

Green Synthesis of Silver Nanoparticles from Medicinal Plants: Review of Methods and Applications

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ABSTRACT: Nanotechnology has been an area of research in recent years since it is possible to synthesize materials with normal properties at the nanoscale. Among them, silver nanoparticles (AgNPs) are particularly interesting due to their antimicrobial, antioxidant, anticancer, and catalytic properties. Conventional physical and chemical routes used for the synthesis of AgNPs involve the use of harmful chemicals, high energy input, or costly apparatus, thereby posing environmental and safety risks. Due to these limitations, green synthesis using plant extracts has been a recent green and sustainable alternative. The process relies on different phytochemicals such as flavonoids, terpenoids, phenolics, proteins, and alkaloids, which work with the dual functions of reducers and capping agents. Natural capping, besides eliminating toxic chemicals, also enhances the biological activity of the nanoparticles. Extract concentration, pH, temperature, and incubation time all play important roles in particle size distribution, morphology, and stability. Structural as well as functional properties of the AgNPs synthesized are demonstrated using characterization by methods such as UV–Visible spectroscopy, FTIR, TEM, and XRD. Plant-mediated AgNPs have potential applications in medicine, such as drug delivery, antimicrobial therapy, and wound healing, and in agriculture and environmental remediation, too. Despite all these advancements, however, challenges in reproducibility, scale-up manufacturing, and the lack of toxicity testing remain to be surmounted. Future endeavors will have to tackle standardization, mechanism elucidation, and biosafety evaluation to facilitate industrial and clinical translation. In general, the green synthesis represents an emerging route to low-cost, safe, and multifunctional silver nanoparticles for varied applications.

Keywords- Nanotechnology, Green Synthesis, Silver Nanoparticles (AgNPs), Phytochemicals, Antimicrobial Activity, Biomedical Applications, Environmental Remediation

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I. INTRODUCTION

Nanoparticles are substances with one or more dimensions in the nanometer range (1–100 nm), which display special physical, chemical, and biological properties as opposed to bulk phases. These specific features, including high surface area, increased reactivity, and controlled optical properties, have made it possible for a wide variety of applications in medicine, electronics, agriculture, and environmental control. Nanotechnology is presently one of the most intriguing areas of research because of its new technologies in agriculture, health, and environmental cleaning ^{1,2,3}. Due to their unique optical, electrical, and antibacterial properties, silver nanoparticles (AgNPs) have been specially highlighted among other nanomaterials. Conventional physical methods of synthesizing AgNPs, such as evaporation-condensation and laser ablation, require costly apparatus and much energy^{4,5}. Decreasing chemicals such as sodium borohydride and hydrazine, which are effective but toxic and harmful to the environment, is often employed in chemical approaches. Owing to these limitations, there is an extreme requirement for safer and greener approaches to synthesizing nanoparticles. One such interesting approach to synthesizing AgNPs in an ecologically friendly manner is green synthesis, which employs biological processes ^{6,7}.

Since they are readily available, cheap, and devoid of pathogenic risks, plants are preferred over microorganisms or fungi among biological conduits. Silver ions can be reduced and stabilised simultaneously by a multitude of phytochemicals present in extracts of plants, including phenolics, flavonoids, terpenoids, tannins, and alkaloids ^{8,9}. Moreover, utilising plants renders the process simple and reproducible by eliminating the necessity of sophisticated culture maintenance. Various studies have shown that phytochemical capping of AgNPs mediated by plants leads to enhanced biological activity along with stability. The widespread use of this approach is supported by the finding that successful synthesis of AgNPs has been reported for more than 150 plant species.

High antibacterial activity has been reported for nanoparticles synthesized from Tulsi (*Ocimum sanctum*), green tea (*Camellia sinensis*), and Neem (*Azadirachta indica*). The ability of aloe vera (*Aloe barbadensis*) and turmeric (*Curcuma longa*) extracts to form stable and bioactive AgNPs has also been intensively studied. Through the selection of specific phytochemical profiles, one is able to tailor the characteristics of nanoparticles due to the variation in plant material.¹⁰⁻¹³ Green synthesis has the added advantage of integrating nanotechnology with conventional medical knowledge alongside sustainability. For instance, nanoparticles of similar enhanced activity are often generated from medicinal plants that have been extensively known to possess antibacterial and antioxidant activity^{14,15,16}. The combination of ethnobotany and nanoscience opens up new possibilities for biomedical uses. As a result, plant-mediated synthesis of AgNPs is becoming more and more popular as a cutting-edge and environmentally friendly nanotechnology platform^{17,18}.

