Research Article **Phytopharmacology Research Journal (PRJ) Open Access**

Received: 16 July 2025 | Revised: 26 November 2025 | Accepted: 1 December 2025 | Published online: 1 December 2025 | ISSN : 2959-9687

Antimicrobial Profile *Azadirachta Indica* (Neem) Against Selected Pathogens

Tayyaba Bibi[1], Syeda Asma Bano[1], Muhammad Shakeel[2]*, Waqar Ali[3]

[1]Department of Microbiology,

The University of Haripur, KP, Pakistan

[2]Institute of Biotechnology and Microbiology,

Bacha Khan University Charsadda KP, Pakistan

[3]Institute of Biotechnology & Genetic

Engineering, Agriculture University, Peshawar,

KP, Pakistan

Correspondence

drshakeel@bkuc.edu.pk

Funding information

NA

Abstract

Azadirachta indica, a famous plant for its therapeutic benefits, has been reported by many authors. This study reported antibacterial and antifungal effects of *A. indica* leaf extracts against numerous fungal strains *i.e. Trichoderma viride*, *Rhizopus stolonifer*, *Aspergillus niger* and *Aspergillus flavus* and also the therapeutically relevant pathogens such as *Salmonella typhi, E. coli, Pseudomonas aeruginosa, and Klebsiella pneumoniae.* Antibacterial and antifungal activities of *A. indica* were determined by measuring the zones of inhibition by the Agar Well Diffusion Method. In the present study, four bacterial strains tested were inhibited by *A. indica* extracts with *K. pnemoniae* showing the highest activity at 0.3g/ml of 18mm and lowest activity shown by *S. typhi* at 0.4g/ml of 11 mm. Antifungal activities against *Rhizopus stolonifer* were inhibited with a zone of inhibition of 20mm. *A. indica* presents an effective and quick approach to introduce alternate therapeutic strategies, keeping in view the global issue of antibiotic resistance.

.

KEYWORDS

Azadirachta indica, Antibacterial, Phytotoxic, Insecticidal Activity

1.0 INTRODUCTION

Azadirachta indica, a highly valued tropical evergreen that belongs to the Kingdom Plantae, is sometimes referred to as the *A. indica* tree, Indian lilac, or margosa. Native to the Indian subcontinent, this amazing tree is now commonly grown across tropical and subtropical regions of the world because of its many advantages and flexibility. With a deep root system and a dense, round or oval crown, it usually reaches medium to large size. The tiny, fragrant white to pale yellow flowers are placed in drooping panicles, and the leaves are pinnate, bright green, and clustered at the ends of branches. The fruit has an oil-rich seed and is olive-sized and semi-sweet. *Azadirachta indica* is known as the "wonder tree," and the "reliever of sickness" (Arishtha in Sanskrit), and its significance extends to traditional medicine, agriculture, and environmental preservation. Using the diffusion method on PDA media, the antifungal activity of aqueous-methanol extracts and acetone exudates from 24 plant species was assessed *in vitro* against *Alternaria alternata*, *Botrytis cinerea*, *Phytophthora cambivora*, and *Fusarium oxysporum*. Salvia officinalis was the most efficient against three infections, while acetone exudates had the strongest antifungal efficacy. Neem is reported to has been used in Unani, Ayurveda and Homeopathic drugs and played key role in developing modern medicines[1]. People have previously used it to treat various health related issues, notably skin disease, inflammatory problems, and dental diseases.

It is also helpful in the treatment of eczema, acne, and psoriasis etc. There are reports that leaves of Neem represent important therapeutic functions and can be used effectively to cure anti-inflammatory effects, antiviral diseases, antimalarial and anti-oxidant, antibacterial, and other key health-related issues [2,3,4,5,6,7,8,9,10,11,12,13,14,15]. Neem branches can be utilized for cleaning teeth, relieving teeth pains, and deodorant. Its bark has been reported to present antimicrobial effects, notably antibacterial effects. The phytochemical composition of the neem tree has demonstrated potent antibacterial activity, which necessitates its detailed study of its phytochemicals. The major active ingredient of this plant is believed to be azadirachtin, which has the potential of possess several antimicrobial effects [15,16]. In addition to its therapeutic role, this plant has also been reported for its use as a skin moisturizer [17].

2.0 MATERIALS AND METHODS

2.1 Cultural Identification

In order to confirm the isolates' identities and rule out contamination, this procedure was crucial. Gram-negative bacteria like *E. coli and K. pneumoniae* were grown on Eosine Methylene Blue Agar (EMB), S. typhi were grown on Salmonella Shigella Agar (SS Agar) P. aeruginosa was grown on MacConkey agar, whereas fungal strains might have been grown on media like Potato Dextrose Agar (PDA).

