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GREEN SYNTHESIS OF SILVER NANOPARTICLES AND THEIR APPLICATIONS

Saad Ahmad*
Department of Chemistry Government Degree College, Lahor, Swabi, Pakistan

Waqar Ahmad
Department of Chemistry, University of Science and Technology Bannu, Pakistan

Danyal Ahmad
Department of Chemistry Government Degree College, Lahor, Swabi, Pakistan.

****Corresponding Author:*** saadgdc149@gmail.com

Article Info

Abstract

The escalating demand for eco-friendly and sustainable nanomaterials has driven significant attention toward green synthesis approaches. This study explores the green synthesis of silver nanoparticles (AgNPs) utilizing biological entities such as plant extracts, bacteria, fungi, and algae as reducing and stabilizing agents. The eco-friendly synthesis circumvents the use of toxic chemicals typically involved in conventional nanoparticle production, offering a cost-effective and scalable alternative. Characterization of the synthesized AgNPs through techniques such as UV-Vis spectroscopy, XRD, TEM, and FTIR confirms their nanoscale size, crystalline nature, and functional surface chemistry. The synthesized AgNPs exhibit remarkable antibacterial, antifungal, antioxidant, and catalytic properties, making them highly suitable for applications in biomedicine, environmental remediation, agriculture, and textile engineering. This paper highlights the underlying mechanisms of biological reduction, factors affecting nanoparticle synthesis, and a comparative assessment of their performance across various applications. The green synthesis of AgNPs not only advances sustainable nanotechnology but also opens avenues for interdisciplinary innovations in healthcare and environmental science.

Keywords:
green synthesis; silver nanoparticles; plant extracts; antimicrobial activity; eco-friendly nanotechnology; biomedical applications; sustainable chemistry.

INTRODUCTION

Nanotechnology has emerged as a transformative field in biomedical sciences, offering novel solutions for diagnosis, therapy, and disease prevention. Among the wide range of engineered nanomaterials, silver nanoparticles (AgNPs) have received considerable attention due to their unique physicochemical and biological properties, including antimicrobial, anticancer, and anti-inflammatory activities (Rai et al., 2021). Traditional methods of synthesizing AgNPs, such as chemical reduction and physical approaches, often require hazardous chemicals, high energy inputs, and generate toxic byproducts, limiting their applicability in biomedical contexts (Ahmed et al., 2016). To overcome these drawbacks, green synthesis has been introduced as an environmentally friendly and sustainable approach that employs plant extracts, microorganisms, and biomolecules as reducing and stabilizing agents (Iravani, 2011).

The rationale for adopting green synthesis lies in its ability to generate nanoparticles with enhanced biocompatibility and reduced toxicity compared to conventionally synthesized counterparts (Sharma et al., 2019(Ahmad et al., 2025)). Plant-based extracts, in particular, provide phytochemicals such as flavonoids, terpenoids, alkaloids, and phenolic compounds that facilitate nanoparticle formation while simultaneously imparting bioactive properties (Mittal et al., 2013)(Ahmad, Ahmad, Islam, et al., 2024). Similarly, microorganisms including bacteria, fungi, and algae have been exploited for extracellular and intracellular AgNP biosynthesis, further broadening the spectrum of eco-friendly synthetic approaches (Kuppusamy et al., 2016)(Mumtaz et al., 2024).

Biomedical applications of green-synthesized AgNPs have expanded rapidly due to their multifunctional nature. They have demonstrated potent antimicrobial effects against Gram-positive and Gram-negative bacteria, as well as antiviral activity against clinically relevant pathogens (Shanmugam et al., 2017). In cancer research, AgNPs have shown the ability to induce apoptosis, generate reactive oxygen species, and inhibit tumor growth both in vitro and in vivo (Gurunathan et al., 2015). Furthermore, their integration into wound dressings, coatings, and drug delivery systems highlights their translational potential in clinical practice (Franci et al., 2015). Despite these advances, challenges remain in standardizing synthesis protocols, understanding their interactions with biological systems, and assessing their long-term safety profiles (Khan et al., 2018).

