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Bio-Catalytic Synthesis of Silver Nanoparticles

V. Veeraputhiran*

Research Department of Chemistry, V. O. Chidambaram College, Tuticorin – 8,TamilNadu, India.

***Corres. Author: liviveera@yahoo.co.in**
Mobile: +919944825986

Abstract: Biosynthesis of nanoparticles is under exploration due to wide biomedical applications and research interest in nanotechnology. Currently, bioreduction of silver nitrate ($AgNO_3$) with the plant extract is the leading trend for the synthesis of silver nanoparticles. More and more research inputs are rendered for this and there has been an upsurge of interest in the biological synthesis of nanomaterials by using several plants, plant pure compounds, plant biochemical and microorganisms in the past few years. In the present study, Green synthesis and their major advantages for the production of silver nanomaterials have been reviewed.

Key words: silver nanoparticles, biosynthesis, microorganism, plant extract.

1. Introduction

Nanostructured materials show many aspects of interesting characteristics, i.e., optical, catalytic, that greatly depends on the size and shape of nanoparticles as an effect of quantum confinement of electrons. Metal nanoparticles are extensively used in many electrochemical, electroanalytical and bioelectrochemical applications owing to their extraordinary electrocatalytic activity. Although metal is a poor catalyst in bulk form, nanometer-sized particles can exhibit excellent catalytic activity due to their relative high surface area-to-volume ratio and their interface-dominated properties, which significantly differ from those of the bulk material [1].These distinct features are directly related to particles size and shape [2].

Recently, various applications have been found for metal nanoparticles in areas such as biology, medicine, and health sciences. Silver nanoparticles, in particular, have been used in medical diagnosis [3], drug delivery systems [4], sanitization [5], water treatment [6], and wound healing [7]. The later study suggests that silver nanoparticles act by diminishing inflammation through cytokine modulation [7]. The growing need for nanoparticles makes the design of new synthetic routes specific for each application of utmost importance.

Generally, nanoparticles are prepared by a variety of chemical and physical methods such as chemical reduction [8–11], photochemical reduction [12,13], electrochemical reduction [14,15], heat evaporation [16,17], etc., which are not environmental friendly. Since most routes utilized for the preparation of nanoparticles involve the use of hazardous reactants, much attention has been paid to the so-called green synthesis [18–20]. The latter technique focuses on the production of nanomaterials by employing natural compounds as reducing and capping agents. In this present paper, we reviewed some of reported articles for synthesis of silver nano particles via biosynthesis.

2. Biosynthesis of silver nanomaterials

The discovery of the fact, that nano silver (Ag) can fight against the microbes, manufacturers have incorporated Ag nanoparticles (Ag NPs) into more than 200 consumer products, including clothing and cosmetics. The development of reliable green process for the synthesis of silver nanoparticles is an important aspect of current nanotechnology research.

Microbes and plants are currently used for nanoparticle synthesis. Recently a number of organisms have been used for the synthesis of nanoparticles, noble metals in particular. Synthesis using bio-organisms is compatible with the green chemistry principles: the bio-organism is eco-friendly as are the reducing agent employed and the capping agent of the reaction [21]. The nanoparticles may either be synthesized intracellularly or extracellularly using bacteria, yeast, fungi and plant materials which have been used for various applications. Synthesis of nanoparticles using microorganisms started with the use of *Pseudomonas stutzeri* AG259, isolated from silver mines, which has been shown to produce silver nanoparticles [22,23]. Recently *Bacillus licheniformis* has been studied to produce silver nanocrystals extracellularly [24]. Eukaryotic organisms like fungi have been used for the synthesis of different types of nanoparticles such as *Verticillium sp*. [25], and *Fusarium oxysporum* [26]. This can be attributed to the fact that silver at low concentrations does not enter the fungal cells, but it is adsorbed onto the bacterial surface just as silver tends to adsorb to other surfaces, thus silver ions immobilize dehydrogenation because respiration occurs across the cell membrane in bacteria rather than across the mitochondrial membrane as in eukaryotic cells of fungi [27].

