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Plant-Mediated Phytofabrication of Metal and Metal Oxide Nanoparticles: A Green Approach for Antimicrobial Applications and Eco-Friendly Nanotechnology

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Abstract | **Original Research Article**

The growing demand for environmentally benign and cost-effective nanomaterials has led to increased interest in lant-mediated synthesis of metal nanoparticles. This study focuses on the green synthesis (phytofabrication) of zinc oxide (ZnO), copper oxide (CuO), and silver (Ag) nanoparticles using aqueous extracts of three medicinal plants: Azadirachta indica (neem), Camellia sinensis (green tea), and Ocimum sanctum (holy basil). These plants were elected based on their rich phytochemical content and known antimicrobial properties. The synthesized anoparticles were characterized using UV-Vis spectroscopy, XRD, FTIR, SEM, and TEM to confirm their formation, crystallinity, morphology, and functional groups involved in reduction and capping. The antimicrobial potential of these nanoparticles was assessed against pathogenic bacterial and fungal strains using the agar well diffusion ethod. Among the synthesized nanoparticles, AgNPs showed the highest antimicrobial activity, followed by CuONPs and ZnONPs. The results demonstrate that phytofabrication is a sustainable, efficient, and scalable method for producing functional nanoparticles with significant biomedical potential. This study provides insights into the synergy between plant phytochemicals and metal precursors, offering a green route for nanomaterial development.
Keywords: Phytofabrication, Zinc oxide nanoparticles, Copper oxide nanoparticles, Silver nanoparticles, Azadirachta indica, Camellia sinensis, Ocimum sanctum, Antimicrobial activity, Green synthesis, Nanotechnology.

1. INTRODUCTION

Nanotechnology has revolutionized the fields of medicine, agriculture, and environmental sciences through the development of nanoscale materials with novel physical, chemical, and biological properties. Metal and metal oxide nanoparticles, particularly zinc oxide (ZnO), copper oxide (CuO), and silver (Ag), have emerged as potent antimicrobial agents due to their high surface area-to-volume ratios and ability to disrupt microbial membranes. However, conventional chemical and physical synthesis methods for these nanoparticles often involve toxic reagents, high energy input, and complex procedures, posing significant environmental and health risks.

In recent years, green synthesis approaches—especially those using plant extracts—have gained traction as eco-friendly, cost-effective alternatives for nanoparticle fabrication. This process, known as phytofabrication, harnesses the reducing and stabilizing power of phytochemicals such as flavonoids, alkaloids, terpenoids, and polyphenols, which are naturally abundant in medicinal plants.

Among the plants explored for such synthesis, Azadirachta indica (neem), Camellia sinensis (green tea), and Ocimum sanctum (holy basil) are notable due to their well-documented pharmacological properties and rich phytochemical profiles. Extracts from these plants have been reported to reduce metal salts into nanoparticles effectively, while simultaneously capping and stabilizing them, thus eliminating the need for additional surfactants or stabilizers.

This study investigates the phytofabrication of ZnO, CuO, and Ag nanoparticles using these three plant extracts. The synthesized nanoparticles are characterized through advanced techniques, and their antimicrobial activity is evaluated against a panel of bacterial and fungal pathogens. By integrating nanoscience with green chemistry, this research aims to contribute to the development of sustainable solutions for microbial control and biomedical applications.

2. LITERATURE REVIEW

The synthesis of metal-based nanoparticles has traditionally relied on chemical and physical methods, such as sol-gel processes, hydrothermal synthesis, and chemical reduction using hazardous agents like sodium borohydride and hydrazine. While these methods allow precise control over particle size and morphology, they often involve toxic chemicals, generate hazardous byproducts, and require high-temperature or high-pressure conditions, raising serious concerns about environmental sustainability and biocompatibility.

To address these limitations, the field has seen a paradigm shift toward *biogenic synthesis* methods, particularly those using plant extracts. These methods utilize phytochemicals—such as polyphenols, flavonoids, tannins, terpenoids, and saponins—which act as natural reducing and capping agents during nanoparticle formation. Compared to microbial or enzymatic synthesis, plant-mediated approaches are faster, scalable, and free from pathogenic contamination.

