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# Antibacterial Activity of Selenium Nanoparticles Synthesized using Mimosa Pudica against Wound Pathogens

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Selenium nanoparticles (Se NPs) have emerged as potent antibacterial agents with potential applications in wound healing. This research examines the antibacterial properties of Mimosa pudica-mediated Se NPs against common wound pathogens, including Streptococcus mutans and Escherichia coli. This study aimed to evaluate the antibacterial efficacy of selenium nanoparticles (Se NPs) synthesized using M. pudica extract against wound pathogens. Se NPs were synthesized using a green synthesis method involving M. pudica extract. The antibacterial activity was assessed using the agar well diffusion technique and a time-kill curve assay. Se NPs showed antimicrobial activity against other pathogens such as S. mutans and E.coli with inhibition zones around 9 mm. M. pudica-mediated Se NPs demonstrated strong antibacterial properties, making them effective candidates for treating wound infections. The study highlights the potential of Se NPs as an alternative antibacterial agent in wound healing applications.

**Keywords:** Antibacterial activity, Green synthesis, Mimosa pudica, Selenium nanoparticles, Wound pathogens.

## 1. Introduction

Bacteria are organisms capable of causing death by spreading infectious diseases, a threat recognized since the fourteenth century. The first antibacterial agent, salvarsan, was introduced in 1910 (Griffiths et al. 2010). Following this, other antibacterial substances such as macrolides, nalidixic acid, and chloramphenicol were widely employed, providing temporary relief from infectious bacterial infections during the 20th century. However, excessive antibiotic use led to the emergence of antibiotic-resistant microorganisms (Bennett et al. 2009). Efforts to combat these resistant strains have involved the development of novel antibiotics, the identification of bacteria producing antibiotics, and discovery of additional antibiotics from natural sources. Despite these efforts, bacterial strains resistant to treatment have continued to proliferate.

Nanotechnology has opened numerous opportunities in biology and medicine, including tissue engineering, drug delivery, diagnosis, imaging, and the fight against bacterial infections (Lay-Ekuakille 2010). Nanoparticles have been suggested as alternative treatments for infections, using mechanisms different from conventional antibiotics. Consequently, nanomaterials are considered potential alternatives to antibiotics in controlling bacterial infections (Alpaslan et al. 2017). Due to their unique properties, nanomaterials have seen tremendous development and study over the past few decades (Granozzi and Alonso-Vante 2019). Metallic nanoparticles can be created using chemical, physical, and biological methods. Chemical methods often involve toxic-reducing agents, while physical methods are energy-intensive and time-consuming (Yang 2012). These limitations make large-scale synthesis of nanoparticles less suitable for chemical and physical techniques.

Biological methods, utilizing plant, bacterial, algal, and fungal extracts, offer an environmentally friendly alternative for nanoparticle production (Shukla and Iravani 2018). Various plants have been successfully used to fabricate nanoparticles, including silver, zero-valent iron, copper, zinc oxide, and gold (Siddhardha et al. 2020; Shukla and Iravani 2018). Selenium, a trace mineral essential for mammals and humans, supports immune system and thyroid function, and prevents cellular damage (Banuelos et al. Yin 2013; Rustum 2019). Its reasonable antibacterial properties have led to its investigation as an anti-carcinogenic agent (Karthik et al. 2019; Fu 2023). selenium nanoparticles (Se NPs) offer low toxicity, biocompatibility, special therapeutic properties, and bioavailability, making them suitable for various biomedical applications, including photovoltaics, sensors, electrical devices, catalysis, antimicrobial agents, and chemoprevention (Sahu and Casciano 2009).

Green synthesis, using plant extracts as reducing agents, has proven to be the most practical and least harmful method for producing Se NPs (Hassan 2016). M. pudica, an annual or perennial creeping herb, is known in Ayurveda for its anti-asthmatic, analgesic, and depressive properties. Traditionally used to treat conditions such as alopecia, diarrhoea, dysentery, insomnia, tumours, and urogenital infections, M. pudica exhibits sedative, emetic, and tonic qualities (Ahmad et al. 2012; Parrotta 2001). Phytochemical studies have identified alkaloids, mimosine, flavonoids, sterols, terpenoids, tannins, and fatty acids in M. pudica (Ahmad et al. 2012; Parrotta 2001). The plant is also known for its rapid leaf movements in response to stimuli and its slow, periodic leaf movements regulated by a biological clock (Shridhar, Narayana, and Yathiraj 2015; Quan et al. 2023).