II. PRINCIPLES OF GREEN SYNTHESIS

The concept of green synthesis is to reduce silver ions into stable metallic nanoparticles using natural proteins. Two roles are being played by plant metabolites in this regard: as capping or stabilising agents and reducing agents^{19,20}. Phytochemicals readily donate electrons to reduce Ag^+ to Ag^0 when aqueous plant extracts are added to a silver nitrate solution. Surface plasmon resonance tends to result in a visible colour change with this decrease, confirming the nanoparticle formation²¹. The process renders the process environmentally friendly by eliminating the need for toxic chemical reductants. As plant-mediated synthesis is not under the requirement of sterile conditions, it is generally considered to be more effective compared to microbial methods^{22,23}.

Moreover, it does away with the slow growth and maintenance needs of fungal or bacterial cultures. Due to the ease with which extracts from roots, stems, leaves, flowers, and seeds can be prepared, plants allow mass production²⁴⁻²⁶. For companies wishing to manufacture sustainable nanomaterials, this is an important advantage. Additionally, high temperature and high pressure are not required for the process to work under mild reaction conditions^{27,28}. Under green synthesis, the reaction condition plays a significant role in determining the nature of nanoparticles. Nucleation and growth rates of the nanoparticles are affected strongly by the concentration of silver nitrate^{29,30}. The size of the particles is finally regulated by the extract-to-salt ratio, which affects the reducing capacity as well. The pH of the reaction medium could alter the ionisation state of the phytochemicals, which may have an impact on the nanoparticle^{31,32}. The reduction kinetics are regulated by temperature; elevated temperatures give rise to smaller particles and accelerated nucleation. As prolonged incubation ensures complete reduction but could also favor aggregation, reaction time is just as important^{33,34}.

The versatility of green synthesis lies in its ability to form nanoparticles that exhibit a vast variety of morphologies, ranging from spherical, triangular, and rod-like shapes. Plant metabolites both reduce and direct the shape, thus allowing these changes^{35,36}. Hence, employment of natural reducing agents from plant extracts for generating stable, biocompatible, and useful nanoparticles is the foundation of green synthesis³⁷.

The major biomolecules, phytochemicals found in the extracts of plants, are responsible for driving the stabilisation and reduction of silver nanoparticles are referred to as phytochemicals³⁸. Gallic acid and catechins are some phenolic compounds that are strong electron donors and are capable of directly reducing Ag^+ ions to metallic Ag^0 . In the process, flavonoids like kaempferol and quercetin are oxidised, releasing electrons that facilitate nucleation^{39,40}.

In addition, these compounds provide nanoparticles with antioxidant defence, enhancing their stability in biological systems⁴¹. As capping molecules, proteins present in plant extracts bind to the surface of nanoparticles through amino and carboxyl groups⁴². Such capping provides nanoparticles with colloidal stability and prevents agglomeration⁴³. AgNPs are also stabilised by polysaccharides such as starch and cellulose derivatives through steric hindrance, reducing the chance of aggregation⁴⁴. Through silver ion reduction and directing the growth of nanoparticles into specific shapes, terpenoids play dual roles⁴⁵. For example, based on their structure, monoterpenes and diterpenes are known to create spherical and anisotropic nanoparticles⁴⁶.

Another major group of compounds, alkaloids, engages in interaction with metal ions to facilitate controlled reduction, ensuring that particle size is controlled⁴⁷. As natural surfactants, saponins facilitate dispersion and prevent nanoparticles from aggregating in solution. Through the formation of active compounds with silver ions and entrapping the product, tannins stabilize nanoparticles^{48,49}. The diversity and abundance of phytochemicals present in different plants and the ability of these to produce nanoparticles of different sizes, shapes, and biological activities can explain why different plants produce nanoparticles. AgNPs are more bioactive compared to chemically synthesised nanoparticles due to the phytochemical corona surrounding them^{50,51}. By collaborating, this natural corona is able to augment its antibacterial, antioxidant, and anticancer properties. Moreover, studies have revealed that phytochemical-modified silver nanoparticles are less toxic compared to those that are not modified^{52,53}. Plant-mediated nanoparticles are characterized by special properties since phytochemicals act not only as reducing and stabilising agents but also as biological functioning⁵⁴.

Table 1. Major phytochemicals involved in plant-mediated synthesis of silver nanoparticles

