

Green Synthesis and Characterization of Silver Nanoparticles Mediated by Carica papaya Leaf Extract and Their Enhanced Antibacterial Activity

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ABSTRACT

Background: The emergence of multidrug-resistant microorganisms has necessitated the development of novel antimicrobial agents. Green synthesis of silver nanoparticles using medicinal plant extracts offers an eco-friendly and cost-effective alternative to conventional nanoparticle synthesis methods. Carica papaya leaves are rich in bioactive phytochemicals that can act as reducing and stabilizing agents during nanoparticle synthesis.

Aim: The present study aimed to synthesize silver nanoparticles using Carica papaya leaf extract, characterize the synthesized nanoparticles, and evaluate their antimicrobial efficacy against selected bacterial and fungal pathogens.

Materials and Methods: Aqueous leaf extract of Carica papaya was prepared and utilized for the green synthesis of silver nanoparticles. The synthesized nanoparticles were characterized using UV–Visible spectroscopy, Fourier Transform Infrared Spectroscopy (FTIR), and Scanning Electron Microscopy (SEM). Antimicrobial activity was assessed using the agar well diffusion method against Staphylococcus aureus, Enterococcus faecalis, Escherichia coli, and Candida albicans at concentrations of 25 and 100 µg/mL.

Results: UV–Visible spectroscopy confirmed nanoparticle formation through the appearance of a characteristic absorption band around 260–280 nm. FTIR analysis revealed the presence of hydroxyl, amine, and phenolic functional groups involved in nanoparticle reduction and stabilization. SEM images demonstrated irregularly shaped and aggregated nanoparticles. Antimicrobial studies revealed significant inhibitory activity against Staphylococcus aureus and Candida albicans, with inhibition zones of 23 mm and 28 mm respectively at 100 µg/mL concentration.

Conclusion: Carica papaya leaf extract-mediated silver nanoparticles were successfully synthesized and exhibited promising antimicrobial activity. The synthesized nanoparticles may serve as potential candidates for biomedical and pharmaceutical applications.

Keywords: Innovation, Sustainability, Carica papaya, Silver nanoparticles, Green synthesis, Antimicrobial activity, SEM, FTIR, UV–Vis spectroscopy.

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1. INTRODUCTION

Antimicrobial resistance has become one of the major global health concerns, leading to increased morbidity, mortality, and healthcare expenditure. The rapid emergence of resistant bacterial strains has prompted the search for alternative antimicrobial agents with enhanced efficacy and reduced toxicity(1).

Nanotechnology has emerged as a promising field in biomedical research due to the unique physicochemical properties exhibited by nanoparticles(2). Among various metallic nanoparticles, silver nanoparticles (AgNPs) have attracted significant attention because of their broad-spectrum antimicrobial, anti-inflammatory, antioxidant, and wound-healing properties(3). Their small size and high surface area facilitate enhanced interaction with microbial cells, resulting in effective microbial inhibition(4).

Green synthesis approaches have gained popularity because they eliminate the use of hazardous chemicals and provide environmentally friendly alternatives to conventional synthesis methods(5). Plant extracts contain a variety of phytochemicals such as flavonoids, alkaloids, tannins, phenolics, and proteins capable of reducing metal ions and stabilizing nanoparticles.

Carica papaya is a medicinal plant widely recognized for its therapeutic properties(6), including antimicrobial, antioxidant, anti-inflammatory, and immunomodulatory activities(7). The leaves contain numerous bioactive compounds that can facilitate nanoparticle synthesis. Therefore, the present study was undertaken to formulate and characterize silver nanoparticles synthesized using *Carica papaya* leaf extract and evaluate their antimicrobial activity against selected microbial pathogens.

2. MATERIALS AND METHODS

Collection and Authentication of Plant Material

Fresh leaves of *Carica papaya* were collected from local sources in Chennai, Tamil Nadu, India. The collected leaves were thoroughly washed under running tap water followed by distilled water to remove dust and other contaminants. The leaves were shade-dried at room temperature for 7–10 days and subsequently pulverized into a fine powder using a laboratory grinder. The powdered material was stored in airtight containers until further use.

Preparation of Aqueous Leaf Extract

Twenty grams (20 g) of dried *Carica papaya* leaf powder was mixed with 100 mL of distilled water in a conical flask. The mixture was heated at 60–70°C for 30 min under continuous stirring to facilitate the extraction of bioactive phytoconstituents. After cooling to room temperature, the extract was filtered through Whatman No.1 filter paper using gravity filtration. The filtrate was collected and stored at 4°C for subsequent nanoparticle synthesis.

