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Green Synthesis and Characterization of Silver Nanoparticles Using *Vachellia seyal* Extract: A Comparative Physicochemical Study with Ascorbic Acid and Sodium Borohydride

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Abstract

The green synthesis of silver nanoparticles (AgNPs) offers an eco-friendly and biocompatible alternative to conventional methods, which often involve toxic reagents. In this study, AgNPs were synthesized via chemical reduction using three different reducing agents: sodium borohydride ($NaBH_4$), ascorbic acid, and a *Vachellia seyal* plant extract. UV-Visible spectroscopy confirmed the formation of AgNPs, with characteristic surface plasmon resonance bands observed at 430 nm for all three reducing agents. Dynamic Light Scattering (DLS) analysis revealed average particle sizes of **18.69 ± 0.04** nm with a polydispersity index (PDI) of **0.639** for $NaBH_4$, **42.44 ± 5.37** nm with a PDI of **0.234 ± 0.074** for ascorbic acid, and **129.53 ± 0.57** nm with a PDI of **0.092 ± 0.016** for *V. seyal* extract. Transmission Electron Microscopy (TEM) confirmed spherical morphology and sizes consistent with DLS measurements. These results indicate that the reducing strength of the agents significantly influences nanoparticle size and distribution. The *V. seyal* extract enabled the successful green synthesis of AgNPs with good size stability, highlighting its potential for pharmaceutical and biomedical applications.

Keywords

Silver Nanoparticles, *Vachellia seyal*, Green Synthesis, DLS, TEM, Polydispersity, Ascorbic Acid, $NaBH_4$

1. Introduction

The rise of nanoscience and nanotechnology has driven significant advances in the fabrication of nanoparticles, particularly silver nanoparticles (AgNPs), which have attracted increasing interest due to their remarkable physicochemical properties and wide-ranging applications in medicine, biotechnology, environmental science, and the pharmaceutical industry. Traditionally, their synthesis has relied on physicochemical processes involving toxic reagents, organic solvents, and energy-intensive production conditions, raising major concerns regarding environmental safety and biological compatibility.

To overcome these limitations, green synthesis has emerged as an innovative, sustainable, and environmentally friendly alternative. This approach uses plant extracts, microorganisms (bacteria, fungi, microalgae), or natural biomolecules that act simultaneously as reducing and stabilizing agents. Green synthesis allows the production of clean, biocompatible, and stable nanoparticles, while offering improved control over size, shape, and distribution [1]-[5]. Moreover, the natural richness of plant extracts in bioactive compounds (phenols, flavonoids, enzymes, tannins, etc.) enhances the biological functionalization of nanoparticles, thereby opening new prospects in diagnostics, targeted therapy, tissue engineering, and antimicrobial coatings [6]-[9].

Several plant species have demonstrated strong potential for the biosynthesis of homogeneous AgNPs with sizes ranging from 10 to 50 nm, characterized by excellent colloidal stability and pronounced antibacterial and photocatalytic properties [1]. Numerous studies have also highlighted the crucial influence of synthesis parameters such as pH, temperature, and reaction time on the size, distribution, and functional properties of the resulting nanoparticles [2] [3]. For instance, AgNPs synthesized from *Ficus carica* have shown notable antibacterial, antibiofilm, and anticancer effects, with an IC_{50} value of 8.4 μg/mL on certain cell lines [4] [10].

Similarly, *Eugenia uniflora* fruit extract has been reported to enable efficient green synthesis of silver nanoparticles with homogeneous morphology, good colloidal stability, and excellent antibacterial properties, highlighting the value of phytochemical-rich plant sources for the eco-responsible production of functional nanomaterials [11]. In another study, AgNPs biosynthesized from *Sida acuta* leaves were compared to nanocomposites combining these nanoparticles with reduced graphene oxide (rGO). The results revealed strong antibacterial activity, particularly against MRSA and *Proteus mirabilis*, as well as promising anticancer potential. These green nanoparticles, which were very small (−5 nm) and well dispersed, also exhibited notable antioxidant, anti-inflammatory, and anti-diabetic properties, underscoring their therapeutic potential [12].

In parallel, the use of microalgae such as *Graesiella emersonii* represents a promising strategy: extracellular metabolites produced by these microorganisms facilitate the reduction of metal ions and the stabilization of AgNPs in a bioinspired and finely regulated environment [5]. Recent developments have further enabled the incorporation of AgNPs into polymeric nanocomposites, opening new avenues for applications in regenerative medicine, drug delivery, and the design of smart antimicrobial materials [6]-[9].

