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A REVIEW OF SILVER NANOPARTICLE SYNTHESIS METHODS AND THEIR FUNCTIONAL APPLICATIONS

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ABSTRACT
Background: The growing interest in the biosynthesis of nanoparticles, particularly silver nanoparticles (AgNPs), has gained massive attention due to their wide range of applications in electronics, biosensors, optics, catalysis, food, and environmental fields. Medical applications of AgNPs are mainly attributed to their substantial antimicrobial property and their relatively low toxicity under appropriate conditions. Plant extracts are more convenient, offering an eco-friendly, cost-effective approach that uses phytochemicals as natural reducing and stabilizing agents. However, a major gap is the lack of standardized synthesis techniques, leading to inconsistent nanoparticle properties and biological efficacy. **Methodology:** This review aims to provide a detailed study of the diverse synthesis routes of AgNPs and their applications. The study focuses on the impact of plant extract composition on other synthesis conditions. Characterization of nanoparticles is also discussed. **Results and Discussion:** Studies have shown that plant-derived AgNPs demonstrate better antimicrobial activity against various microorganisms, including bacteria and fungi. AgNPs synthesized from *Ipomoea asarifolia* exhibited strong antibacterial activity (IC_{50} of 1 µg/mL) against bacterial pathogens. AgNPs produced using *Cocos nucifera* leaf extract showed significant antibacterial activity, with the highest activity against *Citrobacter freundii* (20 mm) and *Pseudomonas aeruginosa* (19 mm). AgNPs synthesized from *Ricinus communis* extracts remained stable at temperatures up to 100 °C and within a pH range of 5–6 for several months. **Conclusion:** The present review details the factors affecting the synthesis of AgNPs. Furthermore, it provides an overview of scalability challenges and the underlying molecular mechanisms in current research.

INTRODUCTION

Nanotechnology is a rapidly evolving field with applications in numerous commercial products worldwide. Scientific communities have observed significant progress in these areas [1-2]. Nanotechnology primarily focuses on nanoparticles around 1 to 100 nm in size. Currently, nanoparticles are widely used commercially across diverse sectors, including electronics, energy, and pharmaceuticals, owing to their unique properties. Among noble metal nanoparticles, silver nanoparticles (AgNPs) hold a prominent position in pharmaceutical and medical

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applications due to their versatility. Biosynthesized AgNPs, produced via green chemistry approaches using plant extracts and microbial agents, are preferred for therapeutic use due to their enhanced biocompatibility compared with those synthesized by chemical routes. The adoption of green chemistry methods for nanoparticle production is attractive because it ensures environmental sustainability and offers economic advantages, given the non-toxic, cost-effective nature of nanoparticles [3].

AgNPs play a key role in numerous biomedical applications, including serving as antimicrobial, drug delivery, and imaging agents, protective coatings for medical devices, and components in diagnostic and optoelectronic systems. Through a variety of synthesis processes, reducing agents and stabilizers can be used to adjust the shape and size of nanoparticles to suit specific physical, optical, and biological properties. Studies have indicated that the optical, physical, and catalytic properties of AgNPs are significantly influenced by variables such as shape, size distribution, and surface features [4].

Since ancient times, silver has been a naturally occurring element on Earth, valued for its therapeutic properties and ability to preserve food. Compared with bulk silver and many silver salts, AgNPs have greater contact with microbes due to their larger surface area, which makes them more effective antibacterial agents [5]. Silver particles are also used as food additives, in textiles, packaging, and plastics to inhibit bacterial growth. Owing to their numerous applications, various techniques have been developed for producing AgNPs and other compounds containing metallic silver (Ag^0) [3]. The present work focuses on various approaches to AgNP synthesis and provides an overview of their properties and uses. The present study primarily addresses the biological production of AgNPs by using a wide variety of biological resources. Among these, plant-derived nanoparticles are targeted for their notable benefits, including environmental sustainability and biocompatibility. Moreover, the work highlights several characterization techniques and different features that affect the synthesis process of AgNPs. AgNPs also have various applications in biomedical and environmental issues.

MATERIAL AND METHOD

An extensive literature survey was conducted to identify relevant studies on the green synthesis of AgNPs using plant extracts and their biomedical applications. The search was conducted using different electronic platforms, such as PubMed, Web of Science, Scopus, and ScienceDirect, with different keywords like "AgNPs", "green synthesis", "plant extract", "phytochemicals", "antimicrobial activity," and "mechanism of action. The literature search was limited to peer-reviewed English-language articles, mostly published between 2018 and 2025, with a few studies published before 2018 included to provide essential background information. The selected literature was evaluated based on its relevance to the objectives of this review, with emphasis on studies relating to the biosynthesis of AgNPs using plant extracts, and quantitative data on nanoparticle size, synthesis conditions, or antimicrobial efficacy (e.g., minimum inhibitory concentration (MIC) or zone of inhibition) were included. To deliver a comprehensive overview of present advances, fundamental mechanisms, challenges, and future perspectives in this fast-evolving field, the collected literature was critically analyzed. This manuscript is a narrative review of the available evidence and was not conducted as a formal systematic review in accordance with PRISMA guidelines.

SYNTHESIS OF SILVER NANOPARTICLES

The key properties of nanomaterials are closely related to their shape, size, interactions with stabilizing agents, the surrounding medium, and the synthesis method. However, achieving uniformity in these nanoparticle parameters remains a major challenge [6]. Consequently, precise control over the synthesis process is essential to produce nanocrystals with specific characteristics. The optical, magnetic, electrical, and catalytic properties of nanoparticles are largely governed by their morphology and chemical surroundings. During the synthesis of AgNPs, two main approaches are followed – top-down and bottom-up approaches [7]. In the top-down approach, physical forces are applied to break down bulk materials into nanoscale particles. Several mechanical techniques are considered for top-down synthesis, such as crushing, grinding, and milling; thermal processes such as vapor condensation; and electrical methods such as laser ablation and arc discharge. Nanoparticles produced via these physical methods are typically highly pure and exhibit a relatively uniform size distribution. Furthermore, these methods avoid the use of harmful chemicals or stabilizing agents, which restrict nanoparticle agglomeration. In other words, the bottom-up approach focuses on forming nanoparticles utilizing precursor salts as starting materials via nucleation and subsequent growth. Some of the bottom-up techniques include photochemical, chemical, and biological (green) approaches. Chemical methods can be made more efficient by using external stimuli such as light, electricity, microwaves, or ultrasound, enabling rapid nanoparticle production across diverse morphologies. However, the presence of toxic reagents in chemical processes can pose safety concerns, especially for healthcare uses. Biological synthesis has received increasing attention in recent years owing to its environmentally friendly nature. This method employs naturally derived compounds from plants and microorganisms—such as flavonoids, alcohols, alkaloids, quinones, phenolic compounds, terpenoids, enzymes, cellulose and exopolysaccharides—to facilitate nanoparticle formation [8]. A detailed comparison of the two approaches across various parameters is provided in Table 1.

Table 1: Comparison between the top-down and bottom-up approaches based on various parameters [8]

Parameter	Physical Method	Chemical Method	Photochemical Method	Biological (Green) Method
Approach	**Top Down**	**Bottom up**		
Examples of Techniques	Laser Ablation Evaporation Condensation Ball Milling	-Chemical Reduction ($NaBH_4$, citrate) --Sol-Gel -Co-precipitation -Microemulsion	- UV/Visible Light Assisted Synthesis - Photosensitizer-Assisted Reduction	- **Plant Extracts** (e.g., neem, tulsi) - **Fungi** (e.g., *Fusarium oxysporum*) - **Bacteria** (e.g., *Bacillus subtilis*) -A**lgae** (e.g., *Sargassum* spp.)
Biological Sub-methodologies	—	—	—	**1. Plant-Mediated Synthesis** (phytochemicals reduce metal ions) **2. Microbial Synthesis** (enzymes/proteins assist reduction) **3. Algal Synthesis** (marine/terrestrial)
Key Reducing Agents	—	Reducing agents (e.g., $NaBH_4$, citrate)	Photons + sensitizers	- Plant polyphenols, flavonoids - Microbial enzymes/proteins - Algal polysaccharides, metabolites
Cost	High	Moderate to High	Moderate	Low
Yield	Moderate to High	High	Moderate to High	Variable
Size Control	Moderate	High	High	Moderate
Scalability	Limited	High	Moderate	Moderate (depends on biological system & culture scalability)
Environmental Safety	Low	Low to Moderate	Moderate	High (minimal toxic waste)
Energy Requirement	High	Moderate	Moderate	Low (ambient conditions)
Reaction Time	Fast	Fast	Moderate	Slow to Moderate (hours to days)
Reproducibility	High	High	Moderate	Variable (biological extract composition varies)
Stability	Moderate	High (with additives)	Moderate – depends on protection from light/oxidation	Good (natural capping)

