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GREEN SYNTHESIS OF SILVER NANOPARTICLES (AgNPs), USING MORINGA OLEIFERA LEAVES FOR ANTIBACTERIAL ACTIVITY

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ABSTRACT

Purpose: In the present work, silver nanoparticles (AgNPs) were synthesized using bio-reduction method.

Research Design: Silver nitrate was used as metallic precursor and the extract of Moringa oleifera leaves with different concentrations was used as reducing as well capping agent. The extract exhibited strong potential in rapid reduction of silver ions for the synthesis of silver nanoparticles. The synthesized silver nanoparticles were characterized by UV-visible spectroscopy, X-ray diffraction (XRD), and scanning electron microscopy (SEM) techniques.

Results: The absorption SPR peaks appeared in the range of 415 to 439 nm. SEM analysis exhibited that particles were spherical in shape with size distribution range from 10 nm to 25 nm. The synthesized silver nanoparticles were pure crystalline in nature as confirmed by the XRD spectra with average crystallite size 7 nm. In vitro antibacterial activity of the prepared silver nanoparticles colloidal samples as well the extract was studied using different concentrations of AgNPs (C1 = 100 µg/ml, C2 = 50 µg/ml, C3 = 25 µg/ml) and C4 = 20 (µg/ml) by well diffusion method against Gram positive Staphylococcus aureus. The antibacterial performance was assessed by measuring the zone of inhibition (ZOI).

Conclusions: The results suggested that AgNPs prepared by green approach can be considered as an alternative antibacterial agent.

Keywords: Moringa oleifera, AgNPs, antibacterial agent, Staphylococcus aureus and green synthesis.

1. INTRODUCTION

Nanotechnology, the manipulation of materials at the nanometre scale, has emerged as a revolutionary field with diverse applications. At this scale, materials exhibit unique properties vastly different from their bulk counterparts, making them ideal candidates for various technological advancements. One of the most promising areas within nanotechnology is the synthesis and utilization of nanoparticles, particularly metallic nanoparticles like silver nanoparticles, which have garnered significant attention due to their exceptional properties.

Silver nanoparticles, ranging in size from one to one hundred nanometres, possess remarkable electrical, chemical, optical, and catalytic capabilities, making them invaluable in numerous fields. Among these, their potent antibacterial activity stands out, sparking interest in their potential role in combating infectious diseases. The ability of silver nanoparticles to disrupt bacterial cell membranes makes them effective antibacterial agents against a wide range of harmful pathogens.

Green synthesis techniques, employing environmentally friendly materials such as plant extracts, have gained prominence in the fabrication of silver nanoparticles. Utilizing plant extracts like Moringa oleifera, known for their medicinal properties, not only offers a sustainable approach but also circumvents the drawbacks associated with conventional chemical synthesis methods. These plant extracts serve as reducing and capping agents, ensuring the precise and stable formation of silver nanoparticles without the use of hazardous chemicals.

Silver nanoparticles synthesized from natural sources like M. oleifera show promise in this regard, offering a sustainable and effective solution to mitigate foodborne infections.

In this context, the synthesis, optimization, characterization, and evaluation of silver nanoparticles derived from M. oleifera leaf extract present a compelling avenue of research. By exploring the antibacterial activity of these nanoparticles against common foodborne pathogens like Staphylococcus aureus.

In conclusion, nanotechnology particularly through the synthesis of silver nanoparticles using green methods, holds immense potential in addressing pressing challenges such as foodborne illnesses. Leveraging the antibacterial properties of silver nanoparticles derived from natural sources like M. oleifera represents a promising approach to combatting infectious diseases and ensuring food safety in a sustainable manner.

Table: 1

Sample (AgNPs)	PrecursorAgNO3 (1 mM)	Reducing agent (MLE)	Reaction Time	Stirring	Temperature
S1	50ml	0.5	3 hours	Constant	60–80°C
S2	50ml	1	3 hours	Constant	60–80°C
S3	50ml	1.5	3 hours	Constant	60–80°C
S4	50ml	2	3 hours	Constant	60–80°C

3. LITERATURE SURVEY

3.1. Neem Gum-Mediated Synthesis of AgNPs: This study explores the green synthesis of silver nanoparticles (AgNPs) using neem gum extract, highlighting its antibacterial properties against Salmonella enteritidis and Bacillus cereus. While effective, it lacks discussion on environmental concerns and reproducibility.