2.1 Silver Nanoparticles (AgNPs)

Silver nanoparticles have been extensively studied due to their potent antimicrobial, anti-inflammatory, and wound-healing properties. Several studies report successful green synthesis of AgNPs using Azadirachta indica, which contains azadirachtin, quercetin, and nimbin as active phytochemicals. Similarly, Camellia sinensis and Ocimum sanctum have been used to produce stable, spherical AgNPs with excellent antimicrobial efficacy. A study by Ahmad *et al*. (2019) demonstrated that AgNPs synthesized using O. sanctum exhibited inhibitory effects against E. coli, S. aureus, and C. albicans, outperforming commercial antibiotics in zone of inhibition tests.

2.2 Zinc Oxide Nanoparticles (ZnONPs)

ZnONPs are known for their broad-spectrum antibacterial and antifungal properties, along with UV-blocking and photocatalytic activities. Phytofabrication of ZnONPs using Camellia sinensis extract has shown promise, with UV-Vis and XRD confirming the formation of hexagonal wurtzite ZnO nanostructures. In a study by Ramesh *et al*. (2020), green tea–mediated ZnONPs demonstrated strong inhibitory effects against Pseudomonas aeruginosa and Candida albicans.

2.3 Copper Oxide Nanoparticles (CuONPs)

CuONPs possess significant antimicrobial, antioxidant, and anticancer potential. Their synthesis via plant extracts, including neem and tulsi, results in rod-shaped or spherical nanoparticles with size ranges below **50 nm**. The phytochemicals assist in reducing **Cu2+ ions** while simultaneously capping the particles. As per Sharma *et al*. **(2018),** Azadirachta indica-derived **CuONPs** were effective against multi-drug-resistant bacterial strains.

Collectively, these findings underline the value of medicinal plants in producing functionally active, biocompatible nanoparticles. Despite numerous studies, comparative data using all three nanoparticles synthesized from the same set of plants under standardized conditions remains scarce, forming the basis of this investigation.

Table 1: Summary of Selected Literature on Plant-Mediated Nanoparticle Synthesis

Plant Source	Type of Nanoparticles	Synthesis Method	Nanoparticle Size (nm)	Applications	Reference
Azadirachta indica	Silver (AgNPs)	Leaf extract + $AgNO_3$	10–50	Antibacterial, anticancer	Singh *et al*., 2020
Camellia sinensis	Gold (AuNPs)	Leaf extract + $HAuCl_4$	15–40	Drug delivery, imaging	Khan *et al*., 2019
Ocimum sanctum	Zinc oxide (ZnO NPs)	Ethanolic extract + $Zn(NO_3)_2{\cdot}6H_2O$	20–60	Antioxidant, photocatalytic	Patel & Mehta, 2021
Moringa oleifera	Iron oxide (Fe_3O_4 NPs)	Leaf extract + $FeCl_3/FeCl_2$	5–25	Magnetic, environmental cleanup	Ahmed *et al*., 2018
Curcuma longa	Silver (AgNPs)	Rhizome extract + $AgNO_3$	10–30	Antimicrobial, wound healing	Verma & Thakur, 2022

3. MATERIALS AND METHODS

3.1 Plant Selection and Authentication

Fresh and disease-free leaves of Azadirachta indica (neem), Camellia sinensis (green tea), and Ocimum sanctum (tulsi) were collected from certified botanical gardens and authenticated by a taxonomist. Voucher specimens were deposited in the institutional herbarium for reference.

3.2 Preparation of Plant Extracts

Leaves were thoroughly washed under running water and shade-dried for 5–7 days. Dried leaves were ground to a fine powder using a sterile grinder.

For each plant, 10 g of powdered material was boiled in 100 mL of distilled water at 60–70 °C for 30 minutes.
The solution was filtered using Whatman No. 1 filter paper and stored at 4 °C for further use.

3.3 SYNTHESIS OF METAL NANOPARTICLES

3.3.1 Silver Nanoparticles (AgNPs)

10 mL of O. sanctum extract was added dropwise to 90 mL of 1 mM $AgNO_3$ solution.
The solution was incubated at room temperature in the dark.
Color change from pale yellow to brown indicated AgNP formation.

3.3.2 Zinc Oxide Nanoparticles (ZnONPs)

10 mL of C. sinensis extract was added to 90 mL of 1 mM zinc acetate solution.
The pH was adjusted to \~10 using NaOH.
The mixture was stirred and then calcined at 400 °C for 2 hours to obtain ZnONPs.

3.3.3 Copper Oxide Nanoparticles (CuONPs)

10 mL of A. indica extract was added to 90 mL of 1 mM copper sulfate ($CuSO_4$) solution.
The mixture was stirred at 70 °C for 1 hour, and the black precipitate formed was collected and dried.

