically and clinically validated worldwide for their bioactive phytochemicals with significant antimicrobial potential against pathogenic microorganisms. *Alstonia scholaris* commonly known as the blackboard tree or devil's tree, is one such plant recognized for its wide range of therapeutic properties. In India, the tree is popularly referred to as "Saptaparni," derived from its characteristic whorls of seven leaves (Singh *et al*., 2017) [8]. It is a fast-growing, evergreen tree belonging to the family Apocynaceae, comprising approximately 40 species distributed throughout tropical regions. *Alstonia scholaris* is a glabrous tree that can attain a height of up to 40 meters. The leaves are glossy on the upper surface and greyish on the underside, occurring in whorls of three to ten with petioles measuring 1-3 cm in length. Flowering occurs during October, with fragrant blooms resembling those of *Cestrum nocturnum*. The seeds are oblong, with ciliated margins and tufts of hair at both ends, measuring approximately 1.5-2 cm. The bark is nearly odorless but intensely bitter and contains a milky, bitter latex. While known for its medicinal properties, the plant exhibits toxicity at high doses, causing organ damage in both rats and mice, as demonstrated in experimental studies.

The therapeutic potential of *Alstonia scholaris* has been extensively investigated, particularly for its bioactive constituents present in the leaves, bark, and roots (Joel James et. al., 2011) [3]. In Mizo traditional medicine, bark extracts of this tree are commonly used to treat bacterial and parasitic infections (Lalngaihmanawmil *et al.*, 2023) [6]. The bark is rich in alkaloids such as ditamine, echitenine, echitamine, and strictamine. Among these, echitamine is considered the most potent and consistently found across different geographical samples.

Phytochemical analyses have revealed that *Alstonia scholaris* contains a diverse range of compounds distributed across its anatomical parts. Over 300 phytochemicals have been isolated from the *Alstonia* genus. The leaves specifically contain compounds like quercetin, ursolic acid, alstonamide, alschomine, scholaricine, isorhamnetin-3-O-β-D-galactopyranoside, streptomycerol, β-sitosterol, akuammidine, angustilobine-B, losbanine, rhuzimanine, and laguamine (Chhajed *et al*., 2023) [2]. Both *in vivo* and *in vitro* models have confirmed the

anti-inflammatory and analgesic properties of its ethanolic leaf extracts and major alkaloids (Verma *et al*., 2024) [10]. In Indian traditional medicine, the leaves, latex and bark of *A. scholaris* are used to treat chronic conditions including asthma, jaundice, cancer, malaria and leprosy (Baliga *et al*., 2012) [1]. The latex is specifically valued for its wound-healing, ulcer-treating, and analgesic properties, particularly in rheumatoid arthritis (Singh *et al*., 2017) [8]. The plant is further documented to possess analgesic, anticancer, anti-inflammatory, antitumor, and laxative activities. Its extracts have shown both antifungal and antibacterial activity. The bark serves as a tonic and is used to treat chronic diarrhea, dysentery, indigestion, and typhoid.

*Staphylococcus aureus* is a Gram-positive, spherical bacterium. It is a common component of the human microbiota, frequently colonizing the skin and upper respiratory tract. It is catalase and nitrate-positive, and facultatively anaerobic. Although *S. aureus* is often a commensal, it can become pathogenic under certain conditions, causing a range of infections. It is a well-known agent of food poisoning due to its ability to produce enterotoxins and it is a major cause of osteomyelitis, septic arthritis and other musculoskeletal infections.

According to Palanisamy *et al*. (2024) [7], antimicrobial screening of 60 plant extracts revealed that many demonstrated significant activity. For instance, the ethyl acetate extract of *Garcinia travancorica* exhibited zones of inhibition against *Lactobacillus acidophilus* (17 mm) and *S. aureus* (16 mm), while chloroform extract of *Orthosiphon aristatus* showed broad-spectrum efficacy. Furthermore, Shetty *et al*., (2014) [9] reported the biosynthesis of stable silver nanoparticles (AgNPs) using *A. scholaris* bark extract. These nanoparticles, with an average size of 50 nm, displayed potent antimicrobial activity against various bacteria and fungi at a concentration of 170 ppm. Khyade and Vaikons (2009) [5] also reported that methanolic extracts of *A. scholaris* exhibited a broad spectrum of antibacterial activity against both Gram-positive and Gram-negative bacteria.

The present study was designed to evaluate the antibacterial potential of *Alstonia scholaris* leaf extracts against *Staphylococcus aureus*. Extracts were prepared using different solvents and their activity was compared with that of standard commercial antibiotics to assess their therapeutic potential.