As shown in Table 1, nanoparticle synthesis methods exhibit notable differences in yield, reproducibility, and stability. Chemical methods typically offer high yields and excellent reproducibility due to well-defined reaction protocols and controlled conditions, often producing stable nanoparticles using synthetic capping agents. However, their dependence on toxic chemicals raises environmental and safety concerns. Physical methods such as milling or laser ablation can also yield large quantities of nanoparticles but often offer limited control over shape and size, resulting in moderate reproducibility and low stability without post-synthesis stabilization. Photochemical methods provide a cleaner, light-driven alternative with moderate-to-high yields and reasonable size control. However, their reproducibility can be affected by light intensity and environmental conditions, and nanoparticle stability may decline under prolonged exposure to light. In contrast, biological methods—especially plant-mediated synthesis—are eco-friendly and inherently produce stable nanoparticles due to natural capping by biomolecules. But they are often limited by lower yields and poor reproducibility owing to variability in biological materials. Overall, although chemical synthesis remains dominant in industrial applications, the growing emphasis on green chemistry is driving interest in biological and photochemical approaches, despite their current limitations in consistency and scalability. A detailed summary of the various methods is provided below:

Physical Approach

In physical processes, evaporation-condensation is the usual technique for synthesizing metal nanoparticles at atmospheric pressure using a tube furnace. This process vaporizes the starting material placed in a boat inside the furnace and carries it with the carrier gas. However, several drawbacks are associated with synthesizing AgNPs using a tube furnace. It takes a longer time to reach thermal stability, requires a large amount of space, consumes a large amount of energy, and increases the temperature of the surroundings. Alternatively, AgNPs can be synthesized through laser ablation using metallic bulk materials in solution [9-11]. A key advantage of laser ablation over traditional methods is that it does not require chemical reagents to produce highly pure colloidal nanoparticles. Sources of physical energy are commonly employed in the physical synthesis of AgNPs to produce uniform-sized nanoparticles, enabling the production of large quantities of AgNP powder in a single process [12].

Chemical Approach

Chemical reduction is a commonly employed technique due to its convenience and simple apparatus requirements. To achieve metal nanoparticles of narrow diameter distribution, spherical shape, and small size, precise control over nanoparticle growth is necessary. AgNPs, in particular, can be produced cost-effectively and with high yield through chemical reactions. Typically, the preparation of AgNPs involves three key components: reducing agents, capping (or stabilizing) agents, and metal precursors. The reduction process proceeds through two phases to generate colloidal solutions: nucleation and subsequent growth, which considerably influence the size and shape of the synthesized nanoparticles. For producing monodisperse AgNPs with a uniform size distribution, all nuclei must form simultaneously. Both initial nucleation and subsequent growth are regulated by parameters such as pH, temperature, precursor concentration, stabilizing agents (like PVP, PVA, and sodium oleate), and reducing agents (like glucose, ethylene glycol, and $NaBH_4$) [13-15].

Photochemical Approach

Photochemical synthesis is an effective and promising method for producing AgNPs with controlled size and well-defined shapes. Under suitable conditions, the photoreduction process enables the efficient conversion of spherical nanoparticles into triangular nanoplates. Compared with conventional chemical methods, photochemical approaches have several advantages: the process can be carried out in the absence of toxic or hazardous chemicals and any expensive equipment under room-temperature, atmospheric-pressure conditions.

This method typically involves a metal precursor (such as a salt or complex), a stabilizing agent, and a reducing agent. The reaction starts with the photocatalytic reduction of metal ions (Mn+) to their zero-valent metallic state (M^0) using a reducing agent under light exposure. These M^0 atoms serve as nucleation sites, which then grow into metallic nanoparticles. Stabilizers or capping agents help to prevent agglomeration and enhance the colloidal stability of the resulting nanomaterials [16]. Huang and Yang synthesized AgNPs by photoreducing silver nitrate ($AgNO_3$) in suspensions of layered inorganic clay to prevent nanoparticle aggregation [13]. Continuous light irradiation helps convert progressively smaller, more uniformly distributed nanoparticles.

Biological Approach

The biosynthetic approach has gained popularity as an effortless, more sustainable alternative to conventional chemical routes for synthesizing AgNPs. These methods utilize naturally occurring reducing agents such as plant extracts, microorganisms (including fungi and bacteria), and polysaccharides, in line with green chemistry principles. Furthermore, green synthesis offers versatility in controlling particle shape, morphology, and size distribution. The efficiency and physicochemical properties of nanoparticles synthesized from plant extracts are strongly influenced by several factors, including temperature, reaction time, pH, concentration, and extract composition.

Biomolecules in biological systems play vital roles in reduction reactions and nanoparticle stabilization. Functional groups such as carbonyl, phenolic, amine, and amide groups, along with various bioactive compounds—including pigments, proteins, flavonoids, alkaloids, terpenoids, and other phytochemicals—enable the nucleation of nanoparticles and their subsequent progression [17].

Bacteria-mediated Synthesis

Bacteria have significant potential as biofactories for nanoparticle synthesis due to their ability to produce inorganic materials either through intracellular or extracellular mechanisms. Kalishwaralal et al. demonstrated the preparation of AgNPs from *the culture supernatant of Bacillus licheniformis* [14]. Huq and Akter reported that bacterial strain *Paenarthrobacter nicotinovorans* MAHUQ-43 is also responsible for the biosynthesis of AgNPs. Characterization of nanoparticles and antimicrobial activity against the Gram-positive bacterium Bacillus cereus and the Gram-negative bacterium *Pseudomonas aeruginosa* strains were also discussed [18]. Ghiuta et al. in 2021 reported the biosynthesis of silver and silver chloride nanoparticles using two Gram-negative bacterial strains from the *Enterobacteriaceae* family: *Pantoea and Raoultella.* These bacteria were able to reduce $AgNO_3$ into Ag/AgCl nanoparticles without the use of capping agents [19].

Fungal-mediated Synthesis

Fungi are eukaryotic microorganisms with non-phototrophic characteristics, primarily composed of a firm cell wall composed of glycoproteins and polysaccharides, including chitin and glucans [20]. Upon contact with metal ions (M+) in aqueous solution, fungi can synthesize nanoparticles via both intracellular and extracellular mechanisms. Formation of larger nanoparticles is possible only when extracellular synthesis occurs more rapidly than intracellular synthesis [21]. Extracellular metabolites released by fungi can act as reducing and stabilizing agents during the synthesis of AgNPs. Further, these metabolites contain proteins, enzymes, flavonoids, polysaccharides, and phenolic compounds [22]. These biomolecules help to reduce metal ions such as Ag^+ or Au^+ in the extracellular medium, leading to nanoparticle formation [23]. During intracellular synthesis, the metal precursor is introduced directly into the fungal mycelial culture. Fungal cells initiate nanoparticle formation by adsorbing metal ions onto their surfaces via electrostatic interactions between the positively charged metal ions and the negatively charged lysine residues on the fungal cell membrane. During adsorption, enzymes and metabolites present in the membrane reduce metal ions (M^+) to neutral metal atoms (M^0), thereby promoting nanoparticle aggregation beneath the fungal cell surface [24].

Yeast-mediated Synthesis

Yeasts have become a promising biological source for the production of nanoparticles because of their strength, genetic controllability, scalability, and reputation. Also, yeast-mediated biosynthesis is a sustainable alternative to conventional synthesis. Furthermore, advancements in genetic and metabolic engineering enable the production of uniform-sized, modified, and functionalized nanomaterials. Several applications of yeast-synthesized nanoparticles were examined across various fields, including cancer treatment, imaging, diagnostics, and drug delivery. Many studies have reported the use of environmentally friendly yeast strains, namely, *Saccharomyces cerevisiae*, *Rhodotorula glutinis*, *Geotrichum candidum,* and *Yarrowia lipolytica*, for the extracellular biosynthesis of AgNPs [25].