S.No.	Phytochemical	Representative Compounds	Function in AgNP Synthesis	Biological Effect on AgNPs	Ref
1.	Phenolics	Gallic acid, Catechins	Strong electron donors; reduce $\text{Ag}^+ \rightarrow \text{Ag}^0$	Antioxidant protection; enhanced nanoparticle stability	38, 39, 41
2.	Flavonoids	Quercetin, Kaempferol	Undergo oxidation, release electrons for nucleation	Improve stability; regulate nanoparticle morphology	40, 41
3.	Proteins	Enzymes, Albumins, Peptides	Bind to AgNP surface via amino & carboxyl groups; act as capping agents	Prevent agglomeration; impart colloidal stability	42, 43
4.	Polysaccharides	Starch, Cellulose derivatives	Steric stabilization; size control	Reduce aggregation; improve dispersity	44
5.	Terpenoids	Monoterpenes, Diterpenes	Reduce Ag^+ ions; direct growth into morphologies	Control nanoparticle shape (spherical/anisotropic)	45, 46
6.	Alkaloids	Nicotine, Berberine	Interact with Ag^+ ; facilitate controlled reduction	Regulate particle size; improve stability	47
7.	Saponins	Triterpenoid saponins	Natural surfactants prevent clumping	Enhance dispersion in solution	48
8.	Tannins	Hydrolyzable & condensed tannins	Form complexes with Ag^+ and cap nanoparticles	Strong stabilization; improved bioactivity	49
9.	Mixed Phytochemicals (Corona)	Polyphenols + proteins	Synergistic reduction + capping	Increase antimicrobial, antioxidant, anticancer activity; reduce cytotoxicity.	50, 51, 52, 53, 54

III. Mechanistic Pathways

A multi-step procedure involving reduction, nucleation, and stabilisation is generally employed in environmentally safe silver nanoparticle production. Phytochemicals donate electrons to minimise Ag^+ ions to metallic Ag^0 atoms during the activation step^{55,56}. Hydroxyl, carbonyl, and amine groups present in the plant metabolites are often involved in this reduction. Small clusters are formed when the initial Ag^0 atoms collide and combine, serving as nuclei for future growth^{57,58}. Particles larger in size are formed due to further deposition of silver atoms onto these nuclei during growth. Some of the parameters that play a critical role in controlling the rate of growth include temperature, pH, and precursor concentration. Smaller and more uniform nanoparticles are often generated by quick nucleation under alkaline pH conditions^{59,60,61}.

FTIR studies, which indicate the presence of functional groups on nanoparticle surfaces, confirm the biomolecule binding. Phytochemicals can also serve as shape-directing agents, states sophisticated mechanistic research⁶²⁻⁶⁴. Terpenoid-rich extracts, for example, could lead to anisotropic morphology such as rods or triangles, but flavonoid-rich extracts generally produce spherical nanoparticles^{65,66}. The unique advantage of green synthesis compared to chemical methods is demonstrated by the role of plant metabolites in controlling shape^{67,68}. Whereas TEM studies reveal the growth and nucleation process, spectroscopic techniques such as UV-Visible and XPS confirm the reduction of silver ions⁶⁹.

In addition, kinetic investigations reveal that, relative to the concentration of silver ions, nanoparticle synthesis often proceeds via a pseudo-first-order mechanism⁷⁰. This mechanistic insight is critical to design reproducible synthesis protocols with targeted nanoparticle properties⁷¹. The step-by-step procedure is depicted in the following Figure 1.