2.2 Antibacterial Activity

The crude methanolic extracts of *F. benghalensis* aerial parts were screened for potential antibacterial activity against the test pathogens (Pseudomonas aeruginosa, Staphylococcus aureus, Salmonella typhi, and Escherichia coli). The antibacterial activity was performed against different test pathogens (Pseudomonas aeruginosa, Staphylococcus aureus, Salmonella typhi and Escherichia coli) by using the agar-well diffusion method. Nutrient agar media was prepared, autoclaved and poured into sterilized petri plates. After incubation, the test organisms were inoculated into the sterile nutrient broth and incubated for 24 hrs at 37 °C. After sterility check, of bacterial lawn was prepared by using 18-24 hrs old bacterial culture. In the laboratory, we used a sterile borer to make small wells (6 mm) in the petri plates. The stock solutions (3 mg/ml of DMSO) were transferred into the respective wells, and the plates were incubated at 37°C for 24 hours. We used amoxicillin as a positive control and less than 1% DMSO as a negative control.

The zones of inhibition were recorded after 24 hrs of incubation at 37 ºC [18].

2.3 Antifungal Activity

Antifungal activity was performed against different fungal species (using the tube dilution method. Sabouraud dextrose agar (SDA) media was prepared, it was autoclaved, and 4 mL was poured into sterilized test tubes along with 66.6µl from the stock solutions (24mg/mL DMSO). To ensure sterility, the slants were placed in an incubator at 28±1 oC for 24 hrs. The 7 day old fungal culture was introduced into the test tubes and allowed to incubate for 7 days at 28±1 oC. The positive control was Micnozole, while the negative control was DMSO. The results were measured on the 7th day by assessing the linear growth of inhibition [19].

RESULTS AND DISCUSSION

3.1 1 Antibacterial activity of A. indica against *S.typhi.* At all three doses, the A. indica extract exhibited strong antibacterial activity against *Salmonella typhi*. The extract suppressed growth with a zone of inhibition of 13 mm at 0.3 g/ml, 11 mm at 0.4 g/ml, and 12 mm at 0.5 g/ml.