Green Synthesis of Silver Nanoparticles

Silver nanoparticles were synthesized using the aqueous leaf extract as a reducing and stabilizing agent. Briefly, 10 mL of freshly prepared aqueous *Carica papaya* leaf extract was added dropwise to 90 mL of 1 mM aqueous silver nitrate (AgNO_3) solution under continuous magnetic stirring.

The reaction mixture was maintained at room temperature and stirred continuously until a visible color change from pale yellow to dark brown was observed, indicating the reduction of Ag^+ ions and the formation of silver nanoparticles. The reaction was allowed to proceed for 24 h to ensure complete reduction of silver ions.

The synthesized nanoparticles were separated by centrifugation at 3000 rpm for 10 min. The pellet obtained was washed three times with distilled water to remove unreacted plant metabolites and residual silver ions. The purified nanoparticles were dried at room temperature and stored in sterile containers for characterization and antimicrobial evaluation.

Characterization of Silver Nanoparticles

UV–Visible Spectroscopy

The optical properties of the synthesized nanoparticles were analyzed using a UV–Visible spectrophotometer. The absorption spectrum was recorded within the wavelength range of 200–800 nm. Distilled water served as the blank. The formation of silver nanoparticles was confirmed by the appearance of characteristic absorption bands due to surface plasmon resonance.

Fourier Transform Infrared Spectroscopy (FTIR)

FTIR analysis was performed to identify the functional groups present in the synthesized nanoparticles and to determine the phytochemical constituents responsible for the reduction and stabilization of silver ions. The dried nanoparticle sample

was analyzed over the spectral range of 4000–400 cm^{-1} .

Scanning Electron Microscopy (SEM)

The surface morphology and structural characteristics of the synthesized silver nanoparticles were examined using Scanning Electron Microscopy (SEM). The dried nanoparticle powder was mounted on aluminum stubs using conductive carbon tape and sputter-coated with a thin layer of gold prior to imaging. Micrographs were obtained at different magnifications to evaluate particle morphology and aggregation patterns.

Antimicrobial Activity

Microbial Strains

The antimicrobial activity of synthesized silver nanoparticles was evaluated against the following microorganisms: *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli*, *Candida albicans*. The microbial cultures were maintained on Mueller–Hinton Agar (MHA) and subcultured prior to experimentation.

Agar Well Diffusion Assay

Antimicrobial activity was assessed using the agar well diffusion method. Fresh microbial suspensions were uniformly spread onto sterile Mueller–Hinton Agar plates using sterile cotton swabs.

Sterile wells/discs were prepared on the inoculated agar plates and loaded with two concentrations of synthesized silver nanoparticles (25 $\mu\text{g/mL}$ and 100 $\mu\text{g/mL}$). Standard antimicrobial discs served as positive controls, while distilled water served as the negative control.

The plates were incubated at 37°C for 24 h. Following incubation, the antimicrobial efficacy was determined by measuring the diameter of the zone of inhibition (ZOI) around each well/disc using a digital Vernier caliper. All experiments were performed in triplicate and the results were expressed as mean inhibition zone diameter (mm).