This study investigates the antibacterial activity of Se Nps synthesized from M. pudica against various microorganisms. The research aims to evaluate the effectiveness of these green-synthesized Se NPs against common wound pathogens, including S. mutans and E. coli. The study employs the agar well diffusion technique to measure the zones of inhibition created by different concentrations of Se NPs, indicating their antibacterial potency. Additionally, the study includes a time-kill curve assay to observe the bactericidal effects of Se NPs over time, providing insight into the kinetics of bacterial inhibition. The results of this research could demonstrate the potential of M. pudica-mediated Se NPs as effective antimicrobial agents, offering a novel and environmentally friendly approach to combating bacterial infections, particularly those associated with wounds.

## 2. Materials and Methods

### 2.1 Preparation of M. pudica-mediated selenium nanoparticles

1g of M. pudica was added to 100 ml of distilled water, heated to 50°C, and boiled for 10-15 minutes. The mixture was filtered by using a muslin cloth to obtain the extract. This plant extract was then mixed with sodium selenite solution.

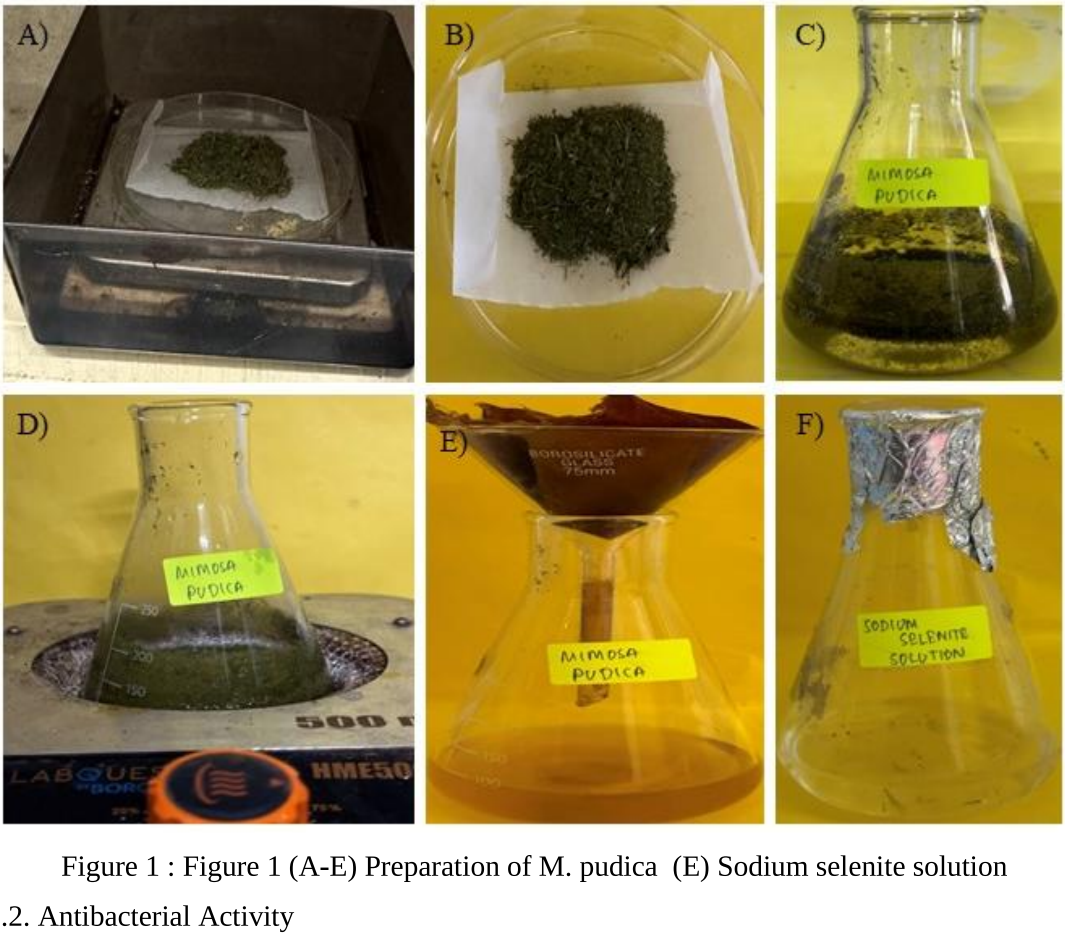


Figure 1 : Figure 1 (A-E) Preparation of M. pudica  (E) Sodium selenite solution

### 2.2. Antibacterial Activity

The antibacterial activity of M.pudica against S. mutans and E.coli was determined using the agar well diffusion technique. Mueller Hinton agar (MHA) plates were prepared and sterilized, and wells were made using a well-cutter. S. mutans and E.coli were swabbed on the agar surface and incubated at 37°C for 24 hours. The plant extract of M.pudica was used as a control. The cultures were treated with Se NPs at different doses (25,50,100 µg/mL). After incubation, the zone of inhibition surrounding the wells was evaluated to determine antibacterial efficiency. (Figure 2)