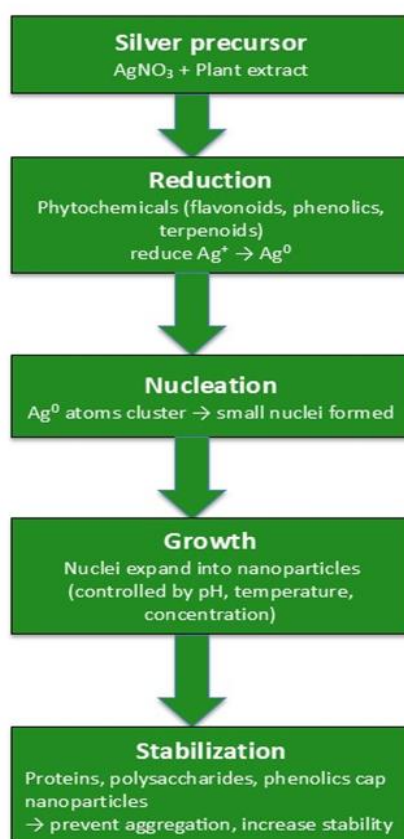


Figure 1. The schematic illustration of the typical pathway

IV. Factors Influencing Synthesis

The physicochemical parameters that exert a strong influence on the properties of silver nanoparticles prepared by green routes are several. Among the most significant parameters is the concentration of silver nitrate, which decides the concentration of available silver ions for reduction^{72,73}. Large precursor concentrations tend to create fast nucleation and the formation of large-sized nanoparticles. Conversely, lower concentration tends to promote the formation of smaller and monodisperse nanoparticles^{74,75}. The proportion between the extract and silver salt is also important in particle formation. Higher extract ratio enhances the reducing capacity of the system and produces smaller nanoparticles^{76,77}. Still, too much extract can add excessive biomolecules and cause aggregation or polydispersity. pH of the reaction medium is another critical parameter in AgNP synthesis^{78,79}. In an alkaline medium, groups like hydroxyls and amines are more active and enhance quick reduction. This tends to form smaller particles with higher stability. In acidic pH, the rate of reduction is slow, and usually, larger or non-uniform particles are formed^{80,81,82}. Temperature has a significant influence on nanoparticle formation kinetics. At higher temperatures, silver ion reduction is rapid, resulting in rapid nucleation and formation of smaller nanoparticles^{83,84}. Heating in the middle range can also improve the crystallinity of nanoparticles, making them more stable. While extreme heating might denature plant metabolites, such that their potential to stabilize the nanoparticles is decreased^{85,86}. Reaction time can both control the size and yield of nanoparticles⁸⁷. Extended incubation times facilitate total silver ion reduction, obtaining a better yield of⁸⁸. However, if the system does not contain adequate stabilizers, extended reactions can result in aggregation and precipitation. Stirring rate and aeration of the reaction mixture also aid in the uniformity of nanoparticles^{89,90}. Controlled mixing leads to silver ions and plant metabolites distribution, ensuring uniformity and avoiding localized supersaturation⁹¹. In addition, the plant part involved (leaf, stem, root, bark, or flower) determines nanoparticle properties because of variations in phytochemical composition. Leaves are used most frequently because they harbor high levels of flavonoids and polyphenols, powerful reducing agents^{92,93}. Roots or seeds can yield nanoparticles with other morphologies due to special biomolecular profiles^{94,95}.

Table 2. Parameters influencing plant-mediated silver nanoparticle synthesis

S.No.	Parameter	Influence on AgNP Synthesis	Outcome	Ref
1.	pH of the reaction medium	Controls the ionization of phytochemicals; affects the reactivity of hydroxyl and amine groups	Alkaline pH → smaller, uniform, stable particles; Acidic pH → slower reduction, larger/irregular particles	79, 80, 81, 82
2.	Temperature	Modulates the reduction kinetics and crystallinity of AgNPs	Higher temp. → faster nucleation, smaller NPs; Moderate heating → better crystallinity; Excessive heating → destabilization/aggregation	83, 84, 85, 86
3.	Silver salt concentration	Determines nucleation density and availability of Ag ⁺ ions	Low conc. → monodisperse, controlled growth; High conc. → rapid nucleation, polydispersity, irregular shapes	72, 73, 74, 75
4.	Plant extract concentration	Defines the availability of biomolecules for reduction and capping	High extract/Ag ⁺ ratio → smaller, stable NPs; Too much extract → aggregation/polydispersity; Low extract → incomplete reduction	76, 77
5.	Reaction time	Affects the completeness of reduction and stability of NPs	Adequate time → complete reduction, stable NPs; Prolonged time without stabilizers → aggregation or precipitation	87, 88
6.	Light/Radiation	Activates phytochemicals; enhances reduction efficiency	Sunlight/UV/microwave → accelerated synthesis, smaller particles	90
7.	Stirring & Aeration	Ensures homogeneous mixing of silver ions and phytochemicals	Improved uniformity; prevents localized supersaturation	89, 90, 91
8.	Plant part used	Different phytochemical composition in leaves, stems, roots, bark, and flowers.	Leaves → rich in polyphenols, yield smaller/stable NPs; Roots/seeds → distinct morphologies	92, 93, 94, 95

V. Characterization Techniques

Characterization of silver nanoparticles (AgNPs) synthesized greenly is significant to confirm successful synthesis and to determine their physicochemical properties, including size, shape, crystallinity, surface chemistry, stability, and quality. Appropriate characterization gives reproducibility, predictability of biological activity, and selection for specific applications. Several spectroscopic, microscopic, and analytical techniques are commonly employed to achieve a general overview of such nanoparticles ⁹⁶.

