

Green Synthesis of Mimosa Pudica Mediated Selenium Nanoparticles and its Antibacterial Activity against Escherichia coli and Staphylococcus Aureus

DhavePrasath V M¹, Prabhakaran A K², Rajeshkumar Shanmugam^{1*},
Dhanyaa Muthukumaran¹

¹*Nanobiomedicine Lab, Centre for Global Health Research, Saveetha Medical College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Chennai, India*

²*Department of Orthopaedics, Saveetha Medical College and Hospital, Saveetha Institute of Medical and Technical Sciences, Chennai, India*

Email: rajeshkumars.smc@saveetha.com

Nanoparticles have gained significant importance in various fields due to their unique properties and potential applications. In this study, we examined the green synthesis of selenium nanoparticles (Se NPs) using *M. pudica* extract and their antibacterial activity against *Escherichia coli* (*E. coli*) and *Staphylococcus aureus* (*S. aureus*). The Se NPs were synthesized through a sustainable and environmentally friendly method, highlighting the growing interest in green synthesis approaches. The antibacterial activity was confirmed by a time-kill assay, which showed a dose-dependent decline in optical density with increasing concentrations of Se NPs. The green synthesis of Se NPs using *M. pudica* extract offers advantages such as sustainability, cost-effectiveness, and reduced environmental impact. The study highlights the successful synthesis of *M. pudica*-mediated Se NPs and their significant antibacterial activity against *E. coli* and *S. aureus*, suggesting their potential as effective antibacterial agents.

Keywords: Antibacterial Activity, Green Synthesis, Novel technique, Selenium Nanoparticles.

1. Introduction

Nanoparticles are increasingly being employed across a variety of industries, including medical, environmental cleanup, and industrial applications (Ziental et al. 2020). In the medical field, they are widely used for imaging and therapy of the central nervous system (Provenziale and Silva 2009). *M. pudica*, commonly known as the touch-me-not, shy, or sensitive plant, is well-known for its therapeutic benefits. It is commonly found in moist waste

ground, lawns, open plantations, and weedy thickets (Ahmad et al. 2012). This plant is a member of the Fabaceae family and the Mimosoideae subfamily. In many countries, plants are used as medicine and as sources of a variety of potent medications (Muhammad et al. 2016). Recently, selenium nanoparticles (SeNPs) synthesized using herbal extract have attracted significant attention due to their unique characteristics and potential applications (Johnson et al., 2024, Kathiravana et al., 2023, SHanmugam et al., 2023, Jayapriya et al., 2022).

In recent studies, *M. pudica* leaf extract has been used as a reducing and stabilizing agent to create SeNPs. Selenium (Se) is an essential micronutrient that plays a crucial role in various biological processes such as DNA synthesis, immune function, and thyroid hormone metabolism (Kieliszek et al. 2022). SeNPs have garnered considerable attention due to their unique properties and potential applications in fields such as medicine, agriculture and industry (Tarmizi et al. 2023; Song et al. 2023; Ferrari et al. 2023). SeNPs possess unique physical and chemical properties such as small size, large surface area, high reactivity, and stability, making them suitable for various applications (Yang et al. 2023). They can be synthesized using chemical reduction, physical methods, and biological methods. Among these, the biological synthesis of SeNPs using microorganisms, plants, and algae is favoured for its eco-friendly and cost-effective approach (Hussein et al. 2022).

S. aureus, often found on healthy people's skin and in their noses, can become opportunistic under specific circumstances, causing infections that range from mild to life-threatening. It can cause skin infections, staphylococcal food poisoning, methicillin-resistant *S. aureus* (MRSA) infections, and osteomyelitis (Lara et al. 2005). *Escherichia coli* (*E. coli*), a Gram-negative bacterium, is naturally found in the intestines of humans and animals. While most strains are benign and beneficial, some can cause serious infections when they spread to other parts of the body or are ingested (Kolenda et al. 2015). *E. coli* infections can range from minor gastrointestinal discomfort to severe, life-threatening conditions such as gastroenteritis, urinary tract infections, hemolytic uremic syndrome, and septicemia (Allocati et al. 2013).

The use of *M. pudica* in the green synthesis of SeNPs offers several benefits over traditional synthesis techniques. It is an economical and environmentally friendly approach with fewer negative effects on the environment and human health (Benelli 2019). Additionally, the SeNPs produced by *M. pudica* exhibit unique properties and have potential applications in medicine, agriculture, and manufacturing.