In this context, the present study aims to investigate the potential of green synthesis of AgNPs from specific biological resources, to characterize the resulting nanoparticles, and to evaluate their colloidal stability. More specifically, the objective is to synthesize silver nanoparticles from *Vachellia seyal* extracts via a chemical reduction process using ascorbic acid, with sodium borohydride serving as a reference, and to characterize them using UV-Visible spectrophotometry, dynamic light scattering, and electron microscopy.

2. Materials and Methods

2.1. Materials

The reagents used in this study were silver nitrate ($AgNO_3$), sodium borohydride ($NaBH_4$) (Aldrich Laboratories), ascorbic acid (National Laboratory for Drug Quality Control), and *Vachellia seyal* extract prepared by the Pharmacognosy Laboratory of Cheikh Anta Diop University of Dakar. Additional reagents included sodium citrate, hydrogen peroxide (H_2O_2) (Gilbert Laboratories), and the solvents ethanol and methanol (Oxford Laboratories). The equipment employed consisted of a Heidolph MR 3001 K magnetic stirrer, a Mettler precision balance (model BC), and standard laboratory glassware and tools, including beakers, Erlenmeyer flasks, pipettes, micropipettes, spatulas, 10 mL test tubes, volumetric flasks, and filter paper.

2.2. Methods

2.2.1. Standard Synthesis of Nanoparticles

The chemical reduction method is commonly employed to synthesize metallic nanoparticles with controlled size and shape. This approach involves the use of a metal precursor (such as silver nitrate, $AgNO_3$), a reducing agent (such as sodium borohydride, $NaBH_4$) to reduce metal ions, and a stabilizing agent to prevent nanoparticle agglomeration [13]-[18]. In this study, silver nanoparticles were synthesized by adding $NaBH_4$ to $AgNO_3$ to reduce $+Ag^+$ ions, in the presence of sodium citrate as a stabilizing agent. To induce the formation of silver nanoprisms, hydrogen peroxide (H_2O_2) was introduced to promote anisotropic growth, while potassium bromide (KBr) was used to control nanoparticle size. Thus, sodium citrate served as a stabilizer, H_2O_2 directed anisotropic growth toward prism-like structures, and KBr regulated particle dimensions [19].

The synthesis was carried out in three steps. First, a control synthesis was performed using sodium borohydride as the reducing agent. In the second step, ascorbic acid was used as an alternative reducing agent. Finally, in the third step, synthesis was conducted using a plant extract as a green reducing agent. Each resulting solution was characterized by UV-Visible spectrophotometry, dynamic light scattering (DLS), and electron microscopy to assess their optical and structural properties.

Despite the different synthesis conditions tested, only spherical nanoparticles were obtained and characterized.

2.2.2. Green Synthesis of Nanoparticles

The previously described protocol was adapted by replacing sodium borohydride ($NaBH_4$) with two biological reducing agents: a plant extract of *Vachellia seyal* and a solution of ascorbic acid. A 10^{-2} mol·L^{-1} (1.7612 g·L^{-1}) ascorbic acid solution was prepared, and the samples were synthesized following the same procedure as for the $NaBH_4$-based method. Similarly, a *Vachellia seyal* extract solution was prepared by dissolving 0.08806 g of the dried extract in 50 mL of distilled water in a 100 mL beaker containing a magnetic stir bar. After stirring for 10 minutes, the mixture was filtered into a 50 mL volumetric flask using filter paper. The corresponding samples were then prepared following the same steps as for $NaBH_4$. For all samples, UV-Visible absorption spectra were recorded to monitor nanoparticle formation and characterize their properties.

2.2.3. Characterizations Methods

Absorption spectrophotometry: UV-Visible spectroscopy is a widely used and reliable analytical technique for the primary characterization of synthesized nanoparticles. It is particularly useful for monitoring nanoparticle formation and assessing the stability of silver nanoparticles [20] [21]. This method relies on the absorption of a monochromatic beam of light in the ultraviolet (UV) or visible range as it passes through a medium containing an absorbing species. The amount of light absorbed is proportional to the concentration of that species, according to Beer-Lambert's law. For nanoparticles, the absorption maxima are associated with surface plasmon resonance and can be used to estimate the average size of nanoprism particles. These values can then be compared to a calibration curve to determine particle dimensions. The UV-Visible absorption spectra of the synthesized samples were recorded using a Thermo Scientific® Evolution 3000 UV-Visible spectrophotometer, operated with Vision Pro software, ensuring accurate and reproducible measurements.