1. **UV-Visible spectroscopy**- The optical properties inherent in silver nanoparticles make UV-Visible spectroscopy the typical first port of call for tracking their synthesis. AgNPs typically have a characteristic Surface Plasmon Resonance (SPR) band at 400-450 nm. Inferred data regarding size, shape, and extent of nanoparticle agglomeration can be gleaned from the position, width, and intensity of this SPR peak. Monodisperse, spherical nanoparticles are reflected in a sharp, intense peak; peak broadening or shifting demonstrates agglomeration or polydispersity. ^{97,98}
2. **Fourier Transform Infrared (FTIR) spectroscopy**- FTIR spectroscopy detects plant extract functional groups responsible for nanoparticle and silver ion stabilisation to some extent. Phytochemical surface interaction with the nanoparticle is established through the presence of peaks for hydroxyl (-OH), carbonyl (C=O), and amine (-NH₂) groups. Organocapping agent functionality, e.g., biomolecules, improves colloidal stability and blocks particle agglomeration. ^{99,100}
3. **X-ray Diffraction (XRD)**- The crystalline character of the synthesized nanoparticles is fully revealed by XRD examination. Bragg reflection peaks characteristic of (111), (200), (220), and (311) planes ascertain metallic silver's Face-Centered Cubic (FCC) structure. According to the Debye-Scherrer formula, XRD patterns can also be utilized to estimate crystallite size, which can provide information about the homogeneity of nanoparticles. ^{101,102}
4. **Transmission Electron Microscopy (TEM)**- The excellence standard in high-resolution images of particle shape and size is TEM images, which are direct nanoscale dimensional evidence and assist in the size correlation with optical and biological properties. TEM images are direct visual proof of the particle shape (sphere, rod, triangle, etc.), size distribution, and aggregation state ^{103,104}.
5. **Scanning Electron Microscopy (SEM) and Energy-Dispersive X-ray Spectroscopy (EDX)**- SEM is used to examine topographical features and the surface morphology of the nanoparticles. SEM also offers elemental composition via analysis in case EDX is used with it, verifying the presence of silver and excluding the possibility of serious contamination. SEM comes in handy in determining the degree of aggregation and the surface roughness on dry samples. ^{105,106}
6. **Dynamic Light Scattering (DLS)**- Diameter of nanoparticles in suspension is determined by DLS; because of hydration layers, the diameter will be greater than a one estimated by TEM. In addition, DLS gives the Polydispersity Index (PDI), a quantitative measure of dispersion and homogeneity of the nanoparticles. Low PDI values are indicative of a monodisperse system ideal for repeated use. ^{107,108}
7. **Zeta potential**- Zeta potential measurements establish the surface charge and colloidal stability of nanoparticles. Aggregation is prevented by electrostatic repulsion, and therefore, nanoparticles with zeta

potentials greater than ± 30 mV are considered to be stable. Measurement is significant from the viewpoint of predicting the behavior of the biological system and storage stability.^{109,110}

8. **Thermogravimetric Analysis (TGA)**- TGA measures organic capping material in plant extracts and also analyzes the thermal stability of nanoparticles. Decomposition temperatures of organics of coatings and the level of interaction of phytochemicals with nanoparticle surfaces are both indicated by this work.^{111,112}
9. **X-ray Photoelectron Spectroscopy (XPS)**- XPS identifies the chemical state of silver and surface-bound protein content. XPS reports on surface contamination or oxidation and distinguishes between metallic silver (Ag^0) and ionic silver (Ag^+). In relating surface chemistry to biological or catalytic activity, XPS is particularly valuable.^{113,114}
10. **Atomic Force Microscopy (AFM)**- AFM provides data on surface roughness, topography, morphology, and aggregation tendency through the creation of three-dimensional topography images. Because it is capable of measuring the height of a particle and also position in space at the nanoscale, AFM is highly suited to study nanoparticles that have been deposited on solid substrates.^{115,116}
11. **Inductively Coupled Plasma Mass Spectrometry (ICP-MS) or Atomic Absorption Spectroscopy (AAS)**- ICP-MS and AAS are employed to find the concentration of silver in nanoparticles and synthesis efficiency. They also facilitate the tracking of the release of silver ions, which is crucial to ascertain potential cytotoxicity and environmental impact.^{117,118}

Hence, the complementarity of these methods for complete characterization of green-synthesized silver nanoparticles confirms their formation, structural stability, and potential application in different areas of medicine, catalysis, and pollution remediation. Effective characterization of green-synthesized silver nanoparticles is guaranteed through the integration of spectroscopic, microscopic, and analytical methods¹¹⁹.

Table 3. Characterization techniques used for plant-mediated silver nanoparticles (AgNPs)

S.No.	Technique	Purpose/Principle	Key Findings for AgNPs	Ref.
1.	UV-Visible Spectroscopy	Detects surface plasmon resonance (SPR) band due to electron oscillation	Confirms nanoparticle formation; SPR peak at 400–450 nm; peak shift indicates particle size and aggregation	97, 98
2.	FTIR	Identifies functional groups of biomolecules attached to nanoparticles	Peaks of $-\text{OH}$, $-\text{C}=\text{O}$, $-\text{NH}$, and $-\text{COOH}$ confirm the role of phenolics, proteins, and flavonoids as reducing/capping agents	99, 100
3.	XRD	Determines crystalline structure and phase	Characteristic peaks at $2\theta \approx 38^\circ, 44^\circ, 64^\circ, 77^\circ$ confirm FCC structure of Ag; Debye–Scherrer equation estimates crystallite size	101, 102
4.	TEM	Provides high-resolution size and morphology	Reveals size distribution, shape, lattice fringes; SAED patterns confirm crystallinity	103, 104
5.	SEM	Visualizes surface morphology and particle distribution	Shows overall shape (spherical, triangular, rod-like); often coupled with EDX for composition	105, 106
6.	EDX (SEM-EDX)	Determines elemental composition	Strong signal at ~ 3 keV confirms silver; minor peaks from capping biomolecules/impurities.	106
7.	DLS	Measures the hydrodynamic diameter and size distribution in the colloidal state	Hydrodynamic size is usually larger than TEM; PDI indicates nanoparticle uniformity.	107, 108
8.	Zeta Potential	Measures surface charge for colloidal stability	Values $\geq \pm 30$ mV indicate stable nanoparticles via electrostatic repulsion	109, 110
9.	TGA	Evaluates weight loss upon heating	Quantifies organic capping layer from phytochemicals; shows thermal stability	111, 112
10.	XPS	Determines oxidation state and surface chemistry	Differentiates Ag^0 (metallic) and Ag^+ (ionic); confirms biomolecule binding.	113, 114
11.	AFM	Provides 3D topographic surface images	Measures nanoparticle roughness, aggregation tendencies on solid substrates	115, 116
12.	ICP-MS / AAS	Quantifies silver content and ion release	Determines synthesis efficiency; monitors Ag^+ release for toxicity evaluation.	117, 118