The green synthesis of selenium nanoparticles (SeNPs) using *M. pudica* extract and evaluates their antibacterial activity against *E. coli* and *S. aureus*. The synthesis process utilizes the leaf extract of *M. pudica* as a reducing and stabilizing agent, emphasizing the sustainable and eco-friendly nature of this approach. The study aims to determine the Antibacterial activity and time-kill curve analysis of the SeNPs against *E. coli* and *S. aureus*, providing insights into their potential as antibacterial agents.

2. Materials and Methods

2.1 Preparation of Mimosa pudica Se NPs:

To prepare *Mimosa pudica*-mediated Se NPs, 1g of powdered *M. pudica* was mixed with 100 ml of distilled water and boiled to obtain a filtered solution. Separately, 0.356 g of sodium

Nanotechnology Perceptions Vol. 20 No. S7 (2024)

selenite was dissolved in 50 ml of distilled water. Subsequently, 50ml of the *M. pudica* solution was mixed with 50ml of the sodium selenite solution.

2.2 Antimicrobial activity

The antimicrobial activity of green synthesized Se NPs was evaluated using the agar well diffusion method on Mueller Hinton agar plates, which were sterilized and inoculated with *E. coli* and *S. aureus*. Wells were filled with varying concentrations (25 µg, 50 µg, 100 µg) of selenium nanoparticles, with plant extract as standard. Plates were incubated at 37°C for 24 hours for bacterial cultures. The inhibition zones around the wells were measured and recorded in millimetresto assess antimicrobial activity.

2.3 Time kill curve assay

The antibacterial activity of *M.pudica*- Se NPs was assessed using a time-kill assay. *S. mutans* and *E.coli* were cultured in Mueller Hinton broth and exposed to different concentrations of 25, 50, and 100 µg/ml and standard antibiotic at 100 µg/ml in 96 well plates for varied time intervals (1,2,3,4 hours). 30 ml of the inoculum was mixed with 15 mL of Mueller Hinton Broth medium that had already been heated and was free of microbes. Then, 90 mL of this mixture was spread evenly over each well of a 96-well ELISA plate. Then, a time-kill curve experiment was done to test the antibacterial qualities.

3. Results

3.1 Visual observation

The green synthesis of *M.pudica*-mediated SeNPs was successfully achieved. The formation of SeNPs was confirmed by the distinct Reddish-orange coloration of the final solution, indicating the successful synthesis of selenium nanoparticles.

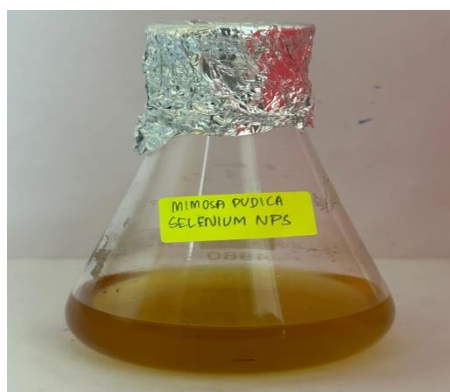


Figure 1: *M.pudica*-Mediated Selenium Nanoparticles

3.2 Anti-bacterial Activity

The antimicrobial activity of *M.pudica*-mediated SeNPs against *E. coli* and *S. aureus* was assessed using the agar well diffusion method. The SeNPs demonstrated significant antibacterial activity, with inhibition zones increasing in a dose-dependent manner. At 100

$\mu\text{g/mL}$, the SeNPs produced the largest inhibition zones, measuring 9 mm for *E. coli* and 13.6 mm for *S. aureus*, comparable to the standard antibiotic used. These results confirm the potential of green-synthesized SeNPs as effective antibacterial agents against both Gram-positive and Gram-negative bacteria.

