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Green Synthesis of Silver Nanoparticles

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Review Article

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ABSTRACT

Development of reliable and eco-accommodating methods for the synthesis of nanoparticles is a vital step in the field of nanotechnology. The remarkable chemical, physical, and biological characteristics of silver nanoparticles make them significant. Nowadays, due to the growing demand for environmentally benign technology for material synthesis, biosynthesis of silver nanoparticles (Ag Nps) has attracted a lot of attention in industrialized nations. Among the various types of nanoparticles and their strategy for synthesis, because of their special physicochemical and biological characteristics, silver nanoparticles created through green synthesis have drawn considerable interest in the biomedical, cellular imaging, cosmetics, drug delivery, food, and agrochemical industries. The size distributions and resultant nanostructure studies from the transmission electron microscopy (TEM) were in perfect agreement with the characteristics of the synthesized colloidal Ag-NPs analyzed by the ultraviolet-visible (UV-vis) spectra. For the creation of silver nanoparticles (AgNPs), biomolecules from various plant parts and microbial species have been investigated. In this review, the available previously released information on AgNPs production, characterisation methods, and applications are compiled and critically analyzed.

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1. INTRODUCTION

Nanotechnology is a new and emerging technology with a wealth of applications. One of the most cutting-edge technologies in many different scientific disciplines, including biology, chemistry, and material science, is nanotechnology [1]. It entails the creation and use of materials with one or more dimensions between the range of 1–100 nm [2]. Given their small sizes, huge surface areas with free dangling bonds, and more reactivity than their bulk cousins, they have surprising and fascinating features [3]. Researchers are becoming more interested in biological processes due to the development of effective green synthesis that uses natural reducing, capping, and stabilizing agents rather than hazardous, expensive, and energy-intensive chemicals [4,5]. Silver has been used for treating many illnesses for ages [6]. “The silver nanoparticles demonstrate better antibacterial” [7], “antifungal and antiviral properties compared with metallic silver and silver compounds” [8,9]. Utilizing biological processes to create nanoparticles is biocompatible because plants, in particular, release functional biomolecules that actively decrease metalions [10].

Both "Top-down" and "Bottom-up" methods can be used to create NPs. “An appropriate bulk material is divided into tiny particles by size reduction using various methods, such as pulse laser ablation, evaporation-condensation, ball milling, etc., in a top-down approach. The bottom-up method allows for the chemical and biological synthesis of NPs through the self-assembly of atoms into new nuclei that develop into nanoscale particles” [11]. In comparison to physical and chemical processes, green synthesis is more environmentally benign, economically advantageous, and readily scaled up for the production of large quantities of nanoparticles (NPs). Green synthesis also does not involve the use of hazardous chemicals, high temperatures, or high energy inputs [12].

Today, extracts from plant parts like fruit, leaves, bark, seeds, and stems have been successfully employed to create nanoparticles. The traditional processes for making NPs are pricey, hazardous, and unfriendly to the environment. Researchers have identified the precise green routes, or the naturally occurring sources and their byproducts, that can be employed for the synthesis of NPs, in order to get around these issues.

2. GREEN SYNTHESIS

The process of creating nanoparticles using microorganisms and plants that have biomedical implications is known as "biosynthesis of nanoparticles." This strategy is economical, safe, biocompatible, green, and safe for the environment [13]. “The two basic ingredients for the ecologically friendly production of AgNPs are a silver metal ion solution and a reducing biological agent. External capping and stabilizing agents are typically not required since reducing agents or other cell components act as stabilizing and capping agents the majority of the time”[14].

2.1 Metal Ion Solution

The main ingredient needed to create AgNPs is the Ag^+ ion, which can be found in a variety of silver salts that are water soluble. However, the aqueous Silver Nitrate solution has been utilized, with Ag^+ ion concentrations ranging from 0.1 to 10 mm.

2.2 Biological Reducing Agents

Silver nanoparticles have been made using biopolymers, microbial cell biomass, or cell free growing media, along with plant extracts. From algae to angiosperms, plants are employed to make AgNps. The manufacture of silver nanoparticles has utilized components like leaves, bark, roots, and stems [14]. The medicinally important plants like Aloe vera [15], *Azadirachta indica* [16], *Cocos nucifera* [17] these were utilized to create silver nanoparticles in a green approach. With the exception of a few instances, all plant extracts served as both potential reducing and stabilizing agents. It was discovered that the plant extracts' metabolites, proteins, and chlorophyll serve as capping agents for synthesized silver nanoparticles [18].