3. RESULTS

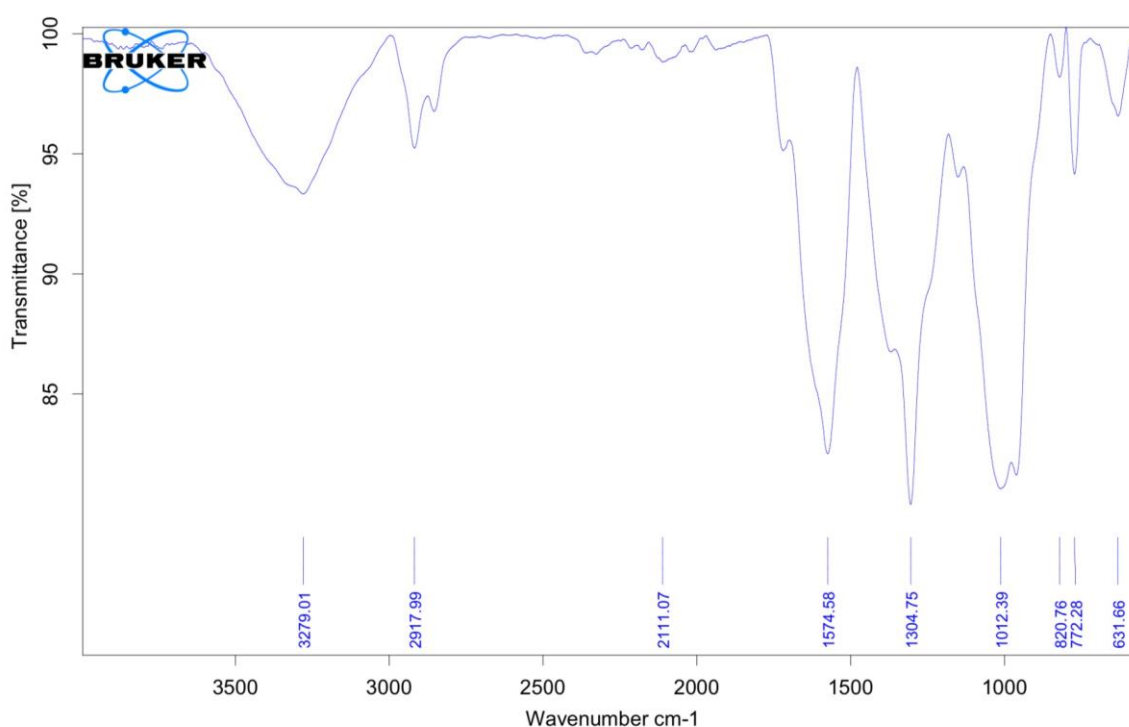


Figure 1:

FTIR analysis revealed characteristic absorption bands at 3279.01, 2917.99, 2111.07, 1574.58, 1304.75, 1012.39, 820.76, 772.28, and 631.66 cm^{-1} , corresponding to hydroxyl, aliphatic, amine, aromatic, and polysaccharide functional groups. These functional groups originate from phytochemicals present in *Carica papaya* leaves and are responsible for the reduction and stabilization of silver nanoparticles.