Algal-mediated Synthesis

Algal-based synthesis is considered a promising technique for the green synthesis of metal nanoparticles. This process offers a sustainable alternative to conventional methods and has potential applications across various industries, including medicine, energy, and environmental remediation. Furthermore, algal-mediated nanoparticles have been found to have many biomedical applications, such as anti-fouling, anticancer, antifungal, and antibacterial activities. Algae, being a rich source of secondary metabolites and owing to their easy cultivation, rapid growth, and scalability, play a significant role in the green synthesis of AgNPs. Depending on the site of nanoparticle formation, synthesis can be either intra- or extracellular. As reported in the literature, the most commonly used algae for nanoparticle synthesis are brown, red, and blue-green algae, as well as microalgae and macroalgae. For example, AgNPs synthesized from the brown alga Gracilaria amansii show potential as anti-fouling coatings for biomedical and environmental applications. AgNPs produced from brown seaweed Padina tetrastromatica efficiently inhibited the growth of Pseudomonas aeruginosa, Bacillus subtilis, and *Klebsiella planticola*. Likewise, AgNPs produced from aqueous extracts of the green marine alga Caulerpa serrulata exhibited strong antimicrobial activity against Shigella spp., Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, and Salmonella Typhi, even at low concentrations [27].

Plant Mediated Synthesis

Among the various available biomaterials, plant biomass or extracts are considered more reliable and nature-friendly alternatives for nanoparticle biosynthesis. They offer various advantages over other biological sources, including wide availability, safe handling, cost-effectiveness, and a diverse array of metabolites that help reduce metal ions. As a result of these benefits, extensive research has been conducted on various plant species to explore their potential for nanoparticle synthesis. Further, this study also emphasizes plant-mediated biosynthesis of AgNPs, covering their characterization, key influencing factors, and applications across multiple fields.

For nanoparticle production, plants can be used either alive or as non-living (inactive) material. Many plants are known to accumulate metals, which they can reduce intracellularly to form nanoparticles. Inactive plant parts are also useful as eco-friendly reducing agents due to the presence of various plant biomolecules. Plant biomass can be used in powdered form or as extracts, typically derived from stems, flowers, leaves, fruits, roots, latex, seeds, and seed coats [26].

During the synthesis of AgNPs, typically silver nitrate ($AgNO_3$) is combined with plant extracts. A flowchart of the plant-mediated synthesis of AgNPs is shown in Figure 1 below. The successful formation of AgNPs is usually signaled by the rapid development of a brownish-yellow color due to surface plasmon resonance, corresponding to the reaction below [26]:

$Ag^{+}NO_3^{-}$ + Plant molecule (OH, C=H, etc.) → Ag^0 nanoparticles

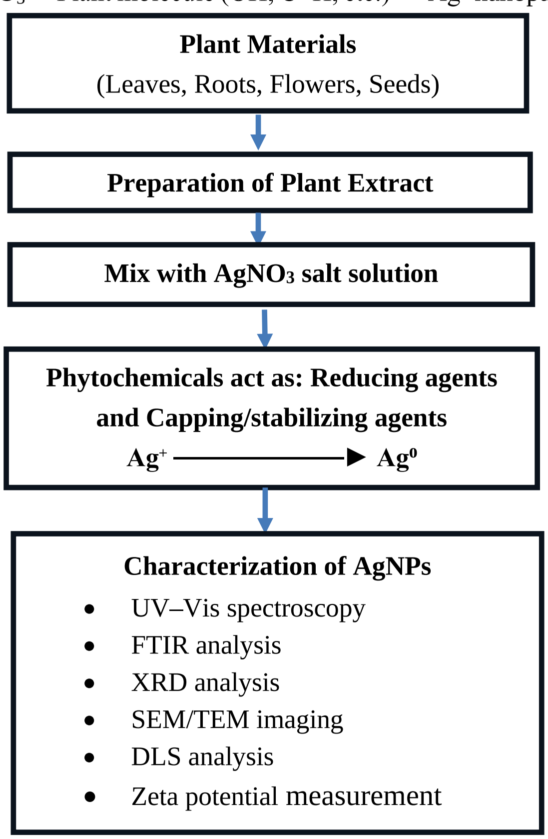

Figure 1: Plant-mediated green synthesis of silver nanoparticles (AgNPs) using plant extracts.

Singh et al. (2025) reported that AgNPs can be produced using neem and turmeric extracts, which act as natural reducing and capping agents. Further, the study focused on characterization and assessment of antimicrobial efficiency of nanoparticles against oral pathogens [28]. In 2023, Ameen et al. explored the effectiveness of AgNPs derived from *the extract of Ipomoea asarifolia (a native African plant)* against cancer and bacterial cells. The antibacterial tests revealed that the synthesized nanoparticles significantly inhibited bacterial growth, with an inhibitory concentration (IC_{50}) of 1 µg/mL [29]. Ansari et al. (2023) reported the preparation of AgNPs using neem leaf extract and explored their effects on tomato seed germination and plant growth under laboratory conditions [30]. Rawat et al. (2020) explored the production of AgNPs using *Polygonatum graminifolium* leaf extract and also tested its antibacterial action against *Escherichia coli* and *Staphylococcus aureus* [31]. Uddin et al. in 2020 established the formation of AgNPs using Cocos nucifera leaf extract as a biological stabilizing and reducing agent, with $AgNO_3$ as the precursor. Their antibacterial studies showed notable zones of inhibition against both Gram-positive (Bacillus subtilis, Staphylococcus aureus) and Gram-negative (Escherichia coli, Salmonella typhimurium, Citrobacter freundii, and Pseudomonas aeruginosa) bacteria, with zones ranging from 10 to 20 mm. The largest zone of inhibition (20 mm) was observed against *C. freundii*, while the lowest (10 mm) was against *Bacillus subtilis*. *Pseudomonas aeruginosa* exhibited the second-largest zone of inhibition at 19 mm. These findings suggest that *C. nucifera* leaf extract-mediated AgNPs hold strong potential as antimicrobial agents [32].

Gul et al. (2021) reported the utilization of *Ricinus communis leaf and root extracts* for the production of AgNPs, and their stability was tested against various conditions. Nanoparticles were found to be stable up to 100 °C and neutral to slightly acidic/basic (pH 5–6) for several months, while unstable below pH 5 [33]. Hashemi et al. (2022) biosynthesized metallic AgNps using *Sambucus ebulus* extract (AgNPs@SEE) and evaluated their anticancer, antibacterial, and photocatalytic activities. They also optimized reaction variables like contact time, temperature, and $AgNO_3$ concentration [34]. Keshari et al. (2020) synthesized AgNPs using *Cestrum nocturnum* leaf extract and evaluated their antibacterial and antioxidant properties. The bacteriostatic and bactericidal activities against *Enterococcus faecalis*, *E. coli*, and *Salmonella Typhi* were examined. Furthermore, the study assessed the antibacterial sensitivity and determined the MIC of the nanoparticles against these pathogens [35].

Microfluidic systems enhance the consistency of nanoparticle (NP) synthesis by enabling precise control of the key physicochemical parameters governing nucleation and particle growth. Unlike conventional batch reactors, which often experience temp. & conc. differences due to poor mixing, microfluidic reactors function in micrometer-scale channels with high surface-to-volume ratios. This design permits quick heat & mass transfer & highly controlled reaction conditions [36].

Flow rate: In microfluidic reactors, the total flow rate (TFR) and flow-rate ratio (FRR) can be adjusted by using syringe or pressure pumps. These factors determine how quickly the reagents mix and directly affect supersaturation, nucleation rate, and nanoparticle growth. Higher flow rates typically reduce mixing times and promote rapid nucleation, leading to smaller, more uniform nanoparticles. Conversely, lower flow rates increase residence time and can allow for more particle growth. Since the flow conditions are controlled and consistent, batch-to-batch variation is greatly minimized [37].

Mixing efficiency: Rapid micromixing is important because nanoparticle formation begins immediately after mixing the precursor solutions. Traditional batch synthesis typically suffers from non-uniform mixing, leading to local concentration gradients that yield particles with broad size distributions. In contrast, microfluidic devices use passive (e.g., serpentine or staggered herringbone channels) or active (e.g., acoustic- or electric-field-assisted) mixers to achieve rapid, homogeneous mixing. Uniform mixing results in simultaneous nucleation in the entire reactor, leading to nanoparticles with narrow size distributions and improved monodispersity [37].