3. MECHANISM OF AgNPs SYNTHESIS

The existence of several organic chemicals, including those that can provide an electron for the reduction of Ag^+ ions to Ag^0, such as carbohydrates, fats, proteins, enzymes & coenzymes, phenols, flavonoids, terpenoids, alkaloids, and gum, is what allows biological matter to produce Ag nanoparticle. Depending on

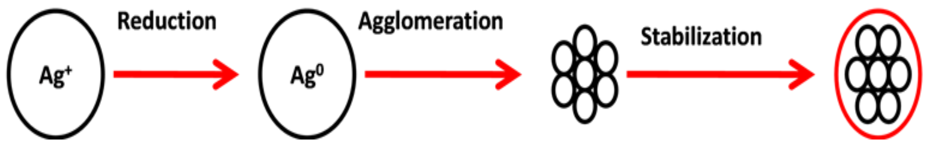

Fig. 1. A schematic diagram showing the silver ion reduction, agglomeration and stabilization to form a particle of nano size

the organism or extract employed, a different active component is responsible for the reduction of Ag^+ ions. The dehydrogenation of acids and alcohols in hydrophytes, keto to enol conversions in mesophytes, or both pathways in xerophytes plants are expected to provide electrons for the nano-transformation of AgNPs. Similar reduction reactions can be carried out by extracellular and cellular oxidoreductase enzymes in microorganisms [19].

4. SEPARATION OF AgNPs

Researchers typically utilize the centrifugation approach to create synthesized silver nanoparticles in pellet or powder form. Additionally, the AgNPs suspensions were dried in the oven to produce the product in powder form [20].

4.1 Cloud-point Extraction

“CPE is simple to perform based on the non-ionic surfactants' solubilization properties and cloud points. This extraction technique, in short, consists of three steps: First, a non-ionic surfactant is added to the sample solution at a concentration greater than its critical micelle concentration (CMC); second, the mixture becomes turbid when the external conditions (such as temperature, pressure, pH, or ionic strength) are changed because it reaches the cloud point (i.e., incomplete solubilization); and third, centrifugation or prolonged standing causes the micelle solution to easily separate into two phases, allowing the analytes to be concentrated and extracted into the surfactant-rich phase due to the analyte-micelle interaction” [21]. Given its numerous advantages, including its low cost, ease of handling, high extraction efficiency, and the preconcentration factor, CPE is the best method for removing contaminants from a variety of environmental and biological samples.

4.2 Field-flow Fractionation

The hydrodynamic separation method known as "field-flow fractionation" (FFF) was created to separate complicated macromolecules, colloids, and particles. It is comparable to a field-driven method and liquid chromatography, with the exception that a stationary phase is not required. In essence, an external field that is applied perpendicular to the axis of the fractionation channel while the flowing stream containing the samples migrates through the FFF channel induces the retention of the analytes. Particles have unique diffusion coefficients due to their diverse physicochemical features and wide size distribution. The retention duration varies because particles remain at varied distances from the accumulation wall to balance this diffusibility and the external field [22].

4.3 Chromatographic Methods

A technique for size-based separation is hydrodynamic chromatography (HDC). Non Porous microparticles are tightly packed in the column, and separation is accomplished by flow velocity and the velocity gradient across them [23].

4.4 Electrophoresis and Capillary Electrophoresis

“Particle size, shape, and surface-chemical alteration of NPs are the key factors that influence electrophoretic separation of NPs. While the electrophoresis of functionalized NPs with surface functional-group modifications is influenced by quantity, chemical groups, and ionization of these functional groups, the electro-charge of NPs without surface modification is primarily from ion adsorption, and the electrophoretic separation greatly depends on particle size” [24].

4.5 Density-gradient Centrifugation

“The density-gradient centrifugation technique, which was used to separate biomacromolecules, has a lot of possibilities for isolating nanoparticles. The development of density-gradient centrifugation has made it possible to employ it with organic solvents, such as nonhydroxylic solvents, which have been used to purify Ag, Au, and CdSe nanoparticles. In the study, cyclohexane and tetrachloromethane mixes with varying densities (by volume 50%, 60%, 70%, 80%, and 90%) were added to create a five-layer gradient. Following the addition of AgNP and centrifugation, distinct coloured zones were seen and are visible under a TEM" [25].