Size and charge measurement: The size distribution and polydispersity index (PDI) of the nanoparticles were measured using a Nanosizer (Malvern Instruments, Orsay, France) through dynamic light scattering (DLS) techniques. All experiments were performed in triplicate to ensure accuracy and reproducibility.

Morphological characterization: The morphology of the nanoparticles was examined using a Philips Morgagni 268 D Transmission Electron Microscope (TEM) operating at 70 kV. No staining agents were used, and the samples were diluted 100-fold with MilliQ water. A drop of the diluted sample was placed onto a carbon-coated copper grid (Type A, 300 mesh, Redding, PA) and dried at 40˚C prior to imaging.

3. Results

The synthesis was carried out by chemical reduction in three steps. First, a control synthesis was performed using sodium borohydride ($NaBH_4$). Next, a second synthesis was conducted using ascorbic acid as the reducing agent. Finally, the last synthesis was carried out with the *Vachellia seyal* extract. Each resulting solution was characterized by UV-Visible spectrophotometry, dynamic light scattering, and electron microscopy (performed for the synthesis using the *V. seyal* extract) to analyze their optical and structural properties.

Upon addition of the $NaBH_4$ solution at 5.0×10^{-3} mol·L^{-1}, the reaction mixture immediately changed color: yellow after one minute, followed by orange, then brown, gray, and finally blue after approximately three minutes. The corresponding absorption spectrum is shown in **Figure 1**.

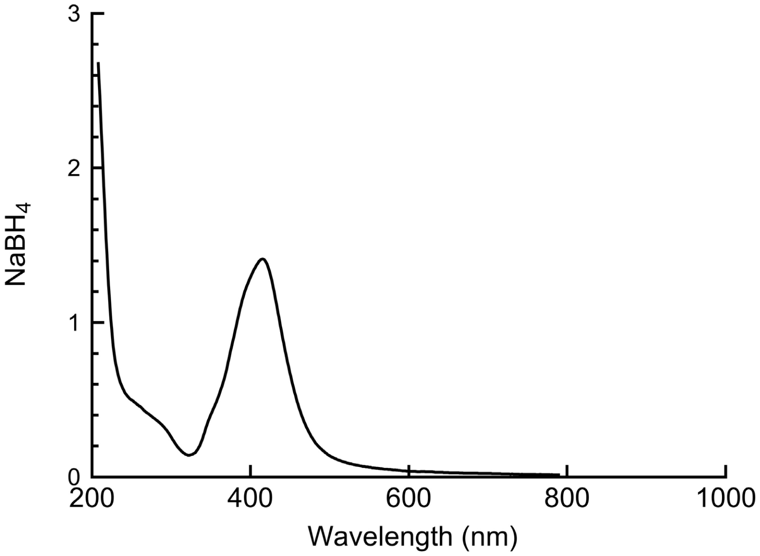

Figure 1. The ultraviolet-visible spectra of synthesized silver nanoparticles using $NaBH_4$.

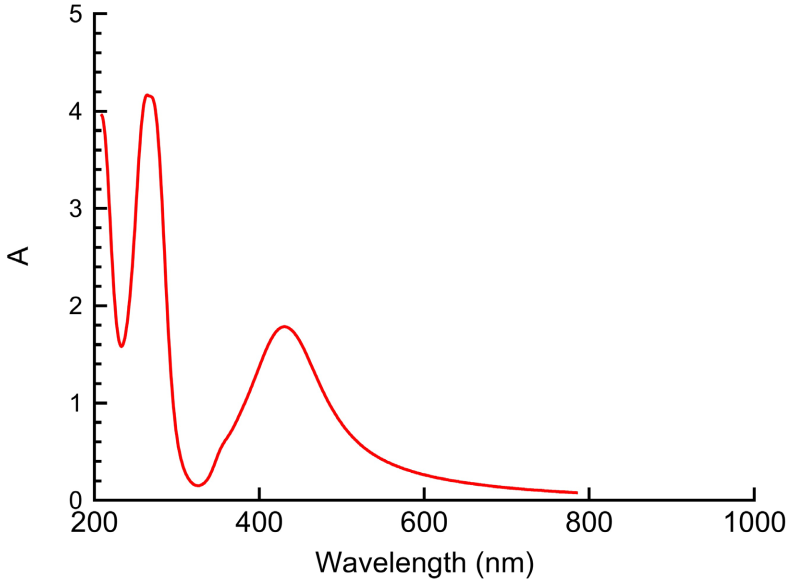

Figure 2. The ultraviolet-visible spectra of synthesized silver nanoparticles using Ascorbic acid.