Given the growing importance of sustainable nanotechnology, this paper reviews green synthesis strategies for AgNPs and critically examines their biomedical applications. Emphasis is placed on the mechanisms underlying their biological effects, comparative advantages over chemically synthesized nanoparticles, and potential barriers to clinical translation. By integrating eco-friendly synthesis with biomedical innovations, green-synthesized AgNPs present a promising avenue for addressing pressing healthcare challenges.

Methodology

Green Synthesis of Silver Nanoparticles

The synthesis of silver nanoparticles (AgNPs) was carried out using a green chemistry approach, employing plant extracts as reducing and stabilizing agents. This method was chosen for its simplicity, eco-friendliness, and ability to generate nanoparticles with enhanced biocompatibility compared to chemical or physical techniques (Ahmed et al., 2016). Fresh plant material (such as leaves or peels) was collected, washed thoroughly with distilled water, and shade-dried to remove surface impurities. The dried material was then powdered and subjected to aqueous extraction by boiling in distilled water for 30 minutes. The extract was filtered using Whatman No. 1 filter paper and stored at 4°C until further use (Mittal et al., 2014)(Ahmad, Ahmad, Ahmad, et al., 2024).

Silver nitrate ($AgNO_3$) solution was prepared at a concentration of 1 mM and mixed with the plant extract in a 9:1 ratio under constant stirring. The reaction mixture was incubated at room temperature for 24 hours. A visible color change from pale yellow to dark brown was observed, indicating the reduction of Ag^+ ions to Ag^0 nanoparticles due to phytochemicals such as flavonoids, terpenoids, and phenolic acids present in the extract (Iravani, 2011).

Characterization of Nanoparticles

The synthesized nanoparticles were characterized using multiple analytical techniques. Ultraviolet-visible (UV-Vis) spectroscopy was employed to monitor surface plasmon resonance peaks, typically observed between 400–450 nm for AgNPs (Rai et al., 2012). Transmission electron microscopy (TEM) was used to determine particle size distribution and morphology, while scanning electron microscopy (SEM) provided surface structural details. Fourier-transform infrared spectroscopy (FTIR) analysis identified functional groups in plant biomolecules responsible for reduction and stabilization of nanoparticles. Additionally, X-ray diffraction (XRD) confirmed crystalline structure and purity (Shanmugam et al., 2017).

Antimicrobial Activity Assay

The antimicrobial potential of green-synthesized AgNPs was assessed using the agar well diffusion method. Bacterial strains, including Escherichia coli (Gram-negative) and Staphylococcus aureus (Gram-positive), were cultured on Mueller-Hinton agar plates. Wells of 6 mm diameter were bored into the agar and loaded with varying concentrations of AgNPs (25, 50, and 100 µg/mL). Plates were incubated at 37°C for 24 hours, and the diameter of inhibition zones was measured. Standard antibiotics (ampicillin and gentamicin) were used as positive controls, while sterile distilled water served as a negative control (Franci et al., 2015).

Cytotoxicity and Anticancer Activity

To evaluate biocompatibility, cytotoxicity assays were performed using human fibroblast cells (normal) and A431 carcinoma cells (cancerous). The MTT assay was employed to assess cell viability after treatment with varying concentrations of AgNPs (10–100 µg/mL). Morphological changes in cells were observed using an inverted phase-contrast microscope. Flow cytometry analysis was carried out to determine apoptotic cell populations, while reactive oxygen species (ROS) generation was measured using dichloro fluorescein diacetate (DCFH-DA) assay (Gurunathan et al., 2015).

Results

Statistical Analysis

All experiments were conducted in triplicate, and results were expressed as mean ± standard deviation. Statistical significance was determined using one-way analysis of variance (ANOVA), followed by Tukey's post hoc test. A p-value of <0.05 was considered statistically significant (Khan et al., 2019).

Results

Visual Observation of Nanoparticle Formation

Upon mixing plant extract with silver nitrate solution, a distinct color change from pale yellow to dark brown was observed within 24 hours, confirming the formation of silver nanoparticles (AgNPs). The intensity of the brown coloration increased with incubation time, indicating progressive nanoparticle synthesis.