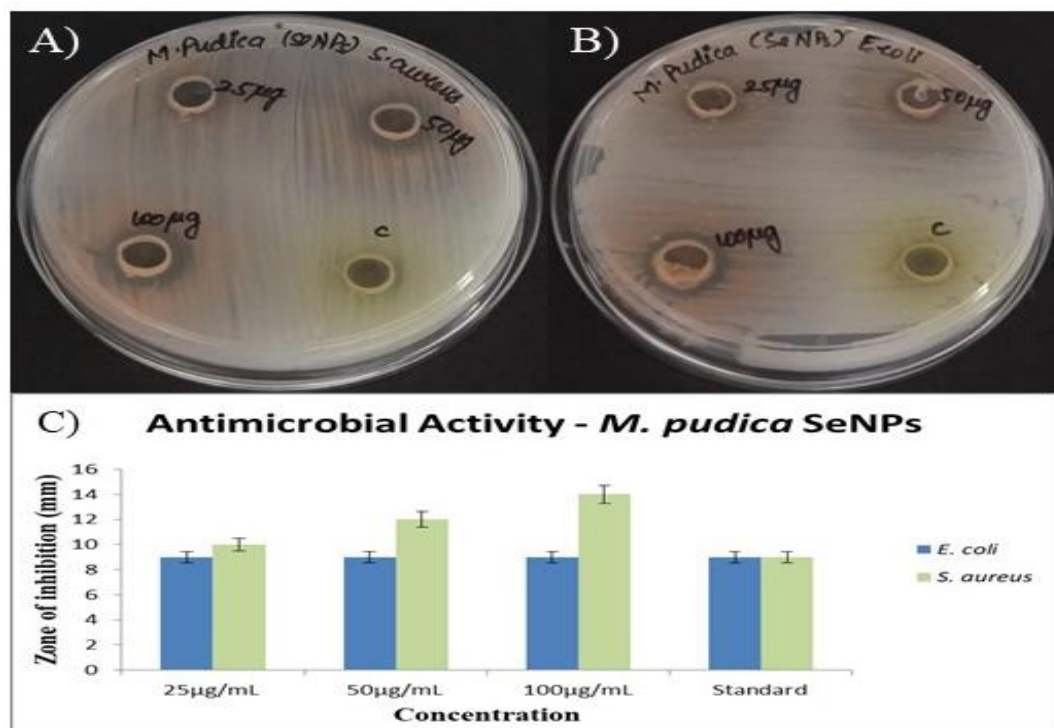


Figure 2: A) Antimicrobial activity of Se NPs against *S. aureus*. B) Antimicrobial activity of Se NPs against *E. coli*. C) Graphical representation of the antimicrobial activity of Se NPs.

3.3 Time kill analysis

The time-kill assay results show that *M. pudica*-mediated SeNPs effectively reduce bacterial cell viability in a dose-dependent manner for both *E. coli* and *S. aureus*. OD measurements at 600 nm over 1, 2, 3, and 4 hours indicate that higher concentrations of SeNPs (25 $\mu\text{g/mL}$, 50 $\mu\text{g/mL}$, 100 $\mu\text{g/mL}$) lead to a significant decline in bacterial growth. The 100 $\mu\text{g/mL}$ concentration exhibited the greatest reduction in optical density, comparable to the standard antibiotic treatment, whereas the control showed the highest bacterial growth. These findings underscore the potent antibacterial activity of green-synthesized SeNPs against both *E. coli* and *S. aureus*.

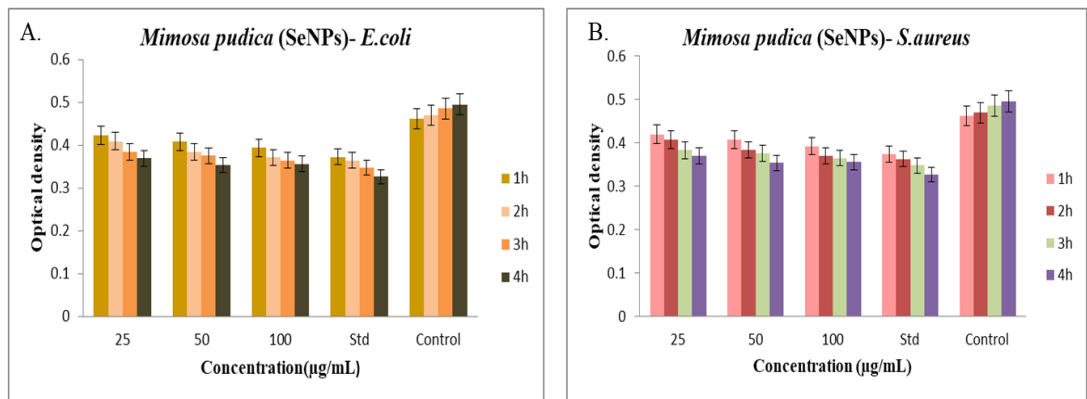


Figure 3: Time-kill assay showing the antimicrobial activity of *M. pudica*-mediated SeNPs against *E. coli* (A) and *S. aureus* (B) at varying concentrations over 1, 2, 3, and 4 hours.