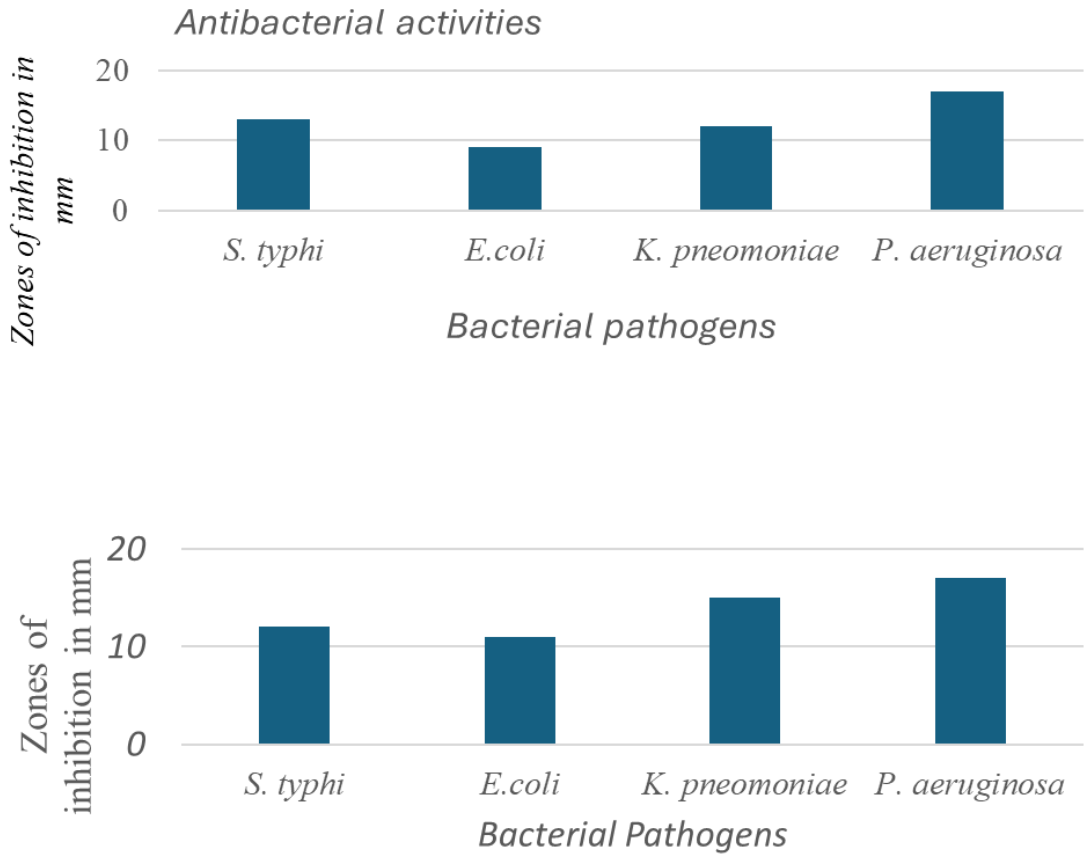

Figure 2. Insecticidal activity of crude methanolic extract of *S. cumini* STD (*Permithrine*)

4.0 DISCUSSION

The antibacterial activity of the was evaluated against

six bacterial strains, including *Bacillus subtilis*, *Staphylococcus aureus*, *Enterobacter*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Salmonella Typhi*. The results demonstrated that significantly inhibited bacterial growth, with the highest inhibition observed for *S. aureus* and *K. pneumoniae* which is consistent with findings in other studies on iron oxide nanoparticles. These exhibited effective bactericidal action, suggesting their potential application in combating multidrug-resistant (MDR) strains, which are a major global health concern [30]. The antimicrobial activity of the S. cumini leaves hydroalcoholic extract may be due to tannins and other phenolic constituents. *S. cumini* is known to be very rich in gallic and ellagic acid polyphenol derivatives [31]. Also, acylated flavonol glycosides, kaempferol, myricetin, and other polyphenols were isolated from *S. cumini* leaves [32].

CONCLUSION

This study demonstrates the successful use of *S. cumini crude* extract, providing an eco-friendly, cost-effective, and scalable method for medicinal production. The presence of key phytochemicals in the extract played a crucial role in the reduction and stabilization. The leaves and roots extract provide significant antibacterial activity against pathogenic bacterial strains, including *S. aureus* and *K. pneumoniae*, highlighting their potential for medical applications. Furthermore, the nanoparticles demonstrated antioxidant properties, supporting their broader therapeutic potential. This approach not only provides an effective method for producing therapeutic products but also contributes to the development of sustainable products for biomedical and industrial use.

ACKNOWLEDGMENTS

We are very thankful to the Institute of Biotechnology & Microbiology Bacha Khan University Charsadda, Khyber Pakhtunkhwa, Pakistan for providing support to conduct this research.

CONFLICT OF INTEREST

Authors express no conflict of interest

1. REFERENCES

1. S. Husain, Z. Urbi, H. Karuniawati, R. B. Mohiuddin, A. Moh Qrimida, A. M. M. Allzrag, & R. Capasso "Andrographis paniculata (burm. F.) wall. Ex needs: an updated review of phytochemistry, antimicrobial pharmacology and clinical safety and efficacy" Life, 11(4), 348 (2021).
2. R. K. Bijauliya, S. Alok, M. Singh, & S. B. Mishra "Morphology, Phytochemistry and pharmacology of Syzygium cumini (Linn.)-an overview" *J. Int. Phar. Sci. Res*, 8(6), 2360-2371 (2017).
3. M. J. Bandar, K. R. Chandra sekhar, & K. M. Kaveriappa "Medical ethno botany of the siddis of Uttar Kannada district, Karnataka, India" *J. ethno*, 47(3), 149-158 **(**2015).
4. A. F. Faria, M. C. Marques, & A. Z. Mercadante "Identification of bioactive compounds from jambolão (Syzygium cumini) and antioxidant capacity evaluation in different pH conditions" Food chemistry, 126(4), 1571-1578 (2011).
5. D. Bhowmik, H. Gopinath, B. P. Kumar, & K. S. Kumar "Traditional and medicinal uses of Indian black berry" *J. Pharma Phytochem*1(5), 36-41 (2013).
6. A. P. Wardana, N. Hidayati & K. Shimizu "An ellagic acid derivative and its antioxidant activity of chloroform extract of stem bark of Syzygium polycephalum Miq.(Myrtaceae*)" J. Indo. Chem*, 18(1), 26-34 (2018)
7. L. A. Craven, & E. Biffin "An infrageneric classification of Syzygium (Myrtaceae)" Blumea-Biodiversity, Evolution and Biogeography of Plants, 55(1), 94-99 (2010).
8. H. Sagrawat, A. S. Mann and M. D. Kharya "Pharmacological potential of Eugenia jambolana: a review" 96-105 (2006).
9. N. Chhikara, R. Kaur, S. Jaglan, P. Sharma, Y. Gat & A. Panghal "Bioactive compounds and pharmacological and food applications of Syzygium cumini–a review" Food & function, 9(12), 6096-6115 (2018).