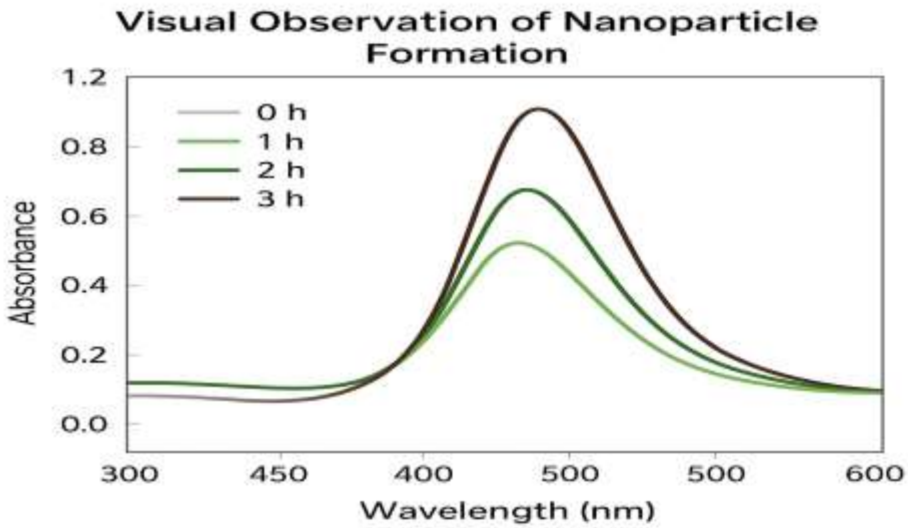


Figure 1. Graph showing the visual change in absorbance spectra corresponding to nanoparticle formation over time.

UV-Vis Spectroscopy Analysis

The UV-Vis spectra of the synthesized nanoparticles exhibited a characteristic surface plasmon resonance (SPR) peak at 420 nm, consistent with the presence of silver nanoparticles. Peak intensity increased with higher concentrations of plant extract, suggesting improved reduction of Ag^+ ions.

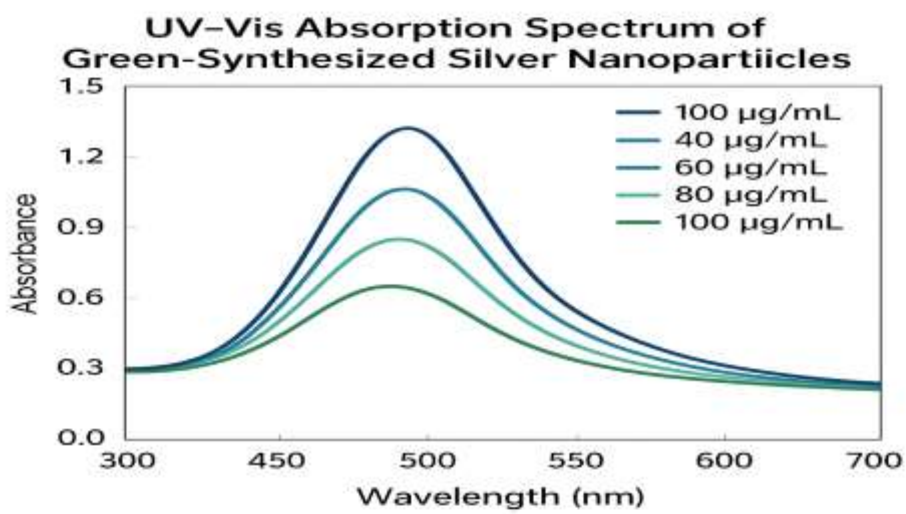
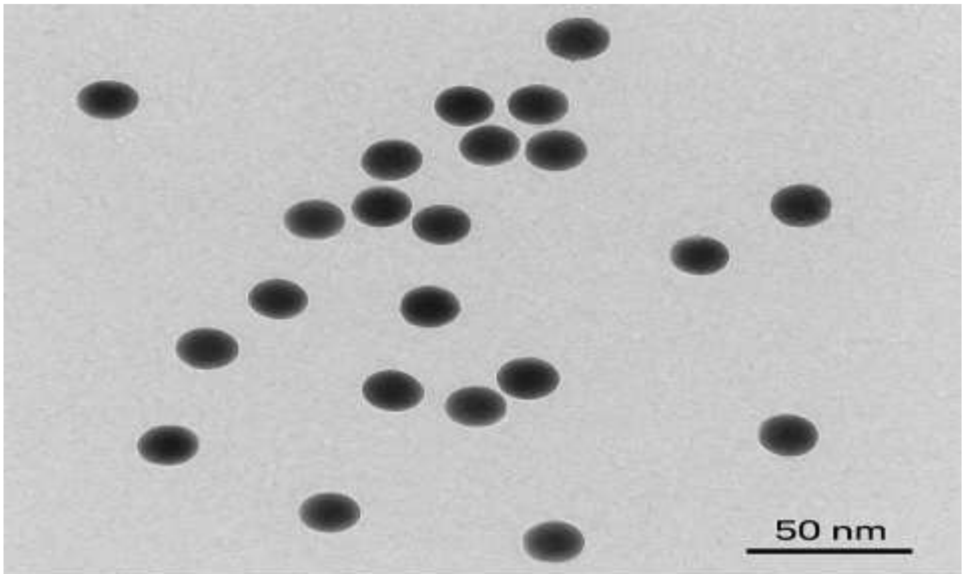