4. Discussion

The application of *M. pudica*-mediated SeNPs as antibacterial agents against *E. coli* and *S. aureus*, demonstrates their potent antibacterial properties. The results from the antimicrobial activity assays indicate that SeNPs exhibit significant antibacterial effects, with a clear dose-dependent inhibition of bacterial growth. In the agar well diffusion method, higher concentrations of SeNPs (100 µg/mL) produced larger zones of inhibition for both *E. coli* and *S. aureus*, comparable to the standard antibiotic used. This highlights the effectiveness of these nanoparticles in inhibiting bacterial proliferation (Mandal et al. 2022). The time-kill assay further supports these findings, showing a substantial reduction in bacterial cell viability over time at various SeNPs concentrations. Specifically, the 100 µg/mL concentration of SeNPs led to a pronounced decline in optical density within 4 hours, indicating a potent bactericidal effect similar to that of the standard antibiotic treatment. This consistency across both bacterial strains underscores the broad-spectrum potential of green-synthesized SeNPs. Comparing the results of both assays, it is evident that *M. pudica*-mediated SeNPs effectively inhibit bacterial growth in a dose-dependent manner (Appiah et al. 2017).

The use of *M. pudica* in the green synthesis of SeNPs offers multiple benefits, including an eco-friendly and cost-effective approach and leveraging the plant's inherent medicinal properties. This study suggests that green-synthesized SeNPs can serve as a viable alternative to conventional antibiotics, particularly in combating the global challenge of antibiotic resistance.

M. pudica-mediated SeNPs demonstrate significant antibacterial activity against *E. coli* and *S. aureus*, as evidenced by the inhibition zones in the agar well diffusion method and the optical density reduction in the time-kill assay. These findings support the potential application of green-synthesized SeNPs as effective and sustainable antibacterial agents.

5. Conclusion

The green synthesis method offers an eco-friendly and cost-effective alternative to conventional antibacterial treatments. These findings highlight the potential of green-synthesized SeNPs as effective antibacterial agents, suggesting their use in addressing antibiotic resistance and promoting sustainable medical practices. Future research should further explore the mechanisms of antibacterial action and broader applications of these nanoparticles.

Funding Details

No funding was acquired for carrying out this study.

Disclosure Statement

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

1. Aguechari, Nassim, Achraf Boudiaf, and Mohand Ould Ouali. "Effect of artificial aging treatment on microstructure, mechanical properties and fracture behavior of 2017A alloy." *Metallurgical and Materials Engineering* 28.2 (2022): 305-318.
2. Negussie, Mengistu, Teshale Woldeamanuel, and Tewodros Tefera. "Analysis of teff market chain: evidence from South Gondar Zone, Ethiopia." *Acta Innovations* (2022).
3. Johnson J, Shanmugam R, Manigandan P (June 30, 2024) Characterization and Biomedical Applications of Green-Synthesized Selenium Nanoparticles Using *Tridaxprocumbens* Stem Extract. *Cureus* 16(6): e63535. doi:10.7759/cureus.63535.
4. Kathiravan, A., Udayan, E., Rajeshkumar, S. et al. Unveiling the Biological Potential of Mycosynthesized Selenium Nanoparticles from Endophytic Fungus *Curvularia* sp. LCJ413. *BioNanoSci.* (2023). <https://doi.org/10.1007/s12668-023-01223-w>.
5. Jayapriya Johnson, Rajeshkumar Shanmugam and Thangavelu Lakshmi A review on plant-mediated selenium nanoparticles and its applications *J PopulTherClinPharmacol* Vol 28(2):e29–e40; DOI <https://doi.org/10.47750/jptcp.2022.870>
6. Shanmugam R, Anandan J, Balasubramanian A K, et al. (December 20, 2023) Green Synthesis of Selenium, Zinc Oxide, and Strontium Nanoparticles and Their Antioxidant Activity - A Comparative In Vitro Study. *Cureus* 15(12): e50861. doi:10.7759/cureus.50861.
7. Allocati, N., Masulli, M., Alexeyev, M., & Di Ilio, C. (2013). *Escherichia coli* in Europe: An Overview. *International Journal of Environmental Research and Public Health*, 10(12), 6235–6254. <https://doi.org/10.3390/ijerph10126235>
8. Appiah, T., Boakye, Y. D., & Agyare, C. (2017). Antimicrobial Activities and Time-Kill Kinetics of Extracts of Selected Ghanaian Mushrooms. *Evidence-based Complementary and Alternative Medicine*, 2017, 1–15. <https://doi.org/10.1155/2017/4534350>
9. Benelli, G. (2019). Green Synthesis of Nanomaterials. *Nanomaterials*, 9(9), 1275. <https://doi.org/10.3390/nano9091275>
10. Ferrari, L., Cattaneo, D. M., Abbate, R., Manoni, M., Ottoboni, M., Luciano, A., Von Holst, C., & Pinotti, L. (2023). Advances in selenium supplementation: From selenium-enriched yeast to potential selenium-enriched insects, and selenium nanoparticles. *Animal Nutrition*, 14, 193–203. <https://doi.org/10.1016/j.aninu.2023.05.002>
11. Gómez, Raúl José Martelo, Piedad Mary Martelo Gómez, and David Antonio Franco Borré. *Nanotechnology Perceptions* Vol. 20 No. S7 (2024)