10. A. Kochhar, M. Nagi, & R. Sachdeva "Proximate composition, available carbohydrates, dietary fibre and anti nutritional factors of selected traditional medicinal plant" *J. Hum. Ecol*. 19(3), 195-199 (2006).
11. D. C. Modi, J. K. Patel, B. N. Shah, & B. S. Nayak "Pharmacognostic studies of the seed of Syzygium cumini Linn"*J. Pharma. Sci. Mon*., 1(1), 20-26 (2010).
12. P. Comoli, S. Binggeli, F. Ginevri, & H. H. Hirsch "Polyomavirus-associated nephropathy: update on BK virus-specific immunity" Transplant Infectious Disease, 8(2), 86-94 (2006).
13. K. S Banu, & L. Cathrine, General techniques involved in phytochemical analysis. *J. Int. Adv. Res. Che. Sci*, 2(4), 25-32. (2015).
14. R. Gul, S. U. Jan, S. Faridullah, S. Sherani, & N. Jahan, Preliminary phytochemical screening, quantitative analysis of alkaloids, and antioxidant activity of crude plant extracts from Ephedra intermedia indigenous to Balochistan. *J. World. Scient*, 2017(1), 5873648 (2017).
15. E. Fachriyaha, D. Kusrinia, & I. B. Haryanto, "Phytochemical Test, Determination of Total Phenol, Total Flavonoids and Antioxidant Activity of Ethanol Extract of Moringa" Jurnal Kimia Sains dan Aplikasi, 23(8), 290-294 (2020).
16. S. V. Thite, Y. R. Chavan, V. T. Aparadh, & B. A. Kore, "Preliminary phytochemical screening of some medicinal plants" *J. Int. Pharm. Chem. Biol. Sci*, 3(1), 87-90 (2013).
17. A. M. Lanjewar, D. Sharma, K. V. Kosankar & K. Thombre, "Extraction and phytochemical screening of Syzygium cumini seeds in Vidarbha region of India" *World J. Pharm. Res*, *7*(5), 1782-91 (2018).
18. J. M. Veigas, M. S. Narayan, P. M. Laxman, & B. Neelwarne, "Chemical nature, stability and bioefficacies of anthocyanins from fruit peel of Syzygium cumini Skeels" *Food ChemisT*, *105*(2), 619-627 (2007). .
19. B. Ahmad, S. Azam, S. Basher, I. Khan, A. Adhikari, & M. I. Choudhary "Anti-inflammatory and enzyme inhibitory activities of a crude extract and a pterocarpan isolated from the aerial parts of Vitexagnus-castus" *J. Biotech*, 5(11), 1207-1215 (2010).
20. M. D. C Sales, H. B. Costa, P. M. B Fernandes, J. A Ventura & D. D Meira. Antifungal activity of plant extracts with potential to control plant pathogens in pineapple. *J. Asia. Paci. Trop Biomed*, 6(1), 26-31 (2016).
21. B. Ahmad, S. Naz, S. Azam, I. Khan, S. Basher, & F. Hassan "Antimicrobial, Phytotoxic, Heamagglutination, Insecticidal and Antioxidant Activities of the Fruits Of Sarcococca Salina (D. Don) Mue". Pak. J. Bot, 47, 313-319 (2015).
22. J. M. McPartland, & Z. Sheikh "A review of Syzygum cumini-based insecticides, Matricides, and repellents" *J. Entomology. Zool. Stud*, 6(6), 1288. (2018).
23. Kumar, A.; Ilavarasan, R.; Jayachandran, T.; Deecaraman, M.; Aravindan, P.; Padmanabhan, N.; Krishan, M. R. V. Anti-diabetic activity of Syzygium cumini and its isolated compound against streptozotocin-induced diabetic rats. J. Med. Plant Res. 2008, 2(9), 246-249].
24. A. Jeyasankar, N. Raja, & S. Ignacimuthu "Insecticidal compound isolated from syzygium lineare wall.(Myrtaceae) against Spodoptera litura (Lepidoptera: Noctuidae)" J. Saudi. Bio. Sci, 18(4), 329-332 (2011).
25. Kumar, A.; Ilavarasan, R.; Jayachandran, T.; Deecaraman, M.; Aravindan, P.; Padmanabhan, N.; Krishan, M. R. V. Anti-diabetic activity of Syzygium cumini and its isolated compound against streptozotocin-induced diabetic rats. J. Med. Plant Res. 2008, 2(9), 246-249].