Residence time: The time the reactants spend inside the microreactor. Microfluidic channels are characterized by well-defined geometries and continuous flow, ensuring that all reacting species have nearly the same residence time. This reduces the variation in crystal growth and particle maturation typically observed in batch reactors, yielding nanoparticles with uniform size, morphology, and crystallinity. Droplet-based microfluidic systems minimize residence-time dispersion by enclosing reactions in separate droplets acting as individual microreactors [37].

Temperature control: Microfluidic channels have microscale dimensions, which lead to excellent heat transfer and rapid thermal equilibration. The uniform temperature in the reactor prevents thermal gradients that can change the reaction kinetics, nucleation rates, and crystal growth. For green synthesis using biological extracts, it is especially important to maintain a constant reaction temperature, as the biomolecules responsible for reduction & stabilization may be temperature-sensitive [38]. In microfluidic systems, controlled pumping is used to deliver precursor solutions and biological extracts at defined concentrations. Precise control of reagent concentration and flow-rate ratio maintains supersaturation during nanoparticle formation, reducing fluctuations in nucleation and growth. This is especially useful in green synthesis where there is inherent batch-to-batch variability in the phytochemical composition of biological extracts. Microfluidics does not eliminate inherent differences in extract composition. Still, it can minimize process-induced variability by providing identical reaction conditions for each synthesis run, thereby improving reproducibility and product consistency [37].

Overall, microfluidic systems provide a highly reproducible reaction environment by precisely controlling flow rate, mixing efficiency, residence time, temperature, and reagent concentration on a continuous-flow platform, thereby enabling uniform nucleation and controlled particle growth. Thus, they produce nanoparticles with narrower size distributions, better monodispersity, greater batch-to-batch reproducibility & greater scalability than conventional batch synthesis methods [39].

Another promising engineering approach to increase the reproducibility and scalability of green nanoparticles is the use of automated bioreactors. A significant limitation of biologically mediated synthesis is batch-to-batch variability due to fluctuations in metabolite production, which are related to variations in breeding conditions, nutrient availability, environmental stress, and growth stage in plant, algal, fungal, or bacterial extracts. Automated bioreactors incorporate real-time sensors and feedback control systems to monitor and control key process parameters, including pH, temperature, dissolved oxygen (DO), agitation rate, and nutrient supply.

Maintaining these parameters within defined ranges ensures steady microbial or plant cell growth and stable biosynthesis of reducing and capping biomolecules, thereby minimizing variations in extract composition. Further, automated feeding strategies and process analytical technologies (PAT) enable continuous optimization of metabolite production, while minimizing operator-dependent errors. The resulting biological extracts exhibit greater compositional uniformity, leading to more reproducible nanoparticle nucleation, controlled growth kinetics, and improved consistency in particle size, morphology, and yield. Furthermore, automated bioreactors facilitate scale-up through controlled and reproducible upstream processing, making them well suited for industrial-scale production of green-synthesized nanoparticles while maintaining product quality and batch-to-batch consistency [40].

CHARACTERIZATION OF SILVER NANOPARTICLES

Characterization of nanoparticles is one of the most vital steps for obtaining complete information about their chemical, structural, and functional properties. Various spectroscopic and microscopic methods are employed to analyze surface chemistry, surface area, morphology, particle size, and compositional consistency, etc.

Among these technologies, UV-Vis spectroscopy is used to determine the metal-like nature of nanoparticles. It helps identify the surface plasmon resonance (SPR)—a phenomenon in which AgNPs typically exhibit a distinct absorption peak between 400 and 500 nm, serving as a reliable indicator of their presence [41]. Fourier transform infrared spectrophotometry (FTIR) is a key method for determining the functional groups present in nanoparticles. It is used to identify biomolecules or capping agents involved in the process, to understand their interactions with nanoparticles, and to analyze surface chemistry to determine how molecules are attached to or bound to the nanoparticle surface [42].

X-ray Diffraction (XRD) analysis is used to determine the crystalline structure of metallic nanoparticles. For AgNPs, XRD peaks are observed at specific 2θ values corresponding to crystal planes such as (122), (111), (200), (220), and (311), thereby confirming the crystalline nature & identity of the particles [43].

Scanning Electron Microscopy (SEM) is widely used to characterize the structural features of nanoparticles, particularly their surface morphology. Elemental composition can also be obtained by coupling SEM with Energy-Dispersive X-ray Spectroscopy (EDX). EDX analysis identifies specific elements by identifying characteristic X-ray peaks [44-45].

Transmission Electron Microscopy (TEM) analysis provides high-resolution imaging that helps analyze nanoparticle shape, size, and crystallographic structure. Only TEM permits detailed analysis of the internal structure and morphology of AgNPs [44]. Dynamic light scattering (DLS) is used to study the average size and distribution of particles dispersed in a liquid. This method is especially useful for assessing molecular size distribution, making it an important tool in nanoparticle characterization [45]. The zeta potential analyzer is an essential instrument for measuring the surface charge of nanoparticles. It is also used to evaluate the electrokinetic behavior of synthesized particles [42]. X-ray Photoelectron Spectroscopy (XPS) is a surface-sensitive method to determine the elemental composition and chemical states of elements on the nanoparticle surface. It is one of the major analyses used to evaluate the surface chemistry and uniformity of AgNPs [41].

The Brunauer–Emmett–Teller (BET) analysis has been used to determine the specific surface area, porosity, and physical structure of nanoparticles. Surface area is directly related to properties such as catalytic efficiency, dissolution rate, and moisture retention, making BET analysis highly valuable in material design and performance optimization. Thus, it is essential to use a wide range of complementary characterization methods to determine the various properties and features of AgNPs [41].

FACTORS INFLUENCING BIOSYNTHESIS OF NANOPARTICLES

Effect of pH

Previous research has shown that the size and shape of biosynthesized nanoparticles can be controlled by adjusting the reaction pH. The pH affects the electrical charges of biomolecules, influencing their ability to capture, stabilize, and facilitate nanoparticle growth. It promotes the synthesis of nanoparticles with specific shapes within certain pH ranges, enhancing their stability. Velgosova et al. addressed this gap by examining the effects of pH and temperature on the synthesis of AgNPs using Rosmarinus officinalis extract as both the reducing and stabilizing agents. UV-vis spectroscopy and TEM analysis revealed that optimal conditions for producing uniform, stable, and spherical AgNPs were achieved at pH 8, with a narrow size distribution (~17.5 nm).

At extreme pH values (≤3 or ≥13), nanoparticle formation was hindered by aggregation or precipitation, while elevated temperatures mainly accelerated the reaction without altering particle morphology. Long-term stability was maintained at pH 6, 8, and 12, although the SPR spectrum at pH 6 was broader and the band symmetry at pH 12 was the lowest. The most favorable properties, such as excellent stability, uniform morphology, controlled size distribution, and the highest yield, were consistently obtained for AgNPs synthesized at a precursor pH of 8 [46-47]. Additionally, pH has been shown to impact the size of AgNPs. Fernando et al. examined how pH influences the stability, aggregation, and dissolution of preformed AgNPs, finding that higher pH reduces aggregation, decreases particle size, and enhances colloidal stability, indicating that pH exerts a stronger effect than other environmental factors, such as ionic concentration. [48].Miranda et al. studied the green synthesis of AgNPs using *Spinacia oleracea* leaf extract at pH values of 4, 5, and 9. Their results showed that nanoparticles synthesized under more alkaline conditions were more stable and exhibited stronger, longer-lasting antimicrobial activity [49]. Under alkaline conditions, the presence of negatively charged functional groups enhances the binding of cationic Ag(I) ions, thereby promoting nanoparticle formation. In contrast, acidic environments favor nucleation-driven growth, resulting in larger nanoparticles.

Effect of Temperature

Temperature is an important parameter in the green synthesis of AgNPs, and its effect on the process has been widely studied. Temperature not only influences particle size, morphology, stability, and yield but also affects the reduction rate of $Ag+$ ions to AgNPs. Many studies have highlighted the significant role of temperature regulation in influencing the nanoparticle size and shape from plant-mediated synthesis. For example, Luckman et al. [50] found that substances released from Medicago sativa seed produced silver nanotriangles only at temperatures above 30°C, as lower temperatures suppressed the form-directing agents.

Effect of Reaction Time

The reaction time is another important parameter in the synthesis of AgNPs and significantly affects nanoparticle properties. The complete duration of the reduction process determines the AgNP dimensions and morphology. The longer the duration, the larger the particles due to high growth rates and nucleation. While shorter durations favor smaller sizes, they often result in incomplete reduction, lower yield, or incomplete formation of AgNPs. Optical properties, stability, and yield of nanoparticles are also affected by the reaction time. Singhal et al. reported the production of AgNPs using Tulsi leaf extract in 8 min [51]. Haridas et al. reported the same nanoparticle formation within 10 min using *Coffea arabica* leaf extract [52].