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. Result

3.1 Antimicrobial activity of Mimosa pudica

The antimicrobial activity of Se NPs synthesized using M. pudica extract was evaluated against Escherichia coli and Streptococcus mutans using the agar well diffusion method. The Se NPs exhibited significant antibacterial effects, with inhibition zones increasing with concentration. For Streptococcus mutans, inhibition zones measured 14 mm at 25 µg/mL, 17 mm at 50 µg/mL, and 19 mm at 100 µg/mL, comparable to the 20 mm zone produced by the standard antibiotic. For Escherichia coli, the inhibition zones were 10 mm at 25 µg/mL, 12 mm at 50 µg/mL, and 15 mm at 100 µg/mL, also demonstrating effective antibacterial activity.

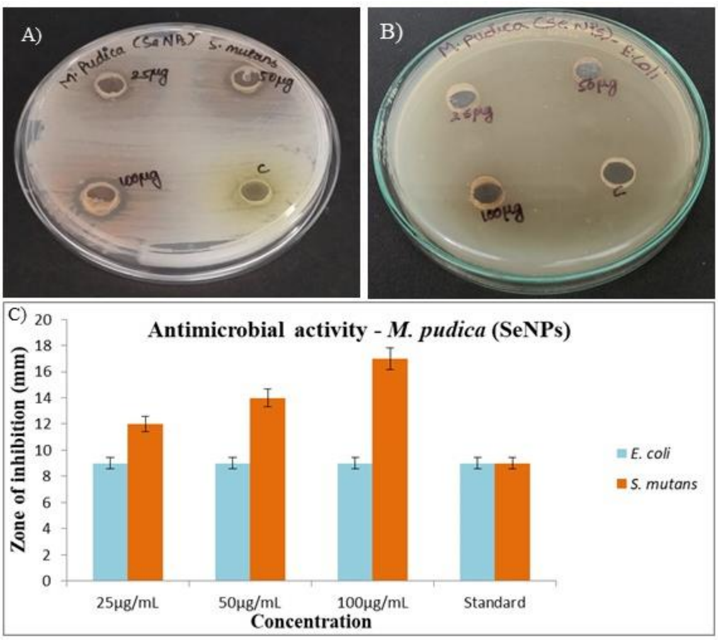


Figure 2: A) and B) show the inhibition zones at different concentrations of M.pudica against

S. mutans and E.coli C) Graphical representation of the antibacterial activity of M.pudica-mediated Se NPs against S. mutans and E.coli.

3.2 Effects of Mimosa pudica in time kill Assay

The time-kill assay results demonstrated that selenium nanoparticles (Se NPs) synthesized using M. pudica extract exhibited strong antibacterial activity against both S. mutans and E. coli. At higher concentrations (50 and 100 µg/mL), Se NPs significantly reduced the optical density of both bacterial strains over 4 hours, indicating potent bactericidal effects. The standard antibiotic showed similar efficacy, while the control group maintained higher optical density values, reflecting continuous bacterial growth. These findings suggest that M. pudica-mediated Se NPs are effective antimicrobial agents, particularly in treating wound infections.