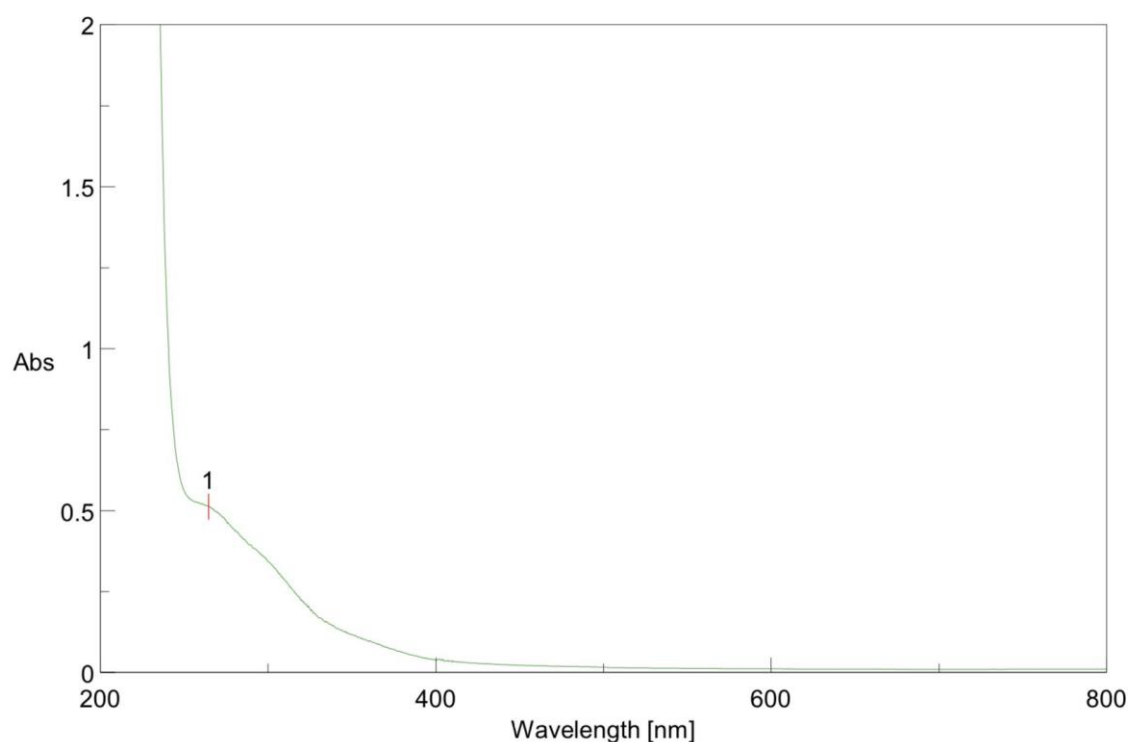


Figure 2: UV–Visible spectroscopy exhibited a characteristic absorption peak around 275–280 nm, confirming the successful biosynthesis of silver nanoparticles through the reduction of silver ions by the plant extract.

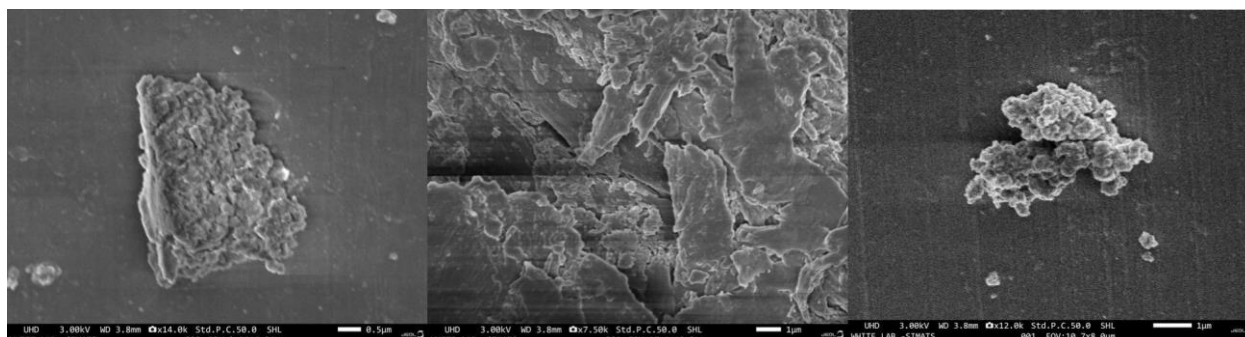


Figure 3: Scanning electron microscopic analysis revealed the formation of silver nanoparticles with irregular, aggregated, and clustered morphology. The synthesized nanoparticles exhibited rough surface characteristics and heterogeneous particle distribution. The agglomerated nature of the nanoparticles is likely due to the presence of phytochemical constituents from *Carica papaya* leaf extract that act as natural reducing and stabilizing agents. The SEM findings confirmed the successful biosynthesis of silver nanoparticles and demonstrated structural features favorable for antimicrobial applications.

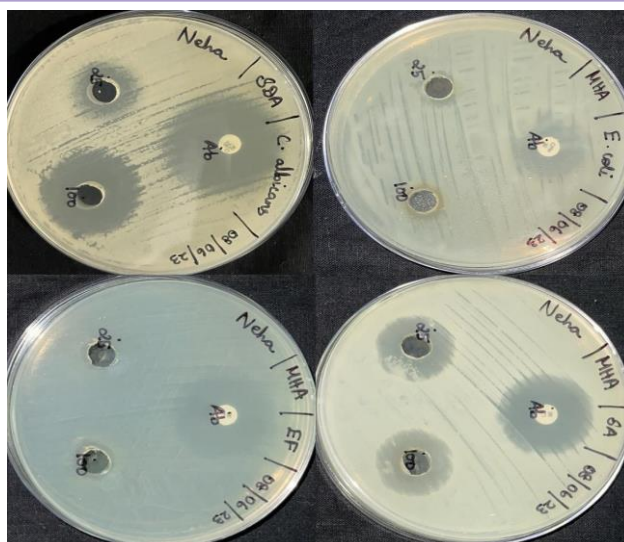


Figure 4:

Antimicrobial activity of *Carica papaya* leaf extract-mediated silver nanoparticles evaluated by agar well diffusion assay against *Candida albicans*, *Escherichia coli*, and *Staphylococcus aureus*. Wells containing AgNPs at concentrations of 25 µg/mL and 100 µg/mL exhibited clear zones of inhibition around the inoculated microorganisms. The antimicrobial activity increased with nanoparticle concentration, demonstrating the potential of biosynthesized silver nanoparticles as effective antimicrobial agents.