Plant extract/biomass dosage

The concentration of plant biomass or extract frequently governs the effectiveness of nanoparticle synthesis. Many studies have also observed that using more starting material boosts nanoparticle production and affects their shape. Therefore, identifying the optimal biomass concentration is crucial for the synthesis process [49].

APPLICATIONS OF SILVER NANOPARTICLES

Nanotechnology promotes growth and sustained competitiveness in a variety of industrial uses. The special features of nanoparticles have significant applications in industries such as biotechnology, materials science, energy, and medicine [53-55]. Nanotechnology finds widespread application in biomedical industries such as orthopedics/implants, drug delivery, labeling, MRI contrast agents, bio-detection, thermal spray coatings, and sunscreens [56].

Furthermore, AgNPs have shown potential for environmental disinfection, effectively removing harmful microorganisms from air and water. AgNPs also help degrade hazardous organic pollutants, contributing to environmental cleanup. Their high surface reactivity and sensitivity make them valuable in environmental detection systems, where they can identify trace contaminants. Moreover, AgNPs' distinct optical and electrical properties are involved in energy conversion technologies, such as photovoltaic cells and fuel cells [57].

More studies have been carried out to enhance the development of well-defined nanoparticles using efficient environmental sensors and catalysts. Figure 2 shows the various applications of plant-mediated AgNPs, whereas Table 2 highlights the comparison between biomedical and environmental pathways and applications of synthesized AgNPs.

Antimicrobial/ Drug delivery

AgNPs consistently exhibit attractive physicochemical properties that make them valuable in biological and medical research. As a strong inhibitor, silver exhibits broad-spectrum bactericidal activity [66]. For ages, silver has been used to cure and prevent a wide range of illnesses, especially infections [67]. AgNPs are also known for their antiviral, anti-inflammatory, antifungal, and antiplatelet characteristics [68].

Various reports state that nanoparticles of silver (Ag^{o}) are generally less dangerous than silver ions (Ag^{+}) [69]. AgNPs are widely used in sectors such as medicine, packaging, agriculture, cosmetics, healthcare & the military due to their antibacterial properties [70].

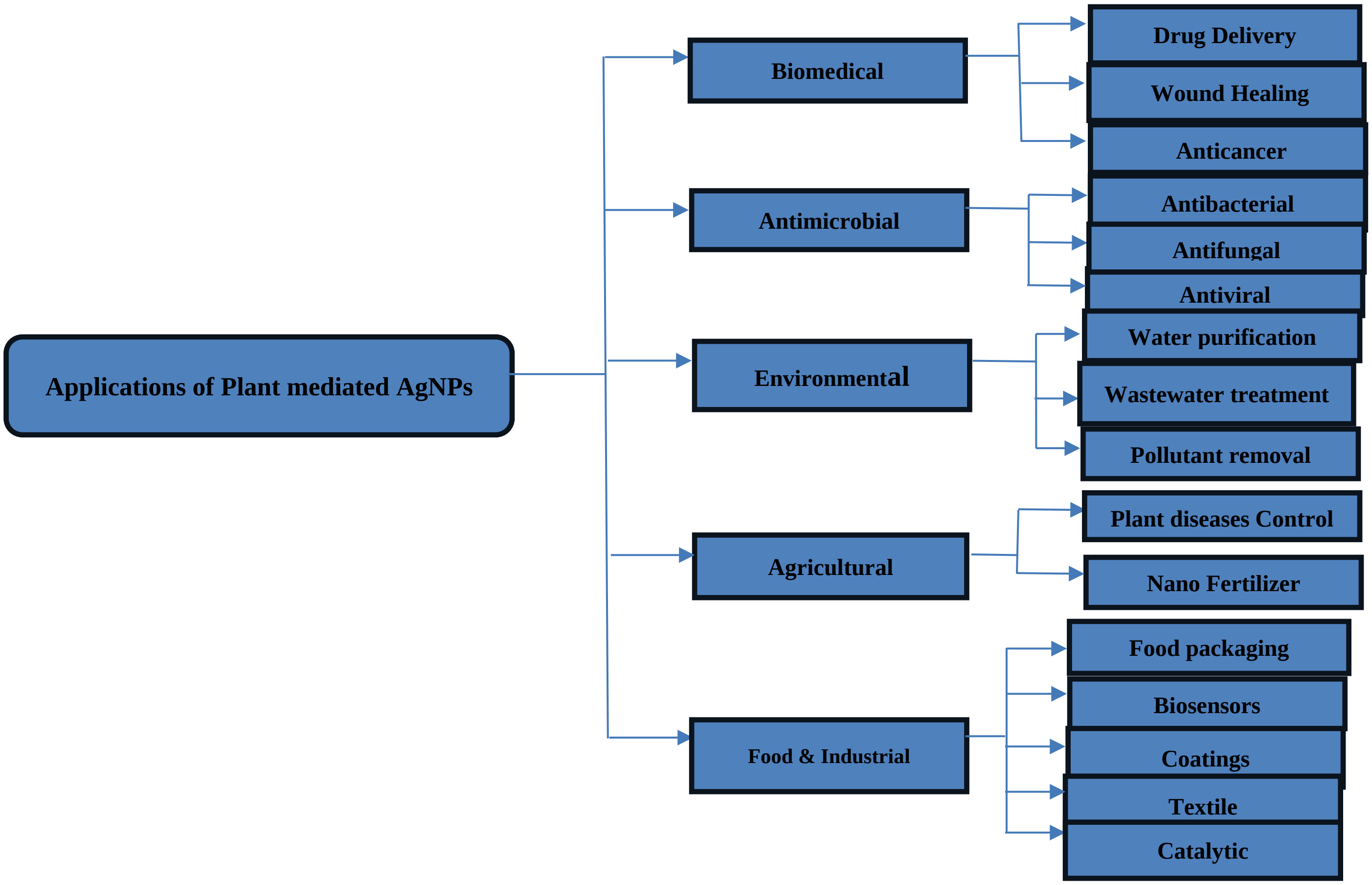

Figure 2: Applications of plant-mediated AgNPs

Table 2: Comparison between biomedical and environmental pathways and applications of synthesized AgNPs

Area	Applications	Description	Pathways	Ref
Biomedical	**Antibacterial Coatings**	Used on surgical instruments and implants for bacterial inhibition	AgNPs disrupt bacterial cell membranes, release silver ions, generate oxidative stress	[58]
	Wound Dressings	Promotes healing and prevents infection	AgNPs prevent bacterial growth on wound surfaces and reduce inflammation	[59]
	Drug Delivery Systems	Enables targeted and controlled release of therapeutics	AgNPs are functionalized to release drugs at specific sites in the body, enhancing bioavailability	[60]
	Cancer Therapy	Enhances photothermal and cytotoxic effects in cancer treatments	AgNPs absorb light (in photothermal therapy) and generate localized heat, killing cancer cells	[61]
	Biosensors	Detect biomolecules like glucose or pathogens.	AgNP surface modifications enhance sensor sensitivity by amplifying detection signals.	[62]
Environmental	**Water & Air Disinfection**	Inactivates bacteria, viruses, and fungi in treatment systems	AgNPs discharge Ag^{+} ions, which will break down microbial cell walls, deactivating pathogens	[58]
	Pollutant Degradation	Catalyzes breakdown of organic pollutants like dyes and pesticides	AgNPs act as catalysts, breaking down complex organic molecules into less harmful substances	[63]
	Environmental Sensors	Detects heavy metals or toxic chemicals with high sensitivity	AgNPs' large surface area interacts with contaminants, facilitating detection of harmful agents	[64]
	Antifouling Agents	Prevents biofilm formation on surfaces like marine equipment or pipelines	AgNPs prevent microbial attachment and biofilm formation through their antimicrobial action	[59]
	Energy Conversion	Improves performance of photovoltaic and fuel cell systems	AgNPs enhance charge transfer and electron mobility in energy devices	[65]

Antimicrobial/ Drug delivery

AgNPs consistently exhibit attractive physicochemical properties that make them valuable in biological and medical research. As a strong inhibitor, silver exhibits broad-spectrum bactericidal activity [66]. For ages, silver has been used to cure and prevent a wide range of illnesses, especially infections [67]. AgNPs are also known for their antiviral, anti-inflammatory, antifungal, and antiplatelet characteristics [68]. Various reports state that nanoparticles of silver (Ag^{o}) are generally less dangerous than silver ions (Ag^{+}) [69]. AgNPs are widely used in sectors such as medicine, packaging, agriculture, cosmetics, healthcare & the military due to their antibacterial properties [70]. The potent antibacterial properties of AgNPs have been well studied due to their high surface-to-volume ratio [65].