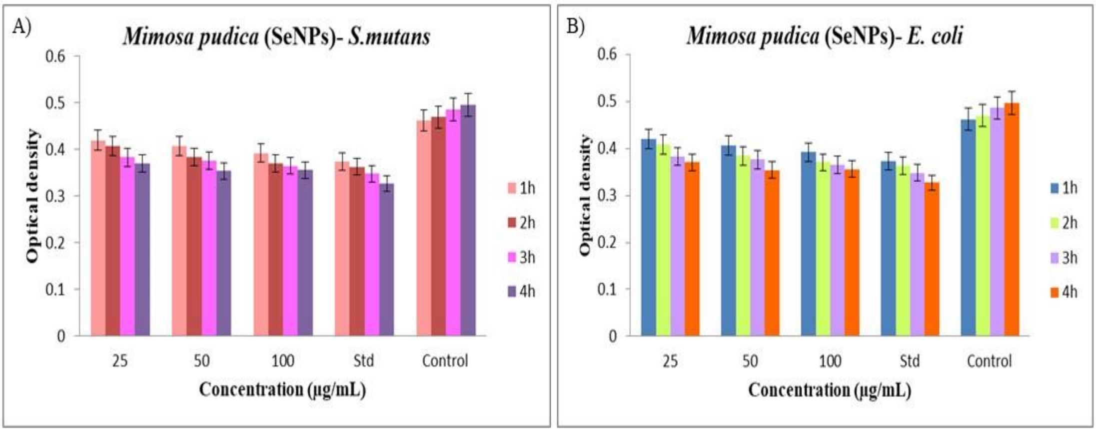


Figure 3: Time-kill assay graph of green-synthesized Selenium nanoparticles against wound pathogens. (A) S. mutans (B) E.coli

## 4. Discussion

The present study evaluated the antibacterial efficacy of selenium nanoparticles (Se NPs) synthesized using Mimosa pudica extract against wound pathogens, specifically S. mutans and E. coli. The results demonstrated significant antibacterial activity, with effects being both concentration and time-dependent, corroborating previous research on the potential of Se NPs as effective antimicrobial agents ((Jayapriya et al., 2022, Johnson et al., 2024, Shanmugam et al., 2023, Kathiravana et al., 2023).

In the time-kill assay, Se NPs at higher concentrations (50 and 100 µg/mL) showed a marked reduction in OD for both S. mutans and E. coli over the 4 hours. These findings align with studies by (Sonkusre et al.2015), which reported that Se NPs exhibited strong bactericidal effects against various bacterial strains, including both Gram-positive and Gram-negative bacteria. The performance of Se NPs in our study, comparable to that of the standard antibiotic, highlights their potential as robust alternatives or complementary antibacterial agents.

The agar well diffusion results further validated the antibacterial efficacy of Se NPs. For S. mutans, the zones of inhibition were significant across all tested concentrations, with the highest concentration (100 µg/mL) yielding an inhibition zone of 19 mm, nearly matching the 20 mm zone produced by the standard antibiotic (Zhang et al. 2021), Previous study reported, similar zones of inhibition for Se NPs synthesized using plant extracts, indicating strong antibacterial activity against Gram-positive bacteria.

For E. coli, the Se NPs demonstrated substantial antibacterial activity, though to a slightly lesser extent than for S. mutans. The inhibition zones for E. coli ranged from 10 mm at 25 µg/mL to 15 mm at 100 µg/mL (Shakibaie et al. 2015). Se NPs had slightly lower antibacterial efficacy against Gram-negative bacteria compared to Gram-positive bacteria. The difference in efficacy might be attributed to the varying cell wall structures and resistance mechanisms between Gram-positive (S. mutans) and Gram-negative (E. coli) bacteria.

The green synthesis of Se NPs using Mimosa pudica extracts not only provides a sustainable and environmentally friendly method but also enhances biocompatibility and reduces the potential toxicity of the nanoparticles. These studies highlighted the advantages of green synthesis techniques in producing biocompatible and effective antimicrobial agents.

Overall, the findings of this study highlight the potential of Mimosa pudica-mediated Se NPs as effective antimicrobial agents, particularly in the treatment of wound infections. The demonstrated antibacterial efficacy, combined with the benefits of green synthesis, suggests that Se NPs could be a valuable addition to antimicrobial treatments, especially in the context of rising antibiotic resistance. Further research should explore the detailed mechanisms of action of these nanoparticles, their long-term effects, and their efficacy in in vivo models to better understand their potential clinical applications.

## 5. Conclusion

The green synthesis approach using M. pudica not only provides an environmentally friendly and sustainable method for producing Se NPs but also enhances their biocompatibility and reduces potential toxicity. These properties, combined with their efficacy in inhibiting bacterial growth, suggest that M. pudica-mediated Se NPs are promising candidates for alternative antimicrobial agents, particularly in the context of wound healing and challenge of antibiotic resistance. Further research is needed to explore the mechanisms of action of these nanoparticles, their long-term effects, and their efficacy in in vivo models to better understand their potential clinical applications. Overall, this study highlights the potential of Se NPs synthesized from M. pudica extract as effective and sustainable antimicrobial agents for the treatment of bacterial infections.

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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.

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