## Materials and Methods

Leaves of *Alstonia scholaris* (commonly known as Saptaparni) were used to evaluate their antibacterial activity against *Staphylococcus aureus*. Fresh leaves were collected, thoroughly washed and air-dried at room temperature for 3-4 days. Once completely dried, the leaves were ground into a fine powder. Leaf extracts were prepared using five different solvents: methanol, acetone, ethanol, chloroform, and distilled water. For each extract, 1 gram of powdered leaf material was mixed with 10 ml of the respective solvent using a vortex mixer for 2 minutes. The mixtures were then allowed to stand at room temperature for 5 days. After the soaking period, the solutions were shaken vigorously, followed by centrifugation at 4000 rpm for 10 minutes. The supernatant was collected and used as the extract for antibacterial testing.

A pure culture of *Staphylococcus aureus* was obtained from Dr. Panjabrao Deshmukh Memorial Medical College, Amravati, (MS). Agar medium was prepared and poured into sterile Petri dishes under aseptic conditions in a laminar airflow cabinet. Once the agar solidified, 5 µl of *S. aureus* culture was inoculated onto the surface using a micropipette and uniformly spread with 'L'-shaped glass spreader. Three Petri plates were prepared for the study.

The first plate was used to assess the antibacterial activity of the solvents alone by placing Whatman paper discs soaked in each solvent.

The second plate was used as a positive control, where standard antibiotics (Vancomycin Hydrochloride and Maczone) were tested.

The third plate was used to evaluate the antibacterial effect of *A. scholaris* leaf extracts prepared in different solvents.

All plates were incubated at 37°C for 24 hours. After incubation, the plates were examined for zones of inhibition around the discs. The diameter of each zone was measured in millimeters using a ruler to determine antibacterial activity.

## Observations

The study aimed to assess the antibacterial activity of *Alstonia scholaris* (Saptaparni) leaf extracts against *Staphylococcus aureus*. Extracts were prepared using five different solvents: methanol, ethanol, acetone, chloroform and distilled water. After preparation, all extracts were centrifuged and filtered to ensure clarity and purity before testing. The *S. aureus* strain used for testing was obtained from Panjabrao Deshmukh Memorial Medical College, Amravati, (MS) and cultured in the laboratory under strict aseptic conditions.

To rule out any antibacterial effect of the solvents themselves, each solvent was tested independently. No zone of inhibition was observed in any of the solvent-only samples, confirming that the solvents did not exhibit any inherent antibacterial activity.

For comparison, commonly used antibiotics—Vancomycin Hydrochloride and Maczone—were tested against *S. aureus*. These antibiotics showed zones of inhibition measuring 28 mm and 20 mm respectively, after 24 hours of incubation.

When the *Alstonia scholaris* leaf extracts were tested, varying degrees of antibacterial activity were observed depending on the solvent used for extraction.

## Results

The distilled water extract of *Alstonia scholaris* showed the highest antibacterial activity against *Staphylococcus aureus*, with a zone of inhibition measuring 15 mm. The methanol extract followed closely with a 14 mm zone, while the acetone extract showed 13 mm. The ethanol extract demonstrated a zone of inhibition of 10 mm, and the chloroform extract showed the least activity with a 6 mm zone. These results clearly indicate that different solvents extract different bioactive components from the leaves, resulting in varied antibacterial effects.

## Conclusion

The findings of this study confirm that *Alstonia scholaris* possesses antibacterial activity against *Staphylococcus aureus*. The variation in the zone of inhibition across different solvents highlights the importance of solvent selection in extracting active antimicrobial compounds. Although the activity of the plant extracts was lower compared to standard antibiotics, their effectiveness—particularly with distilled water and methanol—suggests potential for development as a natural antibacterial agent. Given the rising resistance to conventional antibiotics and associated side effects, such plant-based alternatives warrant further investigation and development.

**Table 1:** Zone of Inhibition for Solvents used

| Test Organism | Zone of Inhibition For Solvents | | | | |
|---|---|---|---|---|---|
| *Staphylococcus Aureus* | Methanol | Acetone | Ethanol | Chloroform | Distilled water |
| | -- | -- | -- | -- | -- |

**Table 2:** Zone of Inhibition for Antibiotic used.

| Test Organism | Zone of Inhibition For Antibiotics | |
|---|---|---|
| *Staphylococcus Aureus* | **Vancomycin Hydrochloride** | **Maczone** |
| | 28 mm | 20 mm |