There is ample evidence of their harmful, toxic effects on a broad range of bacteria. Because of their well-established bactericidal and inhibitory properties, AgNPs are frequently used as antibacterial agents. Studies have shown that, upon attaching to the cell surface, nanoparticles can affect respiratory processes and membrane permeability. Furthermore, AgNPs can extensively infiltrate bacterial cells and adhere to the membrane surface. Compared with Gram-positive bacteria, AgNPs have demonstrated a comparatively stronger antimicrobial effect against Gram-negative bacteria. This increased activity is most likely due to the thin peptidoglycan layer and the presence of porins, beta-barrel proteins in Gram-negative bacteria [72]. Moreover, the surface energy of nanoparticles increases with their surface area, thereby improving their biological efficacy [73]. Various methods have been employed to produce AgNPs in various shapes, including rod-shaped, spherical, triangular, and truncated nanoplates. The most effective antibacterial activity of these has been verified for truncated triangular silver nanoplates [72].

The substantial increase in multidrug-resistant (MDR) pathogenic bacteria presents a serious global health challenge. The Centers for Disease Control and Prevention (CDC) warned that fatalities caused by bacterial infections may soon exceed those from cancer. In response to this growing threat, nanomaterials—particularly AgNPs with diameters of 10 nm or less—have been considered promising substitutes for traditional antibiotics. Their superior antibacterial activity is largely due to their high surface area-to-volume ratio, which facilitates more interactions with bacterial membranes. This allows AgNPs to disrupt cell walls, penetrate cells, and accumulate intracellularly. The smaller size of nanoparticles enhances their capacity to release silver ions (Ag^{+}) and generate reactive oxygen species (ROS), including superoxide anions, hydroxyl radicals, and hydrogen peroxide. These ROS contribute to protein damage and oxidative stress, leading to lipid peroxidation and DNA degradation in microbial cells. In addition to contributing to microbial cell damage, AgNPs were found to generate 2-3 times as many reactive oxygen species (ROS) as controls [74]. In addition, Ag^{+} ions can bind to –SH (thiol) groups in bacterial enzymes and membrane proteins, thereby hindering crucial functions such as respiration and nutrient transport [75]. These mechanisms of membrane penetration depend on particle size, morphology, and surface charge, which may differ between Gram-positive and Gram-negative bacteria owing to their distinct membrane structures. Gram-negative bacteria tend to be more susceptible due to their thinner peptidoglycan layer and negatively charged outer membrane, which facilitate Ag^{+} uptake and membrane destabilization. In contrast, Gram-positive bacteria, with their thicker peptidoglycan walls, often show relatively greater resistance. Despite their effectiveness, AgNPs smaller than 10 nm may pose cytotoxic risks to mammalian cells. Therefore, proper control of the parameters used to produce nanoparticles is necessary to balance strong antimicrobial action with biocompatibility in biomedical applications [76].

Agnihotri et al. studied the antibacterial properties of chemically produced AgNPs with maximum yield and monodispersity, along with average sizes of 5, 7, 10, 15, 20, 30, 50, 63, 85, and 100 nm. The antibacterial efficacy was evaluated by determining the MIC and MBC of these AgNPs against four bacterial strains: *E. coli* MTCC 739, *E. coli* MTCC 443, *Bacillus subtilis* MTCC 441, and *S. aureus* NCIM 5021. Among these, *E. coli* MTCC 443 was more sensitive, while *S. aureus* NCIM 5021 was the least sensitive irrespective of nanoparticle size [77]. The study found that AgNPs smaller than 10 nm displayed significantly improved antibacterial activity, as evidenced by delayed bacterial growth and reduced MIC/MBC values. Among these, 5 nm AgNPs exhibited the most potent antibacterial effect and the fastest bactericidal action against all tested strains compared with 7 nm and 10 nm particles.

Furthermore, the conventional well diffusion method was used to assess the antibacterial efficacy of the synthesized AgNPs against human-pathogenic bacteria, including *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Escherichia coli*, and *Bacillus subtilis* [56]. Determining the zones of inhibition of synthesized nanoparticles against various conventional antibiotics will facilitate a comparative assessment of antibacterial efficacy [78]. A wider zone of microbial inhibition means a stronger antibacterial activity of the investigated drug [79]. AgNPs in the 1–10 nm range are attached to cell membrane surfaces and interfere with processes such as respiration and membrane permeability [80]. The study reveals that AgNPs may limit cell membrane permeability, hinder respiratory activities, and stimulate the generation of free radicals, though the exact mechanism remains unclear. AgNPs were further examined by using fluorescent bacteria. Based on the findings, bacteria die when AgNPs attach to S-containing components of their cells. Measurements of the cell-free supernatant's fluorescence revealed how silver affected bacterial recombination [81]. However, nanoparticles produced from plant extracts exhibit greater antimicrobial activity against a wide range of microorganisms, including fungi, viruses, and bacteria, owing to their abundance of reducing and stabilizing agents. For example, carbohydrates, proteins, alkaloids, flavonoids, terpenoids, and phenols are major reducing agents commonly found in extracts. Some functional groups, such as carbonyl, hydroxyl, or amine groups, act as capping agents during the formation of AgNPs and can be identified through several spectroscopic techniques. Therefore, a plant-based approach has been widely adopted by researchers owing to its accessibility and greater reproducibility related to physical and chemical methods [82]. Antibacterial studies of AgNPs biosynthesized from plant extracts are summarized in Table 3. The table shows that different parts of the plant contain various compounds that may be effective against different types of bacteria.

Table 3: AgNPs biosynthesized from different plant extracts and their targeted bacteria

Sl. No.	Plant name	Targeted Bacteria	Ref.
1	*Tinospora cordifolia* (Stem)	*Pseudomonas aeruginosa*	[83]
2	*Syzygium cumini* (Bark)	*Bacillus licheniformis*	[84]
3	*Cucumis sativus*	*Mycobacterium tuberculosis*	[85]
4	*Juglans regia* (Bark)	*Streptococcus mutans*	[86]
5	*Calotropis procera* (fruit and leaves)	*Vibrio cholerae*	[87]
6	*Cocos nucifera* (Extracted from oil cake)	*Acinetobacter* sp., *Aeromonas* sp. and *Citrobacter* sp.	[88]
7	*Acalypha indica leaf extracts*	*Vibrio cholerae and Escherichia coli*	[89]
8	*Cola nitida* pod extract	*Pseudomonas aeruginosa, E. coli, and Klebsiella granulomatis*	[90]
9	*Annona squamosa* (leaf)	*Escherichia coli*	[91]
10	*Taraxacum officinale* (leaf)	*Pseudomonas syringae and Xanthomonas axonopodis*	[92]
11	*Rosa indica* leaves	*Bacillus subtilis and Pseudomonas aeruginosa*	[93]
12	Pomegranate juice extract	*Escherichia coli and Staphylococcus aureus*	[94]
13	Dried grass	*Acinetobacter baumannii and Pseudomonas aeruginosa*	[95]
14	*Phyllanthus emblica*	*Acidovorax oryzae strain RS-2*	[96]
15	*Phoenix dactylifera*	*Escherichia coli*	[97]
16	*Psidium guajava* (leaf)	*Pseudomonas aeruginosa*	[98]
17	*Rheum palmatum* root extract	*Pseudomonas aeruginosa and Staphylococcus aureus*	[99]
18	*Prunus japonica* leaves extract	*Proteus vulgaris*	[100]
19	Olive leaf extracts	*Pseudomonas aeruginosa, E. coli, and Staphylococcus aureus*	[101]
20	*Boerhaavia diffusa*	*Flavobacterium branchiophilum*	[102]
21	Extract of banana peel	*Escherichia coli and Pseudomonas aeruginosa*	[103]
22	*Citrus Sinensis* peel extract	*Escherichia coli and Staphylococcus aureus*	[104]

In addition, AgNPs are gaining attention in drug delivery systems as a result of their ability to enable controlled, targeted drug release. Further, they can offer a feasible alternative to traditional cancer treatments by reducing side effects, overcoming drug resistance, and improving drug specificity. Their small size facilitates crossing of biological barriers, making them suitable carriers for therapeutic agents such as antisense oligonucleotides and small molecules. Additionally, their unique photophysical properties enable precise delivery, controlled release, and real-time tracking through imaging, highlighting their potential in advanced nanomedicine applications [104].