Recently some biomimetics way have also been tried for the development of silver and nanoparticles by several research groups using parthenium leaf, natural rubber, starch, mushroom, *cinnamomum zeylanicum* leaf, *magnolia kobus* and *diopyros kaki* leaf, geranium leaf, alfalfa plants, rose leaf and other bacterial, viral materials [28–38]. Sastry *et al.* reported the biosynthesis of nanoparticles using plant leaf extracts and their potential application. R. rugosa leaves extract is found suitable for the green synthesis AgNPs within 10 min. Average crystal size calculated from Scherrer equation is found 12nm. They studied bioreduction of silver ions by extracts of geranium [34] and neem leaf [39]. Further, synthesis of silver nanoparticles using Aloe vera plant extracts was reported by Chandran *et al.* [40]. Most of the above research on the synthesis of silver nanoparticles utilizing plant extracts employed broths resulting from boiling fresh plant leaves. Whereas, Huang *et al.* demonstrated the prospect of using sun dried, *Cinnamon camphora* leaf for the synthesis of the nano-sized noble metal Ag at ambient conditions, without any additive protecting nanoparticles from aggregating, template-shaping nanoparticles or accelerants like ammonia[41].

The recent reports include the biosynthesis using *Murraya koenigii* leaf [42], Mangosteen leaf [43], *Mangifera indica* leaf [44], Tansy fruit [36], *Camellia sinensis* [45], Mushroom [36], Honey [46] and so on. Silver nanoparticles prepared using biological materials have the properties of a high surface area, smaller in size and high dispersion. It is well known, that silver is an effective antibacterial agent and possesses a strong antibacterial activity against bacteria, viruses and fungi, although the mechanism and the manner of action are still not well known [47].

Very recently green silver nanoparticles have been synthesized using various natural products like green tea (*Camellia sinensis*) [29], starch [20], lemongrass leaves extract [48,49] leguminous shrub (*Sesbania drummondii*) [50], latex of *Jatropha curcas* [51] etc.

Quercetin, an antioxidant compound present in the plant, *M. sativa* is possess the readily oxidized hydroxyl group in the C-ring next to the carboxyl group was proposed that the possible

mechanism of silver nanoparticles synthesis by plant extract [52]. It also been proposed that the *M. piperita* may reduce the silver and gold nanoparticle into metallic nanoparticles. It was reported that the ketone is the major component in *Cymbopogon flexuosus* extract that renders the liquid like characteristics of the spherical gold nanoparticle [48]. The synthesized silver exhibited a strong antibacterial activity against both *E. coli* and *S. aureus*.

Dubey *et al.* used the aqueous leaves extract of the plant *Sorbus aucuparia* leaf extract as reducing agent for the synthesis of silver nanoparticles from their salt solutions [37]. Silver nanoparticles can be prepared in a size range of 10–40nm with less amounts of leaf extract and without adding chemicals and physical steps like centrifugation, sonication, annealing, etc. *S. aucuparia* leave extract has found suitable for the synthesis silver nanoparticles within 15 min. Average crystal size calculated from Scherrer equation was found as 16 nm.

The quantity of leaf extract, metal concentrations and temperature are playing an important role in the rapid formation of small sized nanoparticles. The developed crystals were found to be stable for more than 3 months as sorbate ion in the leave extract most likely encapsulates the nanoparticles and maintain their stability.

Silver nanoparticles were rapidly synthesized by Krishnaraj *et al.* using leaf extract of *Acalypha indica* and the formation of nanoparticles was observed within 30 min [5]. The average grain size of the silver nanoparticles formed in the bioreduction process was determined using Scherr's formula, and was estimated as 35nm.

Biosynthesis of silver nanoparticles (AgNPs) was achieved by a novel, simple green chemistry procedure using *citrus sinensis* peel extract as a reducing and a capping agent [53]. The size of the particles was found to be 35 and 10nm at 25ºC and 60ºC, respectively as deduced from TEM measurement. An eco-friendly process for rapid synthesis of silver nanoparticles has been reported using aqueous seed extract of *Jatropha curcas* [51]. Formation of stable silver nanoparticles at different concentration of $AgNO_3$ gives mostly spherical particles with diameter ranging from 15 to 50 nm. In the pursuit of making the nanoscale-research greener, the utilization of the reductive potency of a common byproduct of food processing industry i.e. orange peel (*Citrus sinensis*) has been reported to prepare biopolymer templated "green" silver nanoparticles. TEM imaging showed well-dispersed spherical particles of 3–12nmsize. It was also interesting to note that the highest fraction of particles had a diameter of 6 nm.