VI. Biological Applications

Silver nanoparticles synthesized via green pathways have demonstrated a broad spectrum of antimicrobial activity against both Gram-positive and Gram-negative bacteria. Antimicrobial activity is primarily attributed to the release of Ag^+ ions, which degrade bacterial cell membranes and walls^{120, 121}. AgNPs also generate reactive oxygen species (ROS), leading to oxidative stress and elimination of bacterial cells. In addition, nanoparticles can come into contact with proteins and DNA and interfere with replication and enzymatic activity^{122,123}. Green-synthesized AgNPs have been found to possess excellent antifungal activity against the pathogens of fungi such as *Candida albicans* and *Aspergillus* species. They can cause damage to fungal membranes and also interfere with spore germination^{124,125}. Certain research has indicated that they can be employed as antifungal coatings on medical devices as well as in agriculture. AgNPs also have promising antiviral

effects, with evidence of inhibiting HIV, influenza, and SARS-CoV-2^{126,127}. The mechanism is by binding to the virus surface proteins and hence preventing attachment and entry into the host cells. Anticancer treatment is another major application in which AgNPs induce apoptosis in cancerous cells through inducing oxidative stress and mitochondrial injury^{128,129}. They have been reported to suppress the proliferation of breast, lung, and liver cancer cell lines. More importantly, phytochemical-coated nanoparticles are less toxic than chemically produced AgNPs to normal cells^{130,131}. Plant-synthesized AgNPs also possess appreciable antioxidant activity because they are present on the surface with bioactive phytochemicals. Such antioxidant activity makes them of immense value for reducing oxidative stress in food and biomedical sectors^{132,133}. In drug delivery, AgNPs have been explored as drug nanocarriers for controlled drug release. Their large surface area and tiny size facilitate drug loading and target-specific delivery.^{134,135}

AgNPs green-synthesized are also widely used in wound healing, where tissue regeneration is stimulated and microbial infection is inhibited. Topical formulations of AgNPs have been shown to lead to faster wound closure and reduced inflammation in animal models^{136,137}. In agriculture, AgNPs are used as plant growth promoters and nanopesticides. They control phytopathogens with decreased usage of chemical pesticides^{138,139}. Environmental applications include the treatment of water, where AgNPs are utilized to adsorb dyes, heavy metals, and microbial contaminants. They catalyze toxic pollutant degradation in wastewater due to their excellent catalytic activity. The multifaceted applications of green-synthesized AgNPs thus make them a desirable choice for medicine, agriculture, and environmental protection¹⁴⁰⁻¹⁴².