Imaging: Light is scattered by metal nanoparticles, which have both scattering and absorption components in their extinction spectra [105]. The interaction between these nanoparticles and the oscillating electromagnetic field causes the conduction electrons in the metal to oscillate collectively and coherently relative to the nanoparticles' lattices. Localized Surface Plasmon Resonance is the frequency at which this interaction takes place. Compared to existing medicines, nanoparticles have advantages, especially in imaging applications, which have grown in popularity over the last 20 years [106].

Because of their strong and distinct plasmonic resonance properties, silver nanoparticles are an essential component in imaging systems, including scattering, colorimetric, SERS (Surface-Enhanced Raman Spectroscopy), and MEF (Metal-Enhanced Fluorescence) [107]. Nowadays, silver-based biosensors are an effective means of identifying cytochrome P53 in squamous cell carcinoma of the head and neck. Furthermore, AgNPs are useful for detecting sulfide and heavy metal ions, including nickel, cobalt, and mercury, due to their colorimetric sensing capabilities. Since they exhibit greater anisotropy and a more pronounced lightning-rod effect than other forms of AgNPs, triangle-shaped AgNPs are particularly useful for creating plasmonic sensors or detectors to detect mercury ions in solution [108].

Biosensors/nanosensors

AgNPs are extensively used in the development of biosensors and nanosensors due to their unique optical, electronic, and catalytic properties. The ability of AgNPs to enhance signal detection, improve sensitivity, and enable rapid responses makes them suitable for various sensing applications, including detecting biomolecules, pathogens, environmental pollutants, and disease biomarkers. Ding and Wang (2013) reported various nanoparticle-based biosensors, including AgNPs, focusing on their application in SERS and biomolecule detection.

Furthermore, AgNPs boost Raman signals, thereby making them suitable for biosensing applications [109]. SERS-based biosensors utilize the plasmonic properties of AgNPs to detect low concentrations of biomolecules, including proteins, DNA, and small molecules. In 2014, Jin et al. demonstrated the use of AgNPs for colorimetric detection of bacterial pathogens, highlighting their *potential for rapid* diagnostic applications [110]. A colorimetric sensor using AgNPs can detect bacterial DNA or specific cancer biomarkers, offering a simple and fast diagnostic tool. Kumar & Mohammad (2011) discussed the use of nanomaterials, including AgNPs, for biosensing applications, with a focus on DNA and protein detection [111].

Implants/Medical Devices

The process of wound healing is intricate and multi-step, requiring the cooperation of various tissues and cell types [112]. One of the first commercial wound dressings, Acticoat, which contains double-layer membranes of polyamide ester coated with AgNPs, demonstrates the utility of AgNPs in wound curative applications [113]. It is thought that these nanoparticles will accelerate healing and yield better cosmetic results. They may help keratinocytes proliferate and migrate, inhibit collagen production by fibroblasts, and alter cytokine levels [114]. Furthermore, it has been confirmed that *Aspergillus niger-produced* AgNPs promote wound healing in an excision wound rat model by modulating cytokines involved in the process [115]. Furthermore, AgNPs have antiviral, fungicidal [116], antibacterial [117], anticancer [118], antifungal, and anti-inflammatory properties [119], in addition to healing.

Also, dressings that contain AgNPs can protect wound sites from bacterial infection [120]. Silver and AgNPs are added to ointments for medical applications to help treat burns and open-wound infections [121]. Furthermore, biologically produced AgNPs exhibit broad biocidal effects by rupturing bacterial cell membranes and disrupting protein synthesis, thereby impairing enzymatic activity [55]. Silver-containing topical dressings are widely used to treat infections in open wounds, burns, and persistent ulcers. Since diabetic wounds frequently experience several secondary infections, AgNPs and Ag^+ carriers may be especially helpful for delayed diabetic wound healing. These nanoparticles can help people with diabetes heal their wounds more quickly and with less scarring. Silver nitrate remains a common antibacterial agent for treating chronic wounds [122]. Therefore, AgNPs offer a unique therapeutic approach to clinical wound treatment.

Owing to extensive research on the antibacterial properties of AgNPs, novel silver-impregnated catheters are now available for clinical use. Both central venous catheters and urinary Foley catheters have been made with silver. Two medical equipment manufacturers in the UK sell Foley catheters coated with silver. The Bardex IC catheter is coated with a hydrogel layer made by Bard and a silver alloy layer that combines silver with other metals such as gold and palladium. Meanwhile, Tyco Healthcare (now Covidian) unveiled the Dover Silver Foley catheter, which features a silver hydrogel coating. Both the interior and exterior of these catheter types are coated [123].

These catheters furthermore include silver ions bound to an inert ceramic zeolite. To prevent catheter-associated infections, two commercially available medical catheters with AgNPs are the Silverline (by Spiegelberg GmbH and Co. KG, Hamburg, Germany) and the ON-Q Silver SoakerTM (by I-Flow Corporation, CA, USA) [124].

Bone Cement/ Dental Materials

Based on a study, methicillin-resistant *Staphylococcus aureus* (MRSA), methicillin-resistant *Staphylococcus epidermidis* (MRSE), and *Staphylococcus epidermidis* showed no growth in bone cement composed of poly(methyl methacrylate) (PMMA) containing 1% AgNPs. Quantitative and qualitative cytotoxicity testing showed no apparent difference between the non-toxic control group and nanosilver-coated bone cement [125]. Therefore, AgNPs become a useful substitute for antibiotics in orthopedic applications [126].

A comparative investigation between bone cement loaded with nanosilver, cement loaded with gentamicin, and plain cement was carried out in vitro by Alt *et al.* [125]. Each type of cement was evaluated against MRSA, MRSE, and *S. epidermidis*. The cement infused with nanosilver exhibited exceptional antibacterial activity against all bacterial strains. Prokopovich *et al.* investigated the application of tiopronin, a thiol molecule, as a stabilizing component for nanosilver incorporated into bone cement in a separate study [127]. The combination of tiopronin and nanosilver exhibited potent antibacterial activity without impairing the cement's mechanical properties or causing cytotoxicity. Tiopronin further enhanced the stability of nanosilver.

Applications for AgNPs in dental materials are extensive. Studies have shown that incorporating varying concentrations of AgNPs into composite resin yields notable antibacterial activity along with good mechanical properties [128]. Yamamoto *et al.* have shown that a resin composite containing fillers injected with silver ions effectively inhibits oral streptococci [129]. Furthermore, NSPs in endodontic filling materials have demonstrated significantly enhanced antibacterial action against *S. aureus, Enterococcus faecalis* & *Streptococcus milleri* [130].

Other Applications of AgNPs

The wide range of applications of nanomaterials is due to their unique chemical and physical properties. These include scratch-resistant coatings, structural ceramics, fuel cells, propellants, photocatalysts, membranes, and solar cells [56]. Nanostructured metals are very effective as catalytic media due to their high surface energy and surface area. Studies have indicated that stable colloidal particles have less catalytic activity than smaller, actively growing silver particles. It was observed that smaller particles accelerate the reduction of borohydride in various organic dyes compared to larger, more stable silver particles. During reduction of methylene blue dye with $NaBH_4$, it was observed that the catalytic activity of AgNPs produced using Ayurvedic medicines, namely Kashayam and Guggulutiktham, depended on their size [131]. AgNPs derived from the Gloriosa fantastic plant extract exhibited an electron-relay effect that, after 30 minutes, greatly accelerated methylene blue degradation [132]. As a result, AgNPs are beneficial for catalysis. Compared with bulk forms, nanoparticles can have diverse effects on human health [133]. It has been confirmed that human lung epithelial cells exposed to metal-containing nanoparticles produce reactive oxygen species, causing cellular damage and oxidative stress [134-135]. By tracking individual nanoparticles in embryos at different developmental stages, the harmfulness and biocompatibility of AgNPs were assessed. The types of developmental defects observed in zebrafish were directly correlated with the AgNP dosage applied [136]. Besides, the nanomaterial-based cosmetics industry uses them in skin creams, sunscreen, UV-protection ointments, lipsticks, toothpaste, and other products.