Dwivedi *et al.* reported a facile and rapid biosynthesis of silver nanoparticles from *Chenopodium album*, an obnoxious weed [54]. *Chenopodium album* leaf extract was prepared and successfully used for the single-pot biosynthesis of SNPs and GNPs in the size range of 10–30 nm. Only spherical particles were observed at higher leaf extract concentration, as infer from the TEM imaging.

Sathishkumar et al used the bark extract and powder of novel *Cinnamon zeylanicum* for the synthesis of silver (Ag) nanoparticles from silver precursor [55]. Water-soluble organics present in the plant materials were mainly responsible for the reduction of silver ions to nano-sized Ag particles. The average size of the nanoparticles ranged between 31 and 40nm spherical. Also Vidhu *et al.* reported the green synthesis of silver nanoparticles using aqueous seed extract of *Macrotyloma uniflorum* [56]. The effect of experimental parameters such as amount of extract, temperature and pH on the formation of silver nanoparticles was studied. A well-dispersed silver nanoparticle with anisotropic morphology having the size of <12 nm was observed.

A simple one-pot synthesis based on the bioreduction ability of the *P. betle* leaf broth has been developed to produce Ag-protein (core–shell) NPs with spherical shapes in high yield 8 – 30 nm [57]. Authors used culture supernatant of *Pseudomonas aeruginosa* strain BS-161R for the simple and cost effective green synthesis of silver nanoparticles. The culture supernatant of *Pseudomonas aeruginosa* isolate BS- 161R was used for green synthesis of silver nanoparticles with silver nitrate at room temperature. The silver nanoparticles formed were spherical in shape with an average particle size of 13 nm. Motazer *et al.* introduces a new green method synthesis of silver nanoparticles on the cotton fabric surface through using Tollens' reagent [58].

Prathna *et al.* studied the growth kinetics of silver nanoparticles as synthesized on reduction of silver nitrate solution by aqueous extract of *Azadirachta indica* leaves was investigated [59]. The size of the particles was obtained as 10 – 35 nm. Colloidal silver nanoparticles were synthesized by an easy green method using thermal treatment of aqueous solutions of silver nitrate and natural rubber latex (NRL) extracted from *Hevea brasiliensis*. The silver nanoparticles presented diameters ranging from 2 nm to 100 nm and had spherical shape. These silver nanoparticles have face centered cubic (fcc) crystalline structure.

Matos *et al.* reported a simple method to create silver nanoparticles that consists in the irradiation of a silver nitrate and *Euphorbia milii* latex solution by light from a xenon lamp followed by ultrashort laser pulses [60]. After the laser irradiation the silver nanoparticles sizes are reduced drastically to the range of 10–50 nm.

Table 1. The contribution of some biomaterials for the synthesis of silver nanoparticles

Biomaterial	Particle size (nm)	Particle's shape/ structure
Plant origin		
Tansy fruit	16, 11	Spherical, Triangle
Mentha Piperita (leaf extract)	5–30	Spherical
Sesbania drummondii (seeds)	6–20	Spherical
Coriander	6.75–57.91	
Jatropha curcas (latex)	10–20	Face-centered cubic
Jatropha curcas (seed extract)	15–50	Spherical
Lemon grass (leaf extract)		Spherical/triangular
Henna (leaf)	21	Anisotropic
Henna (leaf)	39	Quasi-spherical
Scutellaria barbata D. Don	5–30	Spherical/triangular
Parthenium (leaf extract)	<50	Irregular
Tamarind (leaf extract)	20–40	Triangle
Neem (*Azadirachta indica*) leaf	50–100	Spherical
Geranium (*Pelargonium gra*veolens) (leaf extract)	16–40	Quasilinear superstructures
Eucalyptus hybrida leaf	50–150	Cubical
Medicago sativa (Alfalfa) (biomass)		Icosahedral/irregular
Cinnamomum zeylanicum (Leaf)	25	Spherical/prism
Tea leaves extract	20	Spherical/prism
Acalypha indica leaf extract	20–30	Spherical
Hibiscus Rosa sinensis	14	Spherical/prism
Honey	4	Spherical
Microorganisms		
Bacteria		
Bacillus cereus	4–5	Spherical
Staphylococcus aureus	160–180	–
Klebsiella pneumonia, Escherichia coli, Enterobacter cloacae	<52.5	–
Pseudomonas aeruginosa	15–30	–
Fungi		
Alternaria alternata		
Volvariella volvacea, Mushroom	<15	Spherical
Volvariella volvacea, Mushroom	20–150	Triangular/spherical/hexagonal
Fusarium oxysporum	5–15	
Trichoderma viride	5–40	Spherical/rod-like
Yeast		
Yeast strain MKY3	2–5	