4.6 Miscellaneous Methods

Ag nanoparticles have also been separated using a number of additional methods, such as membrane filtration, ultrafiltration, and dialysis. Centrifugation is a popular method for removing residues from freshly synthesized NPs since it is inexpensive and simple to perform. Membrane filtration, also known as ultrafiltration, has become an effective method for the separation of AgNPs of various sizes due to its straightforward process and lack of the need for additional separating agents. However, during centrifugation or filtering, unwanted aggregation or filter clogging may happen, which could affect the outcomes. Ag^+ and AgNPs may be separated using some methods for determining Ag ions. For the purpose of finding labile metal ions, the diffuse gradients in thin-films method, which is based on Fick's first diffusion law, has generated a lot of interest. In the presence of AgNPs, free Ag ions have been successfully measured by DGT [26,27].

5. CHARACTERIZATION OF AgNPs

AgNPs are frequently characterized using several techniques, such as UV-Vis Spectra, SEM, TEM, FTIR, XRD, and EDAX or EDX/EDS. AgNPs are too small to be detected by conventional optical microscopy due to their nanoscale size. In order to visualize and characterise NMs, many people choose to use electron microscopy (EM) techniques, which are based on the application of an electron beam and have a much greater resolution. Transmission electron microscopy (TEM) and scanning electron microscopy (SEM) are the two techniques that stand out the most. The morphology and status of the particles' aggregation are also revealed by TEM images in addition to the particles' size and form [20]. "The UV-Vis spectral analyses have been used to examine the dependence of pH, metal ion concentration, and extract content on the formation of AgNPs, by showing a red shift in the SPR peak with an increase in nanoparticle size and a blue shift for a decrease in size" [28]. Most studies' SEM morphological analyses showed spherical Ag nanoparticles, however only a handful reported irregular [29], triangular [30], flake [31], flower [32], pentagonal [33] and rod-like structures [34].

6. FACTORS AFFECTING AgNPs SYNTHESIS

The reaction temperature, metal ion concentration, extract content, pH of the reaction mixture, reaction time, and agitation are the main physical and chemical factors that influence the synthesis of AgNP. The size, shape, and morphology of the Ag nanoparticles are substantially influenced by variables such metal ion concentration, extract content, and reaction time [35]. The important parameters in a reaction include the temperature and stirring time. Many researchers employed bio-polymers and plant extracts to synthesize AgNP at temperatures as high as 100℃. The rate of AgNPs production increased as the temperature rose (30–90°C) [36] and also promoted the synthesis of smaller size AgNPs. Most have created AgNPs in a room-temperature (25˚C to 37˚C) range [37].

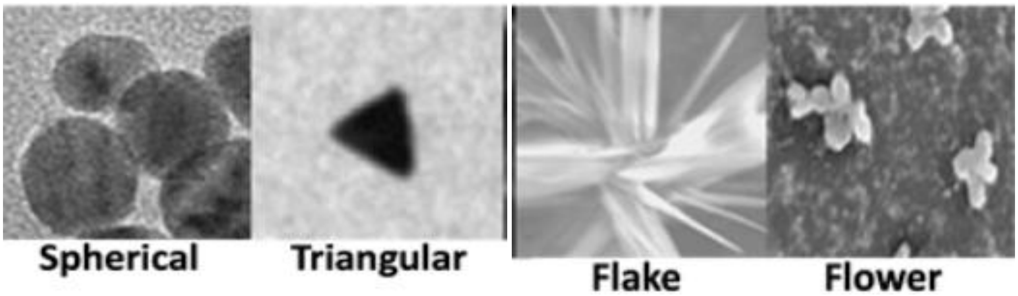

Fig. 2. Various shapes of AgNPs synthesized (from various sources)