The main uses for AgNPs are in environmental applications, such as power-generation equipment and catalytic converters in cars. In these situations, the nanoparticles act as catalysts that interact with toxic gases such as nitrogen oxides and carbon monoxide, helping reduce environmental pollution caused by the burning of coal and gasoline [56]. Additionally, biological systems and wastewater treatment are of great interest in AgNPs [137]. The main areas in which AgNPs are used in environmental applications are air, water, and surface disinfectants.

According to a WHO evaluation, benign microorganisms, including fungal and bacterial contagions, usually originate in air filters, which are responsible for half of the organic impurity in indoor air. These microbes produce mycotoxins that are exceedingly harmful to humans. However, the issue of biological pollution has been partially addressed by developing air filters incorporating AgNPs. Research has shown that activated carbon filters (ACFs) embedded with AgNPs are highly effective at eliminating bioaerosols.

Effective treatment techniques are desperately needed to combat microbial pollution and aquatic illnesses. The potential of Ag-NPs for water disinfection has attracted considerable interest. By employing a connecting agent, 3-aminopropyltriethoxysilane (APTES), chemically generated nanoparticles can be added to form a nanoparticle-containing porous ceramic composite starting from a porous ceramic material. The sterilizing efficacy of water filters composed of this AgNPs–porous ceramic composite was evaluated. The results showed that at a flow velocity of 0.01 L/min, the output count of *E. coli* was zero, despite the input water having a bacterial concentration of about 105 CFU/mL [138]. Paint that contains AgNPs has been shown to have antibacterial properties that are effective against gram-positive and gram-negative strains of bacteria, such as *S. aureus* and *E. coli*, as well as human diseases. By inhibiting microbial growth, AgNPs can also be used to preserve food ingredients and extend their shelf life. Ag-NP-coated paper may be useful for inhibiting microbial growth and assisting in food preservation. Sonochemical coating, a simple technique that uses ultrasonic radiation to apply a colloidal silver coating to paper, was described by Gottesman and associates [139]. According to their research, AgNP-coated paper has promising applications in food packaging.

TOXICITY OF SILVER NANOPARTICLES

AgNPs exhibit notable antimicrobial, anticancer, and drug-delivery properties, although their potential toxicity is a major concern for biomedical applications. The toxic effects of AgNPs depend on their physicochemical properties and the rate at which silver ions (Ag^+) are released. These factors determine the stability of nanoparticles and their interactions with biological systems.

Many cytotoxicity studies have shown that AgNPs can cause oxidative stress. They generate excessive reactive oxygen species (ROS), leading to lipid peroxidation, protein oxidation, mitochondrial dysfunction, and reduced cellular antioxidant defenses. Increased ROS disrupts the mitochondrial membrane potential, activates apoptosis signals, and ultimately leads to programmed cell death. The level of cytotoxicity depends on concentration and typically increases as particle size decreases, because smaller nanoparticles have a greater surface area and enter cells more readily [140].

Genotoxicity studies have found that AgNPs can induce DNA strand breaks, oxidative DNA damage, chromosomal instability, and impaired DNA repair under certain experimental conditions. While many in vitro studies report strong genotoxic effects, in vivo results show variation. Factors such as route of exposure, dose, exposure duration, particle size, and surface chemistry will affect the results [141].

After inhalation, ingestion, skin absorption, or injection, AgNPs can enter the bloodstream and collect in various organs, including the liver, kidneys, spleen, lungs, brain, and reproductive organs. The liver and spleen often show the highest accumulation due to uptake by the mononuclear phagocyte system. Long-term accumulation may lead to chronic inflammation, tissue damage, impaired organ function, and slow nanoparticle clearance. The gradual release of Ag^+ ions from AgNPs is a major factor in their toxicity since these ions interact with proteins, enzymes, and nucleic acids, disrupting essential cellular processes [142].

SAFETY AND REGULATORY CONSIDERATIONS

To use silver nanoparticles safely in clinical settings, it is essential to evaluate their biological effects through standardized safety tests. Various international regulatory bodies have established guidelines to ensure the quality, effectiveness, and safety of biomedical products containing nanomaterials. The International Organization for Standardization (ISO) provides a framework via ISO 10993 for assessing the biocompatibility of medical devices, covering evaluations of cytotoxicity, sensitization, irritation, systemic toxicity, genotoxicity, implantation, and hemocompatibility [143].

The U.S. Food and Drug Administration (FDA) suggests a case-by-case approach to regulate products with nanomaterials. Likewise, the World Health Organization (WHO) and the Organization for Economic Co-operation and Development (OECD) recommend adapting traditional toxicological protocols to account for the unique properties and behavior of engineered nanoparticles [144].

To enhance the safety of AgNPs, strategies include controlling particle size, optimizing dosage, using biocompatible surface coatings, minimizing uncontrolled release of silver ions, and developing targeted delivery systems to reduce off-target toxicity. Moreover, adhering to internationally recognized regulatory standards and conducting thorough preclinical safety evaluations are vital to ensuring the safe, effective, and responsible development of silver nanoparticle-based biomedical products [145].

FUTURE PERSPECTIVES

The future of plant-based silver nanoparticle (AgNP) synthesis holds significant promise, with key trends and challenges shaping its path forward. While plant-based synthesis is simpler, faster, and more cost-effective than microbial methods, challenges such as particle-size variability, instability, and aggregation must be addressed to enable consistent, large-scale production. The growing preference for green synthesis aligns with global sustainability trends, driving efforts to optimize processes and improve scalability. Advanced characterization techniques, including in situ imaging and computational modeling, will significantly contribute to refining nanoparticle properties and enhancing stability. Additionally, addressing the potential toxicity and long-term safety of AgNPs, especially in biological systems, remains a priority. Upcoming research should emphasize optimizing synthesis methods, ensuring consistency, and exploring the mixture of AgNPs with other nanomaterials for multifunctional applications. Moreover, as their use in medical, environmental, and industrial sectors expands, developing regulatory standards for safe production and use will be essential. Ultimately, by overcoming these challenges, plant-synthesized AgNPs could contribute significantly to sustainable healthcare, safer nanotechnology, and environmental protection.

CONCLUSIONS

In the present study, production of AgNPs using plant-based sources is highlighted as a better option than synthesis from fungi, algae, or bacteria because it is simpler, faster, and more cost-effective. Plants provide a wide variety of natural reducing agents, such as flavonoids and polyphenols, that efficiently convert silver ions into nanoparticles without the complex culture conditions required by microorganisms. Additionally, plant-based synthesis is more environmentally friendly, scalable, and less prone to contamination, making it a safer and more sustainable option for producing AgNPs. These green-synthesized AgNPs have valuable properties, including strong antimicrobial activity, biocompatibility, and high conductivity. They are used in drug delivery, wound healing, cancer therapy, and antimicrobial coatings—helping to address issues such as antimicrobial resistance. Their electrical properties also make them useful in electronics, energy storage devices, and sensors. However, AgNPs can generate ROS and bind to protein thiol groups, potentially causing toxicity and oxidative stress, raising concerns about their safe use in biological systems.

Despite these promising applications, challenges remain in optimizing the biosynthesis process for large-scale production, ensuring consistency, and fully understanding the interactions between AgNPs and biological systems. Limitations include the absence of standardized synthesis methods, which can lead to variability in particle size, shape, and stability, affecting their effectiveness. Additionally, the scalability of green synthesis processes remains a challenge, as achieving large quantities of high-quality AgNPs while maintaining reproducibility is difficult. The long-term safety and potential toxicity of AgNPs in humans and in the atmosphere are also not fully understood and require further investigation. Forthcoming research should prioritize refining the synthesis techniques, improving large-scale production, ensuring consistency, and studying their long-term effects and safety. This approach supports global efforts toward sustainable healthcare, safer nanotechnology, and environmental protection.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTION

Neonjyoti Bordoloi contributed to the conceptualization and design of the review; Manash Pratim Sarma contributed to literature collection, data analysis, and the initial drafting of the article; and Moja Kodak contributed to editing and the original drafting of the manuscript. All authors reviewed and approved the final version of the article.

DECLARATION OF USE OF AI TOOLS

The authors used ChatGPT and Grammarly to check grammar and improve sentence clarity. All content, analysis, and conclusions were reviewed and verified by the authors. The authors take full responsibility for the originality, accuracy, and integrity of the work.

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