After the addition of the ascorbic acid solution at 1.0×10^{-2} mol·L^{-1}, the reaction mixture changed color, turning light blue after approximately three minutes. The corresponding absorption spectrum is shown in **Figure 2**.

After the addition of the *Vachellia seyal* extract solution at 1.7612 g·L^{-1}, the reaction mixture rapidly developed an orange color, which remained stable after several minutes of stirring. The corresponding absorption spectrum is shown in **Figure 3**.

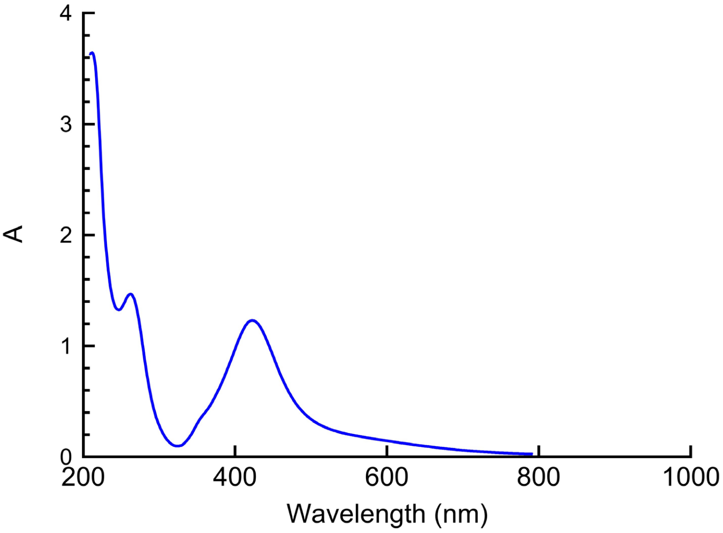

Figure 3. The ultraviolet-visible spectra of biosynthesized silver nanoparticles using *V. seyal*.

3.1. Size and Charges Measurements

Characterization of the preparations by DLS revealed the presence of nanoparticles in all samples, with size distributions varying depending on whether the synthesis was performed using $NaBH_4$, ascorbic acid, or the *V. seyal* extract (**Figure 4**).

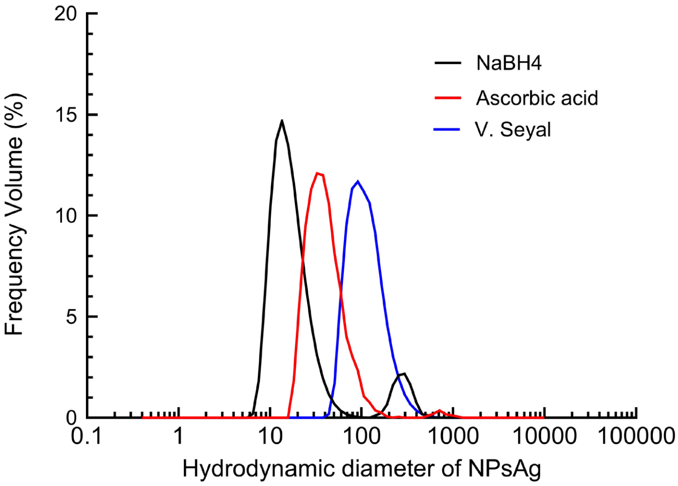

Figure 4. Size distribution of AgNPs obteined with different reductor (Black, $NaBH_4$; Red, Ascorbic; Blue, *V. seyal*).

Table 1. Size distribution and polydispersity according to the reducing agent.

Reducing agent	*Size* (nm)	*PDI*
$NaBH_4$	18.64	0.640
	18.75	0.638
	18.68	0.639
Ascorbic acide	50.51	0.132
	40.36	0.225
	36.47	0.345
Vachellia seyal	130.4	0.118
	128.7	0.077
	129.5	0.083

3.2. Morphological Characterization

Morphological characterization of the nanoparticles (AgNPs) was conducted using Transmission Electron Microscopy (TEM) (see **Figure 5**). The AgNPs (**Figures 5(a)-(c)**) appeared as round spheres, with a size consistent with the dynamic light scattering (DLS) results, showing a distribution centered around a diameter of 130 nm. Some nanoparticles were isolated on the carbon support, while others formed aggregates (see **Table 1**) or clusters, a typical occurrence during sample drying.