4. DISCUSSION

The present study demonstrated the successful green synthesis of silver nanoparticles (AgNPs) using *Carica papaya* leaf extract and confirmed their antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans*. The biosynthesized nanoparticles exhibited concentration-dependent antimicrobial activity, with greater inhibition observed at 100 µg/mL compared to 25 µg/mL. Among the tested microorganisms, *Candida albicans* showed the highest susceptibility, followed by *S. aureus*, while *E. coli* exhibited comparatively lower sensitivity.

The UV–Visible spectroscopic analysis confirmed nanoparticle formation through the appearance of a characteristic absorption peak associated with the surface plasmon resonance (SPR) of silver nanoparticles. Similar observations were reported by Roy et al. (2022)(8), who synthesized plant-mediated AgNPs and observed SPR peaks characteristic of nanoparticle formation. The FTIR analysis revealed the presence of hydroxyl, amine, aromatic, and carbonyl functional groups, indicating the involvement of phytochemicals in the reduction and stabilization of AgNPs. These findings are consistent with those reported by Ghosvandan et al. (2020)(9), who demonstrated that phenolic compounds and flavonoids act as reducing and capping agents during green nanoparticle synthesis.

SEM analysis revealed irregular and aggregated nanoparticle structures with rough surface morphology. Similar morphologies have been reported for biosynthesized silver nanoparticles produced using medicinal plant extracts. The aggregation observed may result from interactions between nanoparticle surfaces and phytochemical constituents present in *Carica papaya* leaves(4). According to Pal et al. (2024)(4), plant-derived biomolecules not only facilitate nanoparticle synthesis but also improve nanoparticle stability by forming protective coatings around the particles.

The antimicrobial results obtained in the present study corroborate previous reports on the broad-spectrum antimicrobial efficacy of green-synthesized silver nanoparticles. The strong inhibitory effect observed against *C. albicans* agrees with the findings of Ting et al. (2021),(10) who reported enhanced antifungal activity of biosynthesized AgNPs against pathogenic *Candida* species. Fungal cells possess negatively charged membrane components that readily interact with silver nanoparticles, resulting in membrane disruption and leakage of intracellular contents.

The moderate activity observed against *S. aureus* is consistent with studies demonstrating that Gram-positive bacteria are highly susceptible to AgNP-mediated cell wall damage(8). The thick peptidoglycan layer of Gram-positive bacteria contains numerous binding sites for silver ions and nanoparticles, facilitating antimicrobial action. Similar findings were reported by Swamy et al. (2015)(11), who observed significant inhibition of *S. aureus* following treatment with plant-mediated AgNPs.

In contrast, *E. coli* exhibited comparatively lower susceptibility to the synthesized nanoparticles. This reduced sensitivity

may be attributed to the complex outer membrane of Gram-negative bacteria, which serves as an additional barrier limiting nanoparticle penetration. A comparable trend was reported by Ahmed et al. (2024), (12) who observed lower antibacterial activity against Gram-negative organisms than Gram-positive bacteria due to differences in cell wall architecture.

The antimicrobial mechanism of AgNPs is multifactorial. (13) Silver nanoparticles can attach to microbial cell surfaces, alter membrane permeability, generate reactive oxygen species (ROS), induce oxidative stress, disrupt electron transport chains, and interfere with DNA replication and protein synthesis. (14) Furthermore, silver ions released from nanoparticles may interact with thiol-containing proteins and enzymes, resulting in cellular dysfunction and microbial death. The presence of bioactive phytochemicals from *Carica papaya* leaves on the nanoparticle surface may further enhance antimicrobial efficacy through synergistic interactions (15).

Overall, the present findings indicate that *Carica papaya*-mediated silver nanoparticles possess significant antimicrobial potential and may serve as promising alternatives to conventional antimicrobial agents (16). The combination of biocompatibility, eco-friendly synthesis, and broad-spectrum antimicrobial activity highlights their potential application in biomedical, pharmaceutical, and dental fields. However, further studies investigating cytotoxicity, nanoparticle stability, and in vivo efficacy are necessary before clinical translation.

5. CONCLUSION

The present study successfully synthesized silver nanoparticles using *Carica papaya* leaf extract through a green and sustainable approach. Characterization analyses confirmed nanoparticle formation, while antimicrobial assays demonstrated significant activity against tested pathogens. These findings suggest that *C. papaya*-mediated AgNPs possess promising potential for future biomedical and antimicrobial applications.

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