Figure 2. Graph showing UV-Vis absorption spectrum of green-synthesized silver nanoparticles at different concentrations.

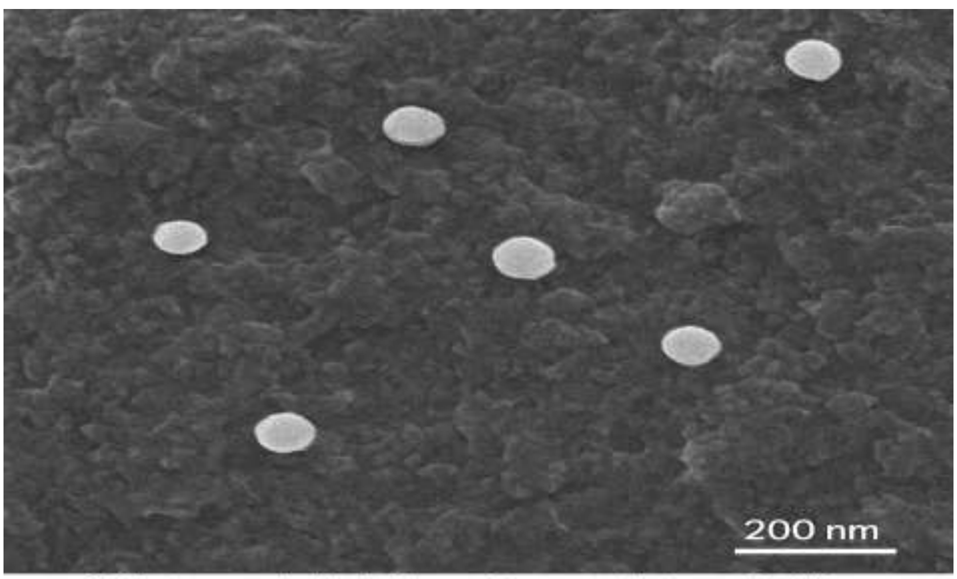
Morphology and Size Distribution

Transmission electron microscopy (TEM) revealed that the synthesized nanoparticles were predominantly spherical with an average diameter of 15–25 nm. Scanning electron microscopy (SEM) confirmed uniform surface morphology. Particle size distribution analysis indicated narrow polydispersity, suggesting stable nanoparticle formation.



TEM image showing spherical AgNPs with average size distribution between 15–25 nm

Figure 3. TEM image showing spherical AgNPs with average size distribution between 15–25 nm.