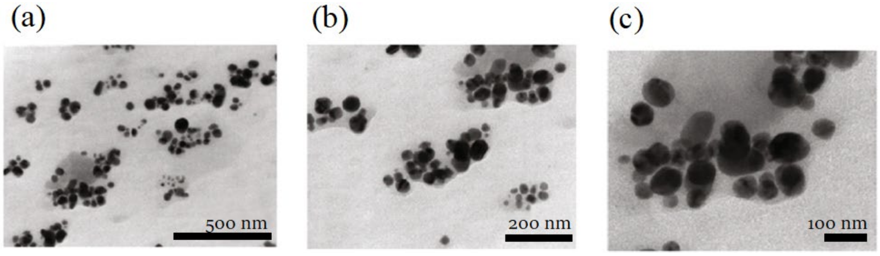

Figure 5. Transmission electron micrograph of AgNPs using the *V. seyal* extract (a) scale bar 500 nm (b) scale bar 200 nm (c) scale bar 100 nm.

4. Discussion

The absorption spectrum of the $NaBH_4$-synthesized sample exhibits a broad band between 350 and 500 nm, confirming the presence of nanoparticles with a highly polydisperse size distribution [22]. In contrast, the sample synthesized with ascorbic acid shows a much narrower band with an absorption maximum at 430 nm, indicating a lower degree of polydispersity and nanoparticles ranging in size from 60 to 70 nm. For the sample synthesized with ascorbic acid, which is a weaker reducing agent than $NaBH_4$, the spectrum displays a very broad band, suggesting a very high polydispersity index, with an absorption maximum at 430 nm.

For the *Vachellia seyal* extract, the absorption spectra of the resulting nanoparticles display moderately narrow bands, suggesting a relatively low to moderate polydispersity. Silver nanoparticles exhibit unique optical properties that enable strong interactions with specific wavelengths of light. Numerous studies have shown that, in general, absorption bands in the 200 - 800 nm range are optimal for characterizing nanoparticles with sizes between 2 and 100 m. In silver nanoparticles, the conduction and valence bands are very close, allowing electrons to move freely. These free electrons give rise to a surface plasmon resonance (SPR) absorption band, resulting from the collective oscillation of electrons in resonance with incident light. The SPR band depends on particle size, the dielectric medium, and the chemical environment. The presence of a surface plasmon peak is well documented for metallic nanoparticles in the 2 - 100 nm range [20] [21] [23].

Characterization by DLS confirmed the presence of silver nanoparticles. $NaBH_4$-synthesized nanoparticles had an average size of 18.69 ± 0.04 nm with a polydispersity index (PDI) of 0.639. Using ascorbic acid, the average nanoparticle size was 42.44 ± 5.37 nm with a PDI of 0.234 ± 0.074. The use of *V. seyal* extract significantly increased particle size to 129.53 ± 0.57 nm, with a PDI of 0.092 ± 0.016. These results indicate an inverse relationship between nanoparticle size and polydispersity index and confirm that nanoparticle size correlates with the reducing strength of the agent [23]-[25].

Overall, these findings demonstrate the feasibility of green synthesis using plant extracts to produce silver nanoparticles with controlled sizes suitable for pharmaceutical applications. Morphological characterization by electron microscopy further confirmed the successful biosynthesis of these nanoparticles.

5. Conclusion

This study demonstrates the successful synthesis of silver nanoparticles (AgNPs) using both conventional ($NaBH_4$, ascorbic acid) and green (*Vachellia seyal* extract) reducing agents. Spectroscopic and microscopic analyses confirmed the formation of AgNPs in all cases, with notable differences in size and polydispersity depending on the reducing agent used. In particular, nanoparticles synthesized with *V. seyal* extract exhibited larger sizes but a lower polydispersity index, indicating good colloidal stability and uniformity. These results highlight the critical role of the reducing agent's antioxidant capacity in determining nanoparticle characteristics. The green synthesis approach using *V. seyal* not only provides an environmentally sustainable alternative but also offers promising potential for biomedical and pharmaceutical applications, owing to its simplicity, eco-compatibility, and the biofunctional properties of the plant-derived compounds. Future studies will focus on optimizing synthesis parameters and evaluating the biological activities of these nanoparticles for potential therapeutic applications.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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