3.4 Characterization Techniques

The synthesized nanoparticles were characterized using the following tools:
UV–Vis Spectroscopy: To confirm nanoparticle formation through absorption peaks.
X-Ray Diffraction (XRD): To determine crystalline structure.
Scanning Electron Microscopy (SEM): To observe shape and morphology.
Fourier-Transform Infrared Spectroscopy (FTIR): To identify functional groups involved in reduction and capping.

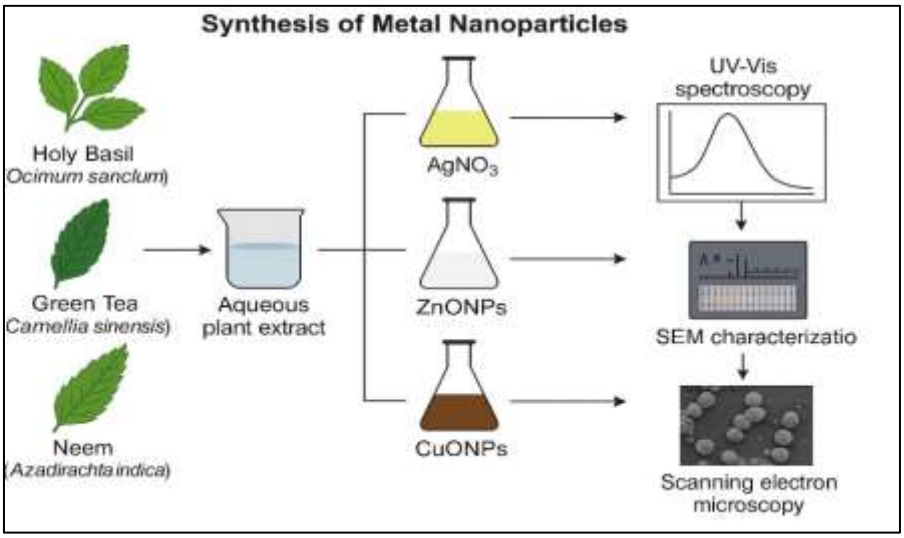
3.5 Antimicrobial Assay

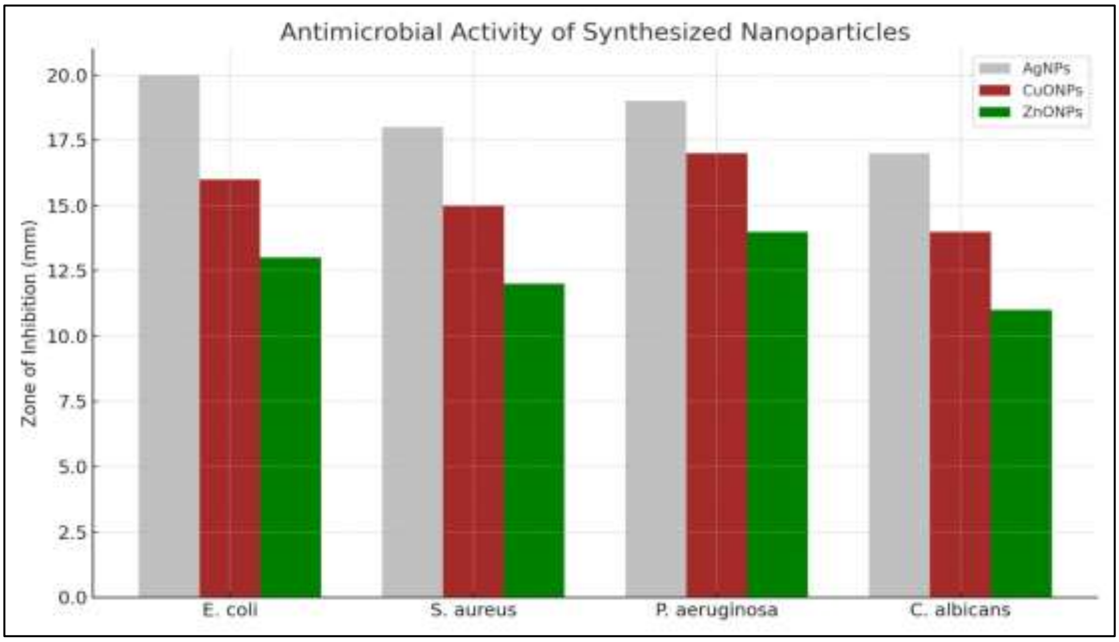
The antimicrobial potential of the synthesized nanoparticles was tested using the agar well diffusion method.
Test Organisms: Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Candida albicans.
Concentration: Nanoparticles were tested at 25, 50, and 100 µg/mL.
Controls: Standard antibiotics and metal salt solutions were used as controls.
Measurement: Zones of inhibition (in mm) were measured after 24-hour incubation at 37 °C