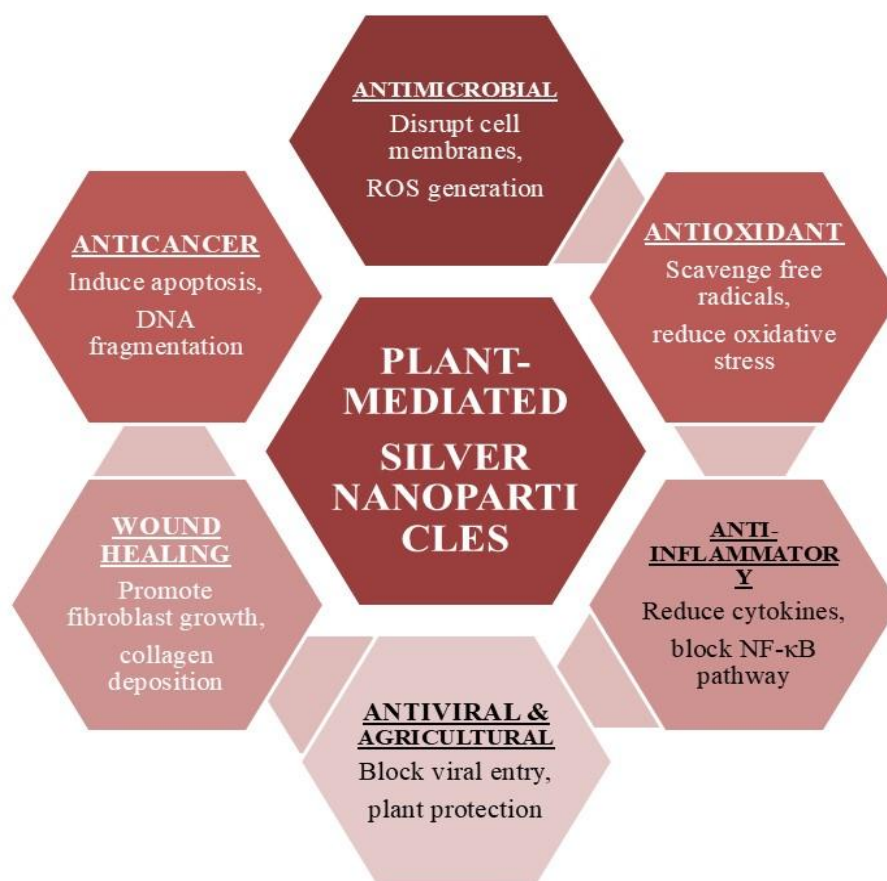


Figure 2. Biological applications of plant-mediated silver nanoparticles (AgNPs)

VII. Toxicity of Green-Synthesized Silver Nanoparticles

Though they are used in many applications, their potential to be toxic is of serious concern. Toxicity is most often because of release of Ag⁺ ions, as the ions have the capacity to interact with cellular components and disrupt homeostasis. AgNPs can cause the generation of ROS, which can induce oxidative stress, lipid peroxidation, and DNA damage¹⁴³⁻¹⁴⁵.

In vitro experiments have established AgNPs' capacity to reduce cell viability in various lines of mammalian cells in a dose-dependent manner. Cytotoxicity is defined by mitochondrial injury, impairment of membranes, and induction of apoptosis^{146,147}. Human lung epithelial cells and liver cells are particularly susceptible to treatment with AgNPs, with implications for biomedical applications¹⁴⁸. In vivo rodent research suggests intravenous or oral exposure to AgNPs can lead to AgNP deposition in vital organs like the brain, spleen,

kidney, and liver ¹⁴⁹. It is preceded by oxidative stress, inflammation, and histopathological alterations. AgNPs pass across biological barriers like the blood–brain barrier, and the possible risk of neurotoxicity is a concern ^{150,151}. Behavioral and neurochemical changes were seen after long-term exposure to nanoparticles in animal models. Ecotoxicological studies confirm that AgNPs have adverse effects on aquatic flora and fauna such as algae, daphnia, and fish ^{152,153}. They can inhibit photosynthesis in algae, cause disruption in the reproduction of aquatic invertebrates, and cause gill injury in fish. Toxicity is highly sensitive to shape, size, coating on the surface, and concentration ^{154,155}. Greater surface area and reduced nanoparticle size of AgNPs release more Ag⁺ ions and are more harmful. Interestingly, the green-synthesized AgNPs have been traditionally reported to be less toxic than chemically synthesized ones due to phytochemical capping ^{156,157}. The chemical capping functionalities utilized during their synthesis can reduce the direct interaction of silver with the biological membrane and oxidative stress. Their biosafety in the long term is not yet completely known, and formal toxicological evaluations are required before they can be applied in clinical or environmental settings ^{158,159}.