7. APPLICATIONS OF AgNPs

"Numerous antibacterial and antifungal applications exist for Ag nanoparticles. Ag nanoparticles have been widely used as antibacterial coatings in medical applications, including heart implants, catheters, wound dressings, orthopedic implants, dental composites, nano-biosensing, and agriculture engineering" [38]. The health sector, food storage, textile coatings, and a number of environmental applications have all made substantial use of Ag nanoparticles as anti-bacterial agents [39]. Ag nanoparticles were used as antibacterial agents in a variety of applications, including water treatment, sanitizing household and medical equipment, and home appliances [40,41]. The Ag nanoparticles exhibited antifungal action against various fungi. Actual mechanism behind the antifungal activity is not fully understood. The antifungal effect has been related to the disruption of the cell membrane's structure by destroying the integrity of the membrane, which inhibits the budding process against C. albicans species. The shape of the Ag nanoparticles has a significant effect on the antimicrobial activity [42,43]. The effectiveness of the Ag nanoparticles as larvicidal agents against dengue vector *Aedes aegypti* [44] *and* malarial vector *A. subpictus* [45]. No attempt has been made to suggest a suitable mechanism for Ag nanoparticles' anti-parasitic effect. Several publications have been presented on the application of Ag nanoparticles in medicine. The Ag nanoparticles have been used as therapeutic agents [38], for disease diagnosis [46], and as nano carriers for drug delivery [47].

Chart 1. Summary of the work related agnps synthesis using green route

S. No.	Author	Reducing agent	Characterization	Particle characteristics	Remarks
1	Kathiraven et al	Marine algae *Caulerpa racemosa* filtered aqueous extract	UV-Vis FTIR TEM XRD	Size—5 - 25 nm Shape—sph, tri. Structure—FCC	Antibacterial effects against *S. aureus* and *P. mirabilis*
2	Rajesh et al	*Ulva fasciata* extract in ethyl acetate	UV-Vis FTIR SEM XRD EDX	Size—28 - 41 nm Shape—sph Structure—cryst Nature—PD	Antibacterial effects against *X. campestrispv malvacearum* pathogen
3	Vivek et al	Aqueous filtrate of *Gelidiella acerosa*	UV-Vis FTIR SEM TEM XRD	Size—~22 nm Shape—sph. Structure—FCC Nature—PD	Antifungal activity against *Mucor inicus* and *Trichoderma reesei*
4	Govindaraju et al	Aqueous filtrate of *Sargassum wightii*	UV-Vis FTIR TEM XRD	Size—8 - 27 nm Shape—sph/variable Structure—cryst	Antibacterial effects against *S. aureus, B. rhizoids, E. coli* and *P. aeruginosa*
5	Kulkarni et al	*Riccia's* ethanol filtrate	UV-Vis SEM EDS	Shape—cub/triang	Antibacterial effects against *p. aeruginosa*
6	Kulkarni et al	Ethanol filtrate of *Anthoceras*	UV-Vis SEM EDS	Size—20 - 50 nm Shape—cub/triang	Gauze cloth with antibacterial effect after inclusion
7	Srivastava et al	Aqueous and ethanol-based *Fissidens*	UV-Vis SEM EDS	Shape—nearly sph	Antibacterial effect against *E. coli, B. cereus,*

S. No.	Author	Reducing agent	Characterization	Particle characteristics	Remarks
		minutes filtrate			*K. pneumoniae, P. aeruginosa*
8	Kulkarni et al	Aqueous filtrate of *Anthoceros*	UV-Vis SEM EDS	Size—20 - 50 nm Shape—cub/triang	Antibacterial effect against *E. coli, B. subtilis, K. pneumoniae, P. aeruginosa*
9	John De Britto et al	Aqueous filtrate of *Pteris argyreae, Pteris confuse* and *Pteris blaurita*	nil	nil	Antibacterial effect against *Shigella boydii, Shigella dysenteriae, S. aureus, Klebsiella vulgaris* and *Salmonalla typhi*
10	Bhor et al	*Nephrolepis sexaltata L.* fern aqueous filtrate	UV-Vis SEM XRD	Size—avg 24.76 nm Shape—sph. Structure—FCC	Antibacterial effect against many human and plant pathogens

8. CONCLUSION

It is determined that over the past ten years, significant efforts have been undertaken to advance green synthesis. Green synthesis advances over chemical and physical approaches because it is affordable, environmentally friendly, and can be scaled up successfully for large-scale synthesis [48]. Ag nanoparticles have a wide range of important pharmacological properties, and they are more affordable than local medications. In green synthesis mediated by plants Cu nanoparticles with an average size of 48 nm and high crystalline properties were produced by the *A. indica* leaf broth and remained stable for two months at 4°C [49,50]. In addition to plant-mediated green synthesis, AgNPs' numerous bioassay capabilities have received particular attention. The form and size of the Ag nanoparticles produced employing biological reducing and capping agents vary greatly. The anti-microbial effect of Ag nanoparticles has been extensively researched among other applications.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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