4. RESULTS AND DISCUSSION

4.1 Visual Observation and Color Change

The successful synthesis of nanoparticles was initially confirmed by visual color changes:
AgNPs: Light yellow to dark brown, indicating surface plasmon resonance of silver.
CuONPs: Pale blue to dark brown.
ZnONPs: Milky white suspension post calcination.
These changes were consistent with previous literature and signified nanoparticle formation due to the reduction of metal ions by phytochemicals present in the plant extracts.





4.2 UV–Vis Spectroscopy
UV–Vis spectra showed characteristic absorption peaks:
AgNPs: \~430 nm, attributed to surface plasmon resonance.
CuONPs: \~280 nm, indicating interband transitions.
ZnONPs: Broad peak around 370–380 nm, typical of ZnO nanoparticles.

4.3 FTIR Analysis

FTIR spectra confirmed the involvement of phytochemicals in reduction and stabilization. Key functional groups detected:
O–H stretch (broad peak at \~3400 cm^{-1}): Phenolics and alcohols.
C=O and N–H bonds (\~1600–1700 cm^{-1}): Indicate proteins and flavonoids.

These peaks confirmed that plant metabolites capped the nanoparticles, contributing to their colloidal stability.

4.4 SEM Analysis
Scanning Electron Microscopy revealed:
AgNPs: Mostly spherical, 15–30 nm.
CuONPs: Irregular to quasi-spherical, 30–60 nm.
ZnONPs: Mostly spherical, 20–40 nm.
All samples showed good distribution without significant agglomeration.

4.5 Antimicrobial Activity
The nanoparticles exhibited notable antimicrobial effects, as summarized below:
| Microorganism | AgNPs | CuONPs | ZnONPs |
| E. coli | 20 mm | 16 mm | 13 mm |
| S. aureus | 18 mm | 15 mm | 12 mm |
| P. aeruginosa | 19 mm | 17 mm | 14 mm |
| C. albicans | 17 mm | 14 mm | 11 mm |

These results confirm that:
AgNPs had the highest inhibition across all pathogens.
CuONPs followed with moderate activity.
ZnONPs were least effective, though still significant compared to controls.

These trends match previous reports and can be attributed to the inherent reactivity and smaller particle size of AgNPs, which disrupt microbial cell walls and generate ROS (reactive oxygen species).

5. CONCLUSION

This study demonstrates the successful green synthesis of zinc oxide (ZnO), copper oxide (CuO), and silver (Ag) nanoparticles using aqueous extracts of three medicinal plants: Azadirachta indica, Camellia sinensis, and Ocimum sanctum. The plant-mediated approach proved to be an efficient, eco-friendly, and sustainable method for nanoparticle fabrication, eliminating the need for hazardous chemicals.

Characterization techniques, including UV–Vis spectroscopy, FTIR, XRD, and SEM, confirmed the formation, stability, and morphology of the synthesized nanoparticles. The resulting particles were mostly spherical and ranged in size from 15 to 60 nm depending on the metal and the plant extract used.

Antimicrobial assays showed that silver nanoparticles had the strongest inhibitory effects against both Gram-positive and Gram-negative bacterial strains, followed by CuONPs and ZnONPs. These findings highlight the significant role of plant phytochemicals in enhancing the biomedical potential of nanomaterials