3. Characterization of silver nanoparticles

Characterization of nanoparticles is important to understand and control nanoparticles synthesis and applications. Characterization is performed using a variety of different techniques

such as transmission and scanning electron microscopy (TEM, SEM), atomic force microscopy (AFM), dynamic light scattering (DLS), X-ray photoelectron spectroscopy (XPS), powder X-ray diffractometry (XRD), Fourier transform infrared spectroscopy (FTIR), and UV–Vis spectroscopy [61,62]. These techniques are used for determination of different parameters such as particle size, shape, crystallinity, fractal dimensions, pore size and surface area. Moreover, orientation, intercalation and dispersion of nanoparticles and nanotubes in nanocomposite materials could be determined by these techniques.

For instance, the morphology and particle size could be determined by TEM, SEM and AFM. The advantage of AFM over traditional microscopes such as SEM and TEM is that AFM measures three-dimensional images so that particle height and volume can be calculated. Furthermore, dynamic light scattering is used for determination of particles size distribution. Moreover, X-ray diffraction is used for the determination of crystallinity, while UV–Vis spectroscopy is used to confirm sample formation by showing the plasmon resonance.

4. Applications

AgNPs have been used extensively as anti-bacterial agents in the health industry, food storage, textile coatings and a number of environmental applications. It is important to note that despite of decades of use, the evidence of toxicity of silver is still not clear. Products made with AgNPs have been approved by a range of accredited bodies, including the US FDA, US EPA, SIAA of Japan, Korea's Testing and Research Institute for Chemical Industry and FITI Testing and Research Institute [63-68]. As anti-bacterial agents, AgNPs were applied in a wide range of applications from disinfecting medical devices and home appliances to water treatment [69]. Moreover, this encouraged the textile industry to use AgNPs in different textile fabrics. In this direction, silver nanocomposite fibers were prepared containing silver nanoparticles incorporated inside the fabric [70]. The cotton fibers containing AgNPs exhibited high anti- bacterial activity against Escherichia coli [64].

Furthermore, the electrochemical properties of AgNPs incorporated them in nanoscale sensors that can offer faster response times and lower detection limits. For instance, Manno *et al.* [65] electrodeposited AgNPs onto alumina plates gold micro-patterned electrode that showed a high sensitivity to hydrogen peroxide [66]. Catalytic activities of nanoparticles differ from the chemical properties of the bulk materials. Furthermore, AgNPs was found to catalyze the chemiluminescence from luminol–hydrogen peroxide system with catalytic activity better than Au and Pt colloid [67].

The optical properties of a metallic nanoparticle depend mainly on its surface plasmon resonance, where the plasmon refers to the collective oscillation of the free electrons within the metallic nanoparticle. It is well known that the plasmon resonant peaks and line widths are sensitive to the size and shape of the nanoparticle, the metallic species and the surrounding medium. For instance, nanoclusters composed of 2–8 silver atoms could be the basis for a new type of optical data storage. Moreover, fluorescent emissions from the clusters could potentially also be used in biological labels and electroluminescent displays [68-70].

5. Conclusion

The use of plants and microbes for the fabrication of nanoparticles is a rapid, low cost, eco-friendly and a single step method for biosynthesis process, however understanding the mechanism of involvement of biomolecules is lacking. This simple, efficient and rapid green synthesis of Ag NPs can be used in various biomedical and biotechnological applications. Production of Ag nanoparticles by biomaterials can be even more attractive if the size of the nanoparticles can be controlled. This can be achieved by studying the suitable reaction timings. However, any Ag product that claims its antibacterial properties must prove the product is safe to be released into the environment. Therefore, there is a great need for understanding of their sources, release interaction with environment, and possible risk assessment would provide a basis for safer use of nanomaterials with limited or no hazardous impact on environment.

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