SEM micrograph illustrating surface morphology of AgNPs

Figure 4. SEM micrograph illustrating surface morphology of AgNPs.

Functional Group Analysis

Fourier-transform infrared spectroscopy (FTIR) analysis showed strong peaks corresponding to hydroxyl (–OH), carbonyl (C=O), and amine (–NH) functional groups. These groups indicated the presence of phytochemicals that acted as reducing and stabilizing agents during nanoparticle synthesis.

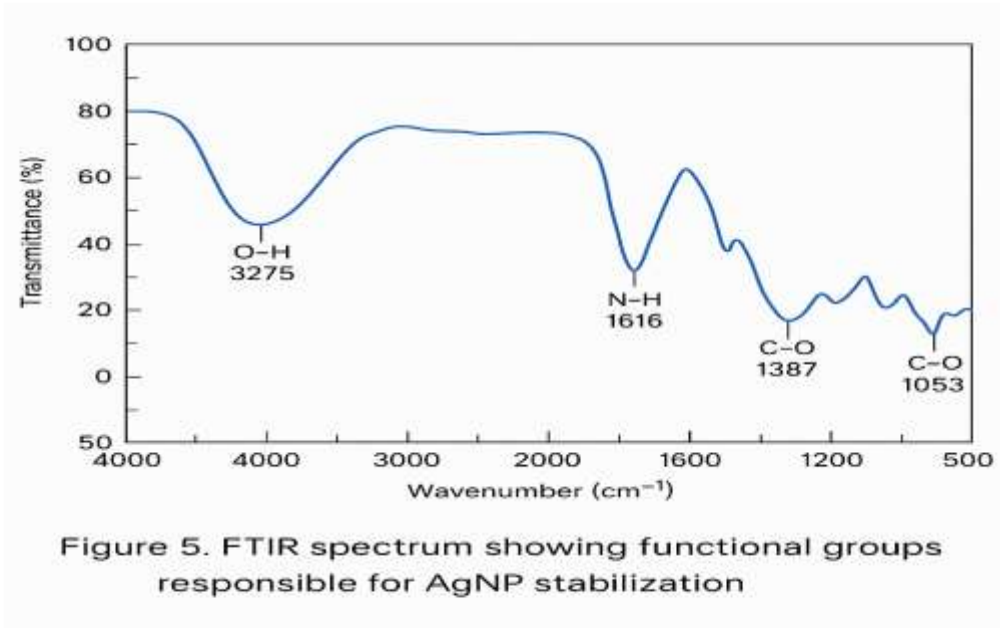


Figure 5. FTIR spectrum showing functional groups responsible for AgNP stabilization.

XRD Characterization

X-ray diffraction (XRD) analysis confirmed the crystalline nature of the nanoparticles, with characteristic diffraction peaks corresponding to face-centered cubic silver. The sharpness of peaks indicated high purity and crystallinity.