- "Assessment of Parameters in Integrated Photonic Systems Through the Analysis of Interdependencies with the MICMAC Technique." *Nanotechnology Perceptions* 20.2 (2024): 39–52.
12. Hussein, H. G., El-Sayed, E. S. R., Younis, N. A., Hamdy, A. E. H. A., &Easa, S. M. (2022). Harnessing endophytic fungi for biosynthesis of selenium nanoparticles and exploring their bioactivities. *AMB Express*, 12(1). <https://doi.org/10.1186/s13568-022-01408-8>
13. Kieliszek, M., Bano, I., & Zare, H. (2021). A Comprehensive Review on Selenium and Its Effects on Human Health and Distribution in Middle Eastern Countries. *Biological Trace Element Research*, 200(3), 971–987. <https://doi.org/10.1007/s12011-021-02716-z>
14. Kolenda, R., Burdukiewicz, M., & Schierack, P. (2015). A systematic review and meta-analysis of the epidemiology of pathogenic *Escherichia coli* of calves and the role of calves as reservoirs for human pathogenic *E. coli*. *Frontiers in Cellular and Infection Microbiology*, 5. <https://doi.org/10.3389/fcimb.2015.00023>
15. Lowy, F. D. (1998). *Staphylococcus aureus*Infections. *New England Journal of Medicine*~ the æNew England Journal of Medicine, 339(8), 520–532. <https://doi.org/10.1056/nejm199808203390806>
16. Mandal, A. K., Pandey, A., Sah, R. K., Baral, A., & Sah, P. (2022). In Vitro Antioxidant and Antimicrobial Potency of *Mimosa pudica* of Nepalese Terai Region: Insight into L-Mimosine as an Antibacterial Agent. *Evidence-based Complementary and Alternative Medicine*, 2022, 1–11. <https://doi.org/10.1155/2022/6790314>
17. Muhammad, G., Hussain, M. A., Jantan, I., & Bukhari, S. N. A. (2015). *Mimosa pudica* L., a High-Value Medicinal Plant as a Source of Bioactives for Pharmaceuticals. *Comprehensive Reviews in Food Science and Food Safety*, 15(2), 303–315. <https://doi.org/10.1111/1541-4337.12184>
18. Provenzale, J., & Silva, G. (2009). Uses of Nanoparticles for Central Nervous System Imaging and Therapy. *American Journal of Neuroradiology*, 30(7), 1293–1301. <https://doi.org/10.3174/ajnr.a1590>
19. Song, J., Yu, S., Yang, R., Xiao, J., & Liu, J. (2023). Opportunities for the use of selenium nanoparticles in agriculture. *NanoImpact*, 31, 100478. <https://doi.org/10.1016/j.impact.2023.100478>
20. Tarmizi, A. a. A., Ramli, N. N. N., Adam, S. H., Mutalib, M. A., Mokhtar, M. H., & Tang, S. G. H. (2023). Phytofabrication of Selenium Nanoparticles with *Moringaoleifera* (MO-SeNPs) and Exploring Its Antioxidant and Antidiabetic Potential. *Molecules*, 28(14), 5322. <https://doi.org/10.3390/molecules28145322>
21. Yang, Z., Hu, Y., Yue, P., Li, H., Wu, Y., Hao, X., & Peng, F. (2023). Structure, stability, antioxidant activity, and controlled-release of selenium nanoparticles decorated with lichenan from *Usnealongissima*. *Carbohydrate Polymers*, 299, 120219. <https://doi.org/10.1016/j.carbpol.2022.120219>
22. Ziental, D., Czarczynska-Goslinska, B., Mlynarczyk, D. T., Glowacka-Sobotta, A., Stanis, B., Goslinski, T., & Sobotta, L. (2020). Titanium Dioxide Nanoparticles: Prospects and Applications in Medicine. *Nanomaterials*, 10(2), 387. <https://doi.org/10.3390/nano10020387>