**Table 3:** Zone of Inhibition for Extracts in Solvents used.

| Test Organism | Zone of Inhibition for Extracts In Solvents | | | | |
|---|---|---|---|---|---|
| *Staphylococcus Aureus* | **Extract in Methanol** | **Extract in Acetone** | **Extract in Ethanol** | **Extract In Chloroform** | **Extract in Distilled water** |
| | 14 mm | 13 mm | 10 mm | 06 mm | 15 mm |

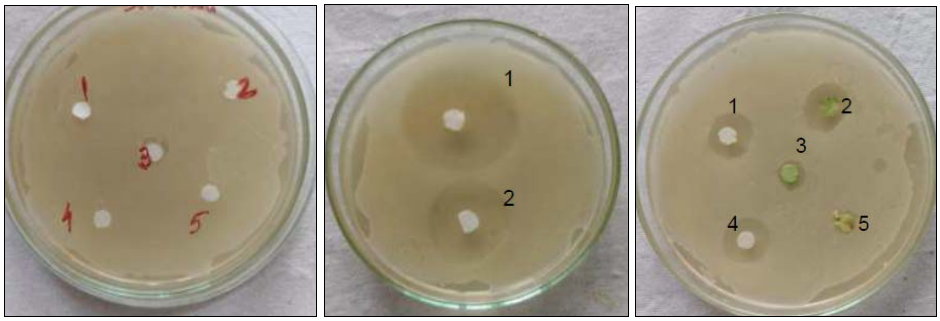


Plate 1 Plate 2 Plate 3

**Plate 1:** Solvents: Antimicrobial Test-1) Methanol, 2) Acetone, 3) Ethanol, 4) Chloroform, 5) Distilled Water

**Plate 2:** Antibiotics: Antimicrobial Test- 1) Vancomycin Hydrochloride, 2) Maczone

**Plate 3:** leaves extract in solvents Antimicrobial Test- 1) extract in Methanol 2) extract in Acetone 3) extract in Ethanol 4) extract in Distilled Water 5) extract in Chloroform

## References


1. Baliga MS. Review of the phytochemical, pharmacological and toxicological properties of *Alstonia scholaris* Linn. R. Br. (Saptaparna). Chin J Integr Med. 2012;28(1):1-4. https://doi.org/10.1007/s11655-011-0947-0
2. Chhajed M, Jain A, Pagariya A, Dwivedi S, Jain N, Taile V. *Alstonia scholaris* Linn. R. Br.: An assessment of its botany, conventional utilization, phytochemistry and pharmacology. Indian J Pharm Educ Res. 2023;17(33):184-203.
3. James J, Thaliyil VK, Dev L, Thankamni V. Phytochemical screening and antimicrobial activity of *Alstonia scholaris* flowers. Int J Pharm Res Dev. 2011;3(3):172-178.
4. Kachhawa JBS, Sharma N, Tyagi S, Gupta RS, Sharma K. Antibacterial activity of *Alstonia scholaris*: An in-vitro study. Int J Pharm Sci Rev Res. 2012;12(2):37-40.
5. Khyade MS, Vaikons NP. Phytochemical and antibacterial properties of leaves of *Alstonia scholaris* R. Br. Afr J Biotechnol. 2009;8(22):6434-6436.
6. Lalngaihmanawmil L, Lalthanpuii PB, Tlau L, Lalawmpuii L, Lalnundanga, Lalchhandama K. Phytochemical analysis and antioxidant and antibacterial activities of *Alstonia scholaris* from Mizoram, India. Plant Sci Today. 2023;X(X):1-7,15.
7. Palanisamy B, Pachaiyappan SK, Subramanian M, Das R, Savarimuthu I. Unraveling the antimicrobial efficacy and chemical fingerprinting of medicinal plants against the WHO's prioritized pathogens. Bull Natl Res Cent. 2024;48:11.
8. Singh H, Bhushan S, Arora R, Buttar HS, Arora S, Singh B. Alternative treatment strategies for neuropathic pain: Role of Indian medicinal plants and compounds of plant origin - A review. Biomed Pharmacother. 2017;92:634-650. https://doi.org/10.1016/j.biopha.2017.05.079
9. Shetty P, Supraja N, Garud M, Prasad TNVKV. Synthesis, characterization and antimicrobial activity of *Alstonia scholaris* bark-extract-mediated silver nanoparticles. J Nanostruct Chem. 2014;4:161-170. https://doi.org/10.1007/s40097-014-0132-z
10. Verma PK, Tukra S, Singh B, Sharma P, Abdi G, Bhat ZF. Unrevealing the ethnomedicinal potential of *Alstonia scholaris* (L.) R. Br.: A comprehensive review on its phytochemical, pharmacological and other applications. J Food Chem Adv. 2024;6:1-12.