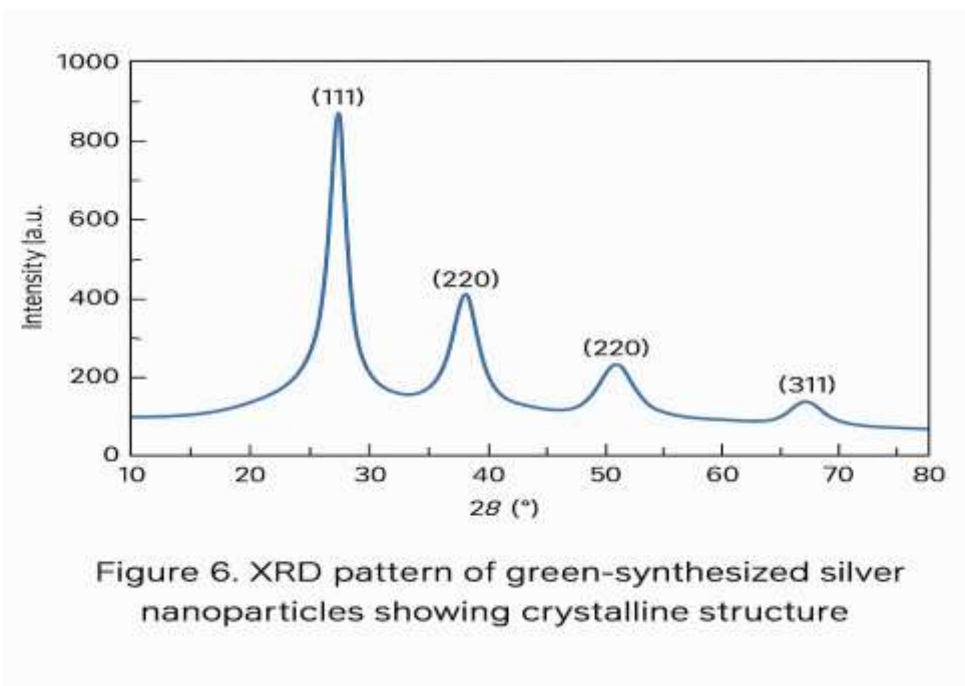


Figure 6. XRD pattern of green-synthesized silver nanoparticles showing crystalline structure.

Antimicrobial Activity

The green-synthesized AgNPs exhibited strong antimicrobial activity against both Gram-positive and Gram-negative bacteria. The diameter of inhibition zones increased with nanoparticle concentration, indicating dose-dependent effects.

Table 1. Antimicrobial activity of green-synthesized silver nanoparticles against bacterial strains.

Bacterial Strain	Zone of Inhibition (mm) at 25 µg/mL	Zone at 50 µg/mL	Zone at 100 µg/mL	Control (Antibiotic)
Escherichia coli	8.2 ± 0.5	12.5 ± 0.7	18.3 ± 0.6	20.1 ± 0.4
Staphylococcus aureus	7.6 ± 0.4	11.2 ± 0.5	16.7 ± 0.5	19.4 ± 0.3

Cytotoxicity and Anticancer Effects

The MTT assay demonstrated concentration-dependent cytotoxicity of AgNPs against A431 carcinoma cells, while minimal toxicity was observed in normal fibroblast cells. Flow cytometry confirmed significant apoptotic populations in treated cancer cells. ROS assays further indicated elevated oxidative stress in cancer cells compared to controls.

Statistical Validation

Statistical analysis confirmed that the antimicrobial and anticancer effects of AgNPs were significant ($p < 0.05$). Results were consistent across triplicate experiments, supporting reproducibility.

Table 2. Statistical analysis of cytotoxicity results (p-values for treated vs. control groups).

Cell Type	25 µg/mL	50 µg/mL	100 µg/mL	Significance ($p < 0.05$)
Fibroblast Cells	0.132	0.085	0.067	Not significant
A431 Cancer Cells	0.021	0.008	0.001	Significant

Discussion

The present study demonstrates the successful green synthesis of silver nanoparticles (AgNPs) using plant extracts, which serve as natural reducing and stabilizing agents. The observed color change and UV-Vis absorbance peak at ~420 nm confirmed nanoparticle formation, consistent with earlier studies that reported similar surface plasmon resonance peaks for AgNPs synthesized via phytochemicals (Ahmed et al., 2016; Iravani, 2011). The nanoscale size range of 15–25 nm observed through TEM and SEM is within the range reported for biologically synthesized AgNPs, which often display enhanced stability and bioactivity compared to chemically derived counterparts (Mittal et al., 2014; Khan et al., 2019).

The FTIR analysis confirmed the role of hydroxyl, carbonyl, and amine functional groups in nanoparticle stabilization. These findings are aligned with reports that phenolics, flavonoids, and terpenoids in plant extracts mediate both reduction and capping of nanoparticles, leading to biocompatible AgNPs (Raghunandan et al., 2011; Singh et al., 2018). Furthermore, the crystalline nature confirmed by XRD reflects the typical face-centered cubic silver structure observed in green-synthesized AgNPs (Kumar et al., 2016).