3.2. AgNPs Synthesis from Moringa Oleifera Seed Cake: Utilizing Moringa oleifera seed cake extract, this research demonstrates successful AgNPs synthesis with significant antimicrobial activity against Escherichia coli. However, it lacks comprehensive biocompatibility assessment and scalability considerations.

3.3. AgNPs Synthesis from Moringa Oleifera Leaves: Green synthesis using Moringa oleifera leaf extract yields AgNPs with potent antimicrobial and cytotoxic properties. Nevertheless, there's a need for further biocompatibility investigation and scalability assessment.

3.4. AgNPs Synthesis from Moringa Oleifera Leaves under Sunlight: Synthesizing AgNPs from Moringa oleifera leaf extracts under sunlight proves effective against various microorganisms, but scalability challenges and synthesis time need addressing.

3.5. AgNPs Synthesis from Moringa Oleifera Leaf targeting Quorum Sensing & Biofilms: This study focuses on inhibiting quorum sensing and biofilm formation in Gram-negative bacteria using AgNPs from Moringa oleifera, yet scalability and environmental concerns remain challenges.

3.6. AgNPs Synthesis from Abelmoschus Esculentus Flower Extract: Using okra flower extract, this study successfully synthesizes AgNPs with promising cytotoxic and antimicrobial properties. However, mechanistic insights and scalability need further exploration.

3.7. Multivariate Optimization for AgNPs Synthesis using Moringa Oleifera Extracts: Employing multivariate optimization, AgNPs are synthesized from different parts of Moringa oleifera, showing varied properties. Yet, the reliance on water extracts and interactions between factors require more investigation.

3.8. Vanadium Nanoparticles Synthesis using Moringa Oleifera Leaf Extract: Green synthesis of vanadium nanoparticles using Moringa oleifera leaf extract exhibits antimicrobial efficacy against bacteria, but it lacks optimization for antifungal activity and synthesis scalability.

4. MATERIALS AND METHODS

4.1. M.oleifera Leaf Extract Preparation

Objective: To obtain M. oleifera leaf
Extract, which will act as a reducing agent in the synthesis of AgNPs.

Process: Leaves are powdered, boiled in deionized water, filtered, and stored. The boiling helps in extracting the phytochemicals from the leaf powder into the water.

4.2. Precursor Preparation

Objective: To prepare a silver nitrate solution, which provides the Ag+ ions needed for nanoparticle synthesis.

Process: A 1 mM solution of AgNO3 is prepared and stored, ensuring that the concentration of silver ions is controlled for a consistent reaction.

4.3. Biosynthesis of Silver Nanoparticles

Objective: To synthesize AgNPs using a green chemistry approach, utilizing the natural reducing properties of M. oleifera extract.

Process: The leaf extract is added to the AgNO3 solution and heated. The change in color is a visual indication of nanoparticle formation.

4.4. Substrate Cleaning and Sample Preparation

Objective: To prepare clean glass slides for nanoparticle deposition, ensuring that the surface is free from organic or inorganic residues.

Process: Slides are washed, treated with solvents, and dried. Clean slides are crucial for subsequent analyses and applications to avoid contamination.

4.5. Characterization of Silver Nanoparticles

UV-VIS Absorption Spectroscopy: Used to confirm the presence and quality of AgNPs by measuring their absorption in the UV-Vis spectrum, which is characteristic of their size and shape.

Scanning Electron Microscopy (SEM): Offers a detailed view of the nanoparticles' morphology, including shape and size.

X-Ray Diffraction Analysis (XRD): Provides information on the crystalline structure of the nanoparticles, confirming their composition and crystallinity.

4.6. Antibacterial Analysis

Objective: To evaluate the antibacterial efficacy of the synthesized AgNPs against Staphylococcus aureus.

Process: The growth of Staphylococcus aureus in the presence of MLE-AgNPs is monitored over time, comparing it to a control (without AgNPs) to assess the nanoparticles antibacterial activity.

4.7. Growth of Bacteria with MLE-AgNPs

Objective: To observe the growth pattern of Staphylococcus aureus. when exposed to MLE-AgNPs under aerobic conditions.

Method: Staphylococcus aureus is cultured in nutrient broth supplemented with MLE-AgNPs and incubated, with optical density (OD) measurements.

4.8. Well Diffusion Method

Objective: To visually assess the antibacterial activity of MLE-AgNPs and the leaf extract itself against Staphylococcus aureus.

Method: After sterilization, agar plates are prepared and inoculated with the bacterial culture. Wells are created in the agar, into which samples of MLE-AgNPs at various concentrations are introduced.