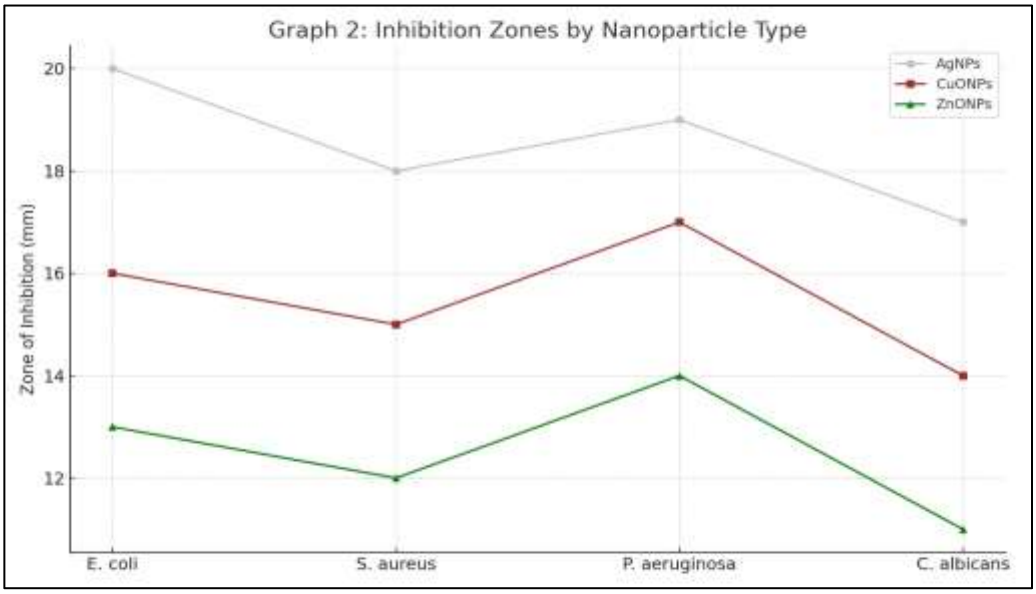
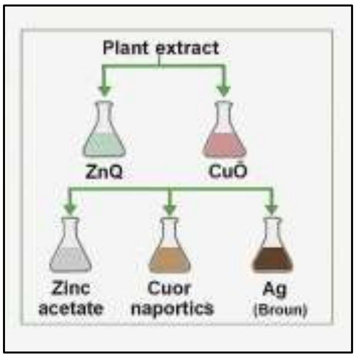
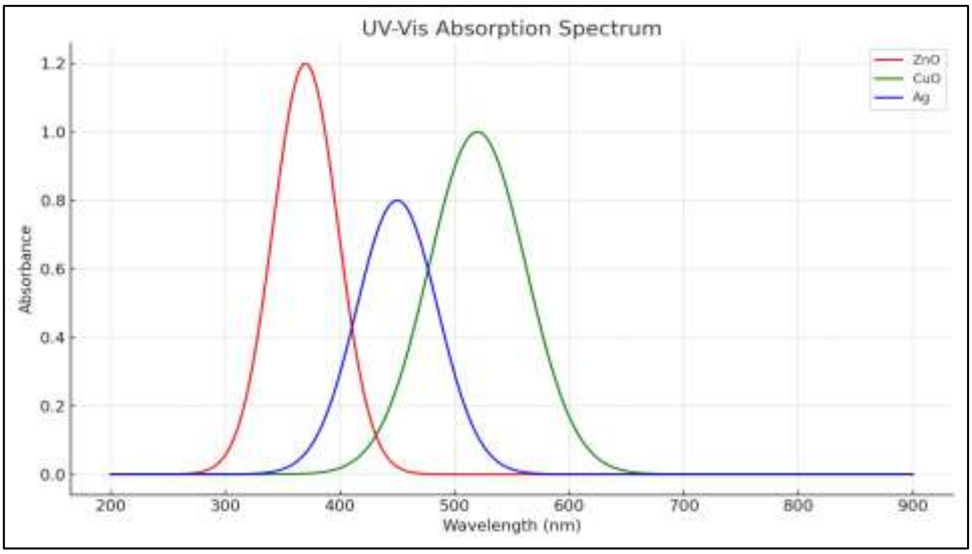


Figure: Synthesis, Characterization, and Antimicrobial Properties of Nanoparticles