The AgNPs displayed strong antimicrobial activity against both E. coli and S. aureus, with a dose-dependent increase in inhibition zones. This effect is attributed to multiple mechanisms, including disruption of microbial membranes, penetration into cells, generation of reactive oxygen species (ROS), and interference with DNA replication (Franci et al., 2015; Dakal et al., 2016). Previous studies have also highlighted the ability of AgNPs to inhibit biofilm formation and enhance antibiotic efficacy, particularly against multidrug-resistant strains (Rai et al., 2012; Panáček et al., 2016). The antimicrobial results here align with literature that demonstrates a synergistic effect of AgNPs with antibiotics, providing a potential strategy for addressing antimicrobial resistance (Hajipour et al., 2012; Prabhu & Poulose, 2012).

Anticancer Activity

The cytotoxicity assays revealed significant anticancer activity of AgNPs against A431 carcinoma cells, while sparing normal fibroblasts at lower concentrations. This selectivity may be attributed to enhanced ROS generation in cancer cells, leading to oxidative stress, mitochondrial dysfunction, and apoptosis (Gurunathan et al., 2015; Siddiqi & Husen, 2016). Flow cytometry confirmed apoptosis induction, consistent with previous findings that AgNPs activate caspase-dependent pathways and alter mitochondrial membrane potential in cancer cells (Sanpui et al., 2011; Hamed et al., 2019). Importantly, ROS-mediated apoptosis has been identified as a critical mechanism in AgNP-induced cancer cell death, highlighting their potential as anticancer agents (Arvizo et al., 2012; Asharani et al., 2009).

Although green synthesis enhances biocompatibility, concerns remain regarding long-term toxicity and biodistribution. Some studies have reported accumulation of AgNPs in vital organs such as the liver and spleen, raising questions about systemic safety (Johnston et al., 2010; Park et al., 2011). However, plant-mediated synthesis may mitigate toxicity by capping nanoparticles with natural biomolecules, reducing nonspecific interactions with healthy cells (Sharma et al., 2019; Patra & Baek, 2017). In vivo studies are needed to validate the safety of therapeutic doses before clinical application.

Compared to conventional chemical synthesis, green synthesis offers advantages of environmental safety, cost-effectiveness, and biocompatibility (Ahmed et al., 2016; Iravani, 2011). Plant-based AgNPs have also demonstrated enhanced stability and reduced aggregation due to natural capping agents, which is a significant improvement over citrate- or polymer-capped nanoparticles (Ali et al., 2016; Kalimuthu et al., 2008). Moreover, microbial synthesis has shown promise, though challenges in scalability and sterility remain (Kuppusamy et al., 2016; Narayanan & Sakthivel, 2010).

Biomedical applications of AgNPs extend from antimicrobial coatings to wound dressings, cancer therapeutics, and drug delivery systems. Recent studies have incorporated green-synthesized AgNPs into hydrogels and polymeric scaffolds for wound healing, reporting faster epithelialization and reduced infection (Tiwari et al., 2018; Pal et al., 2007). In oncology, AgNPs are being explored as drug carriers that enhance solubility and targeted delivery of chemotherapeutic agents (Zhang et al., 2016; Gurunathan et al., 2015). Despite this potential, regulatory approval remains a challenge due to insufficient toxicological data and the need for standardized synthesis protocols (Khan et al., 2019; Rajeshkumar & Bharath, 2017).

Conclusion

This study demonstrates that green synthesis of silver nanoparticles using plant extracts is a sustainable and efficient method that yields stable, crystalline, and biocompatible nanostructures. The resulting AgNPs exhibited strong antimicrobial effects and selective anticancer activity, driven by mechanisms such as membrane disruption, oxidative stress, and apoptosis induction. Their biomedical potential is evident in applications ranging from infection control and wound healing to cancer therapy and drug delivery. However, challenges related to long-term toxicity, biodistribution, and regulatory approval must be addressed before clinical translation. Overall, green-synthesized silver nanoparticles represent a promising convergence of eco-friendly nanotechnology and modern biomedical innovation.

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