4.9. Determination of Minimum Inhibitory Concentration (MIC)

Objective: To find the lowest concentration of MLE-AgNPs that effectively inhibits the growth of Staphylococcus aureus, providing a clear measure of antibacterial potency.

Method: Staphylococcus aureus is cultured in the presence of varying concentrations of MLE-AgNPs. After incubation, the cultures are examined for growth by measuring OD at 600 nm.

Figure: 1

5. Interpretation and Importance

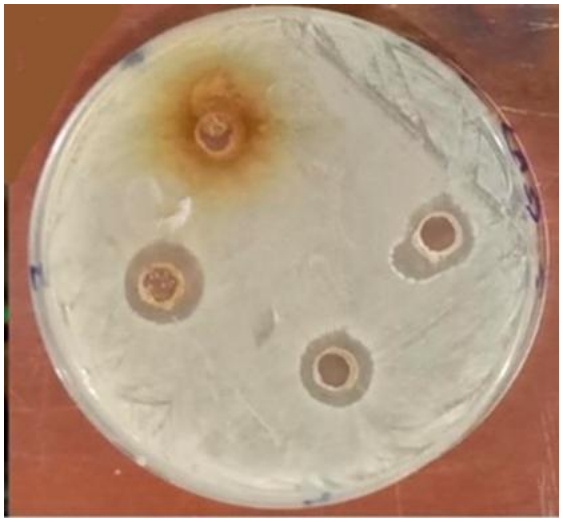
5.1. Growth Observation: By comparing growth curves of treated and untreated S. aureus cultures, the impact of AgNPs on bacterial proliferation can be quantified. A reduction in growth rate or a lag in reaching the stationary phase indicates antibacterial activity.

5.2. Well Diffusion Method: Offers a direct and visual method of assessing antibacterial activity. The presence of clear zones around wells (zones of inhibition) directly correlates to the effectiveness of the AgNPs against S.aureus.

5.3. MIC Determination: MIC is a critical value in antibacterial studies, providing a quantitative measure of the antimicrobial strength of a substance. It informs on the minimal concentration needed to inhibit bacterial growth, which is crucial for potential therapeutic applications.

6. RESULT

Using the agar well diffusion method, the antibacterial activity of produced silver nanoparticles against Staphylococcus aureus was examined. Three distinct nanoparticle concentrations (C1: 100 µg/ml, C2: 50 µg/ml, and C3: 25 µg/ml) were examined in conjunction with pure water and leaf extract as negative controls. The findings demonstrated the silver nanoparticles' strong antibacterial activity, with bigger zones of inhibition correlating to higher concentrations. In particular, no inhibition zones were seen for the negative controls, but the zones of inhibition for C1, C2, and C3 were measured to be 19 mm, 17 mm, and 15 mm, respectively. This suggests that the antibacterial activity of silver nanoparticles and their concentration are related in a dose-dependent manner. These results highlight the manufactured silver nanoparticles' potential as powerful antibacterial agents against Staphylococcus aureus.



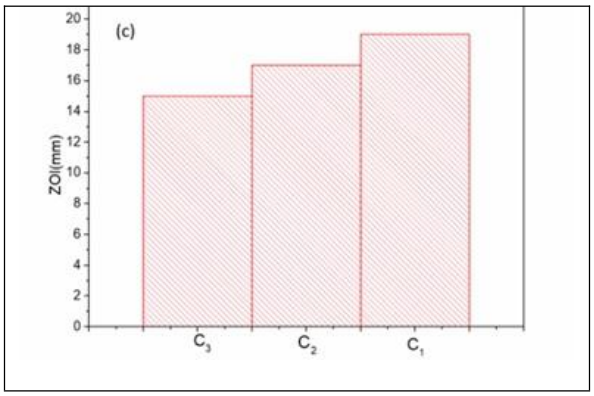


Figure: 3

7. CONCLUSION

The utilization of a green synthesis approach to produce silver nanoparticles (AgNPs) using Moringa oleifera leaf extract has yielded promising results in terms of antibacterial activity. Characterization techniques such as UV-visible spectroscopy, X-ray diffraction (XRD), and scanning electron microscopy (SEM) confirmed the successful synthesis of AgNPs with desirable properties, including spherical morphology and pure crystallinity. The in vitro antibacterial evaluation against S. aureus demonstrated the effectiveness of the synthesized AgNPs, with increasing concentrations correlating with larger zones of inhibition. Finally, to combating bacterial infections, suggesting their possible application as antibacterial agents in various medical applications.

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