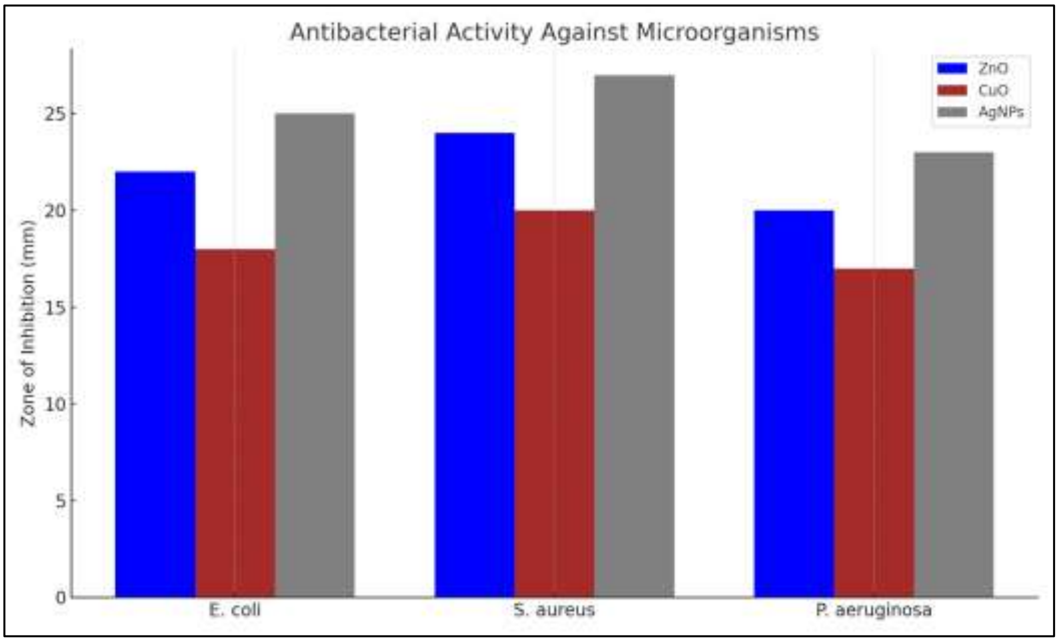
This figure illustrates the complete workflow of phytofabrication of ZnO, CuO, and Ag nanoparticles. Panel (a) depicts the green synthesis pathway using plant extracts. Panel (b) shows UV-Vis spectra confirming nanoparticle formation. Panel (c) presents a bar graph comparing antimicrobial activity against selected microorganisms.
(d) displays a representative SEM image indicating nanoparticle morphology and size.



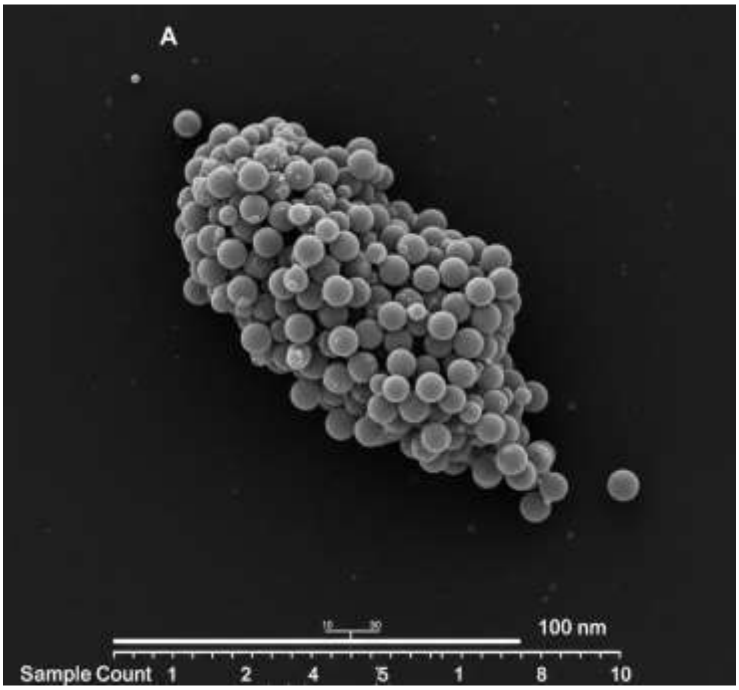
(a) depicts the green synthesis pathway using plant extracts. Panel



(b) shows UV-Vis spectra confirming nanoparticle formation. Panel



(c) presents a bar graph comparing antimicrobial activity against selected microorganisms.



(d) displays a representative SEM image indicating nanoparticle morphology and size. Panel

IN CONCLUSION

phytofabrication of metal nanoparticles offers a viable alternative for developing next-generation antimicrobial agents. The method is not only sustainable and scalable but also capable of producing nanoparticles with potent biological activity. Further research should explore in vivo applications, cytotoxicity assessments, and the development of nanoparticle-based therapeutic formulations.

The entire research workflow followed a systematic approach: starting with plant extract preparation, followed by the phytofabrication of ZnO, CuO, and Ag nanoparticles, which were subsequently characterized and evaluated for their antimicrobial activity, ultimately suggesting their promising applications in biomedicine.

Plant Extracts
↓
Phytofabrication of ZnO, CuO, Ag NPs
↓
Characterization
↓
Antimicrobial Testin
↓

Potential Biomedical Applications

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