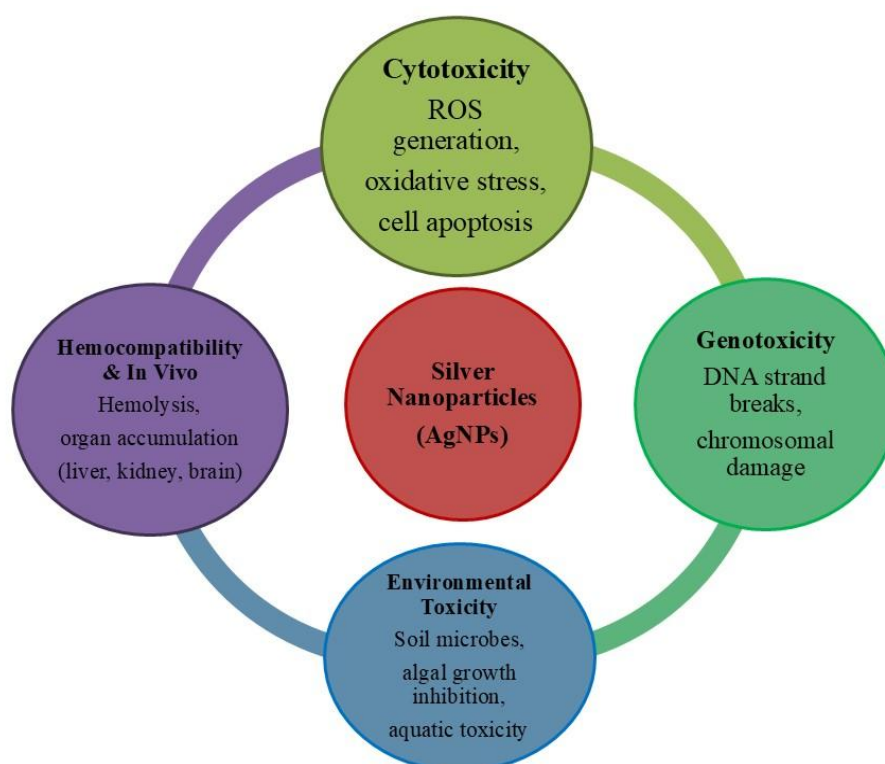


Figure 3. Toxicity pathways of silver nanoparticles (AgNPs).

VIII. Challenges and Future Prospects

Green synthesis of silver nanoparticles is a highly promising strategy, but various challenges lie ahead for large-scale application. One of the main concerns is a lack of reproducibility, wherein different plants or even the same plant harvested in different seasons can produce nanoparticles with differing properties ^{160,161}. Inhomogeneity of the phytochemical composition poses difficulties in standardizing synthesis protocols in laboratories. Another shortcoming is the inability to transfer laboratory practices to large-scale production, keeping nanoparticle size and stability constant ^{162,163}.

Batch-to-batch consistency tends to lead to polydispersity, decreasing application reliability in medicine and industry. Toxicological issues also discourage the commercialization of AgNP-based products ^{164,165}. Even though green-synthesized nanoparticles are likely to be safer than chemically synthesized nanoparticles, long-term biosafety assessment is still inadequate. Regulatory agencies need precise toxicological information prior to sanctioning clinical or agricultural applications ^{166,167}.

There is another challenge posed by the limited mechanistic insight into phytochemical interactions upon synthesis. There is a necessity for sophisticated studies combining omics technologies and molecular modeling to determine the specific biomolecules involved in reduction and stabilization ^{168,169}. Cost-effective strategies for purification are also required to isolate nanoparticles from plant residues and achieve high purity. Improper purification can downgrade nanoparticle quality and restrict biomedical applications ^{170,171}. Application-wise, the laboratory-to-clinical application translation is still restricted. The majority of studies target in vitro antimicrobial or anticancer activities, but few have proceeded to clinical trials ^{172,173}.

Closing this gap demands interdisciplinary efforts between nanotechnologists, pharmacologists, and clinicians¹⁷⁴. Future studies should focus on standardizing the protocols to ensure scalability and reproducibility for industrial applications¹⁷⁵. The combination of green nanotechnology with machine learning and artificial intelligence can enhance the speed of optimization of synthesis parameters¹⁷⁶. Further, the integration of green synthesis with other green nanotechnologies, like the use of biopolymer-based stabilization, has the potential to enhance efficacy and safety further¹⁷⁷. Finally, the future of green-synthesized silver nanoparticles is in harmonizing their vast potential applications with stringent evaluations of safety, reproducibility, and environmental sustainability¹⁷⁸.

IX. Conclusion

Green synthesis of silver nanoparticles has become a green and sustainable methodology compared to the traditional physical and chemical processes¹⁷⁹. The employment of plant extracts offers a cost-effective, facile, and scalable method for nanoparticle fabrication independent of hazardous chemicals¹⁸⁰. Phytochemicals like flavonoids, terpenoids, and phenolics not only reduce silver ions but also stabilize the nanoparticles, increasing their biocompatibility¹⁸¹. Detailed studies have validated the multifaceted applications of green-synthesized AgNPs, such as antimicrobial, antifungal, antiviral, anticancer, and antioxidant activities¹⁸². These nanoparticles also have potential applications in agriculture, drug delivery, wound healing, and environmental remediation as versatile functional materials¹⁸³.

Challenges such as reproducibility, mass production, and toxicity evaluation remain key challenges in spite of such progress¹⁸⁴. There is a shortage of standardized protocols as well as inadequate long-term safety evaluations, which impede clinical and industrial translation¹⁸⁵. Optimization of synthesis parameters, attainment of consistency with different plant sources, and the incorporation of cutting-edge analytical tools for mechanistic insight will be the focus of future studies¹⁸⁶.

Interdisciplinary research involving nanotechnology, pharmacology, and environmental science will be needed to realize the full potential of green-synthesized AgNPs¹⁸⁷. Therefore, though the area promises much, judicious balancing of innovation with safety and sustainability will make the successful use of these nanoparticles a reality in the future¹⁸⁸.

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