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Green Synthesis, Characterization And Antibacterial Activity Of Silver Nanoparticles Using Soybean Pod Dry Waste Extract

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Abstract

Nanotechnology has attracted considerable interest due to its wide-ranging applications in healthcare and environmental management; however, many conventional nanoparticle synthesis techniques rely on toxic chemicals and energy-intensive processes that raise environmental concerns. In this study, an environmentally benign and sustainable method was developed for the green synthesis of silver nanoparticles (AgNPs) using soybean pod dry waste extract as a natural reducing and stabilizing agent. The synthesis was conducted under mild reaction conditions and was initially evidenced by a visible color change, followed by UV–Visible spectroscopic analysis, which showed a characteristic surface plasmon resonance band at 420 nm. Morphological examination using scanning electron microscopy revealed predominantly spherical nanoparticles with slight agglomeration. Energy dispersive X-ray analysis confirmed silver as the principal elemental component and indicated the presence of plant-derived biomolecules contributing to nanoparticle stabilization. Fourier transform infrared spectroscopy further demonstrated the involvement of phenolic, flavonoid, and proteinaceous functional groups in the reduction and capping processes. The synthesized AgNPs exhibited significant, concentration-dependent antibacterial activity. Overall, the findings underscore the potential of soybean pod dry waste as a low-cost and renewable resource for the eco-friendly synthesis of biologically active silver nanoparticles with promising antibacterial applications.

Keywords: Green synthesis, Silver nanoparticles, Soybean pod dry waste, Antibacterial activity, Nanotechnology

Introduction

Nanotechnology has emerged as a valuable tool for advancements in healthcare and environmental protection. However, many commonly used methods for nanoparticle synthesis pose serious environmental concerns. These conventional approaches frequently rely on toxic chemicals, require high energy consumption, and produce hazardous by-products that negatively impact ecosystems [1,4,5]. In particular, traditional chemical routes for silver nanoparticle synthesis employ aggressive reducing and stabilizing agents, resulting in environmentally unsafe waste streams [4,5,10]. At the same time, large volumes of soybean pod (husk) waste generated after harvesting are often burned or discarded, contributing to pollution rather than being utilized productively [6,9].

Green synthesis has gained attention as an eco-friendly alternative, as it utilizes plant-based biomolecules to act as natural reducing and stabilizing agents, allowing nanoparticle formation under mild and environmentally safe conditions [1,2,3]. In this strategy, phytochemicals present in plant materials serve as biological nano-factories, promoting the synthesis of stable and biologically active silver nanoparticles without the need for toxic reagents or complex infrastructure [2,4,8].

Soybean husk represents an attractive raw material for green synthesis due to its abundant availability as an agricultural by-product, low economic value, and rich composition of phenolics, proteins, and polysaccharides capable of reducing silver ions and preventing nanoparticle aggregation [6,9]. Transforming this underutilized agro-residue into valuable silver nanoparticles aligns well with the concepts of waste valorization and circular bioeconomy, particularly in soybean-producing regions [6,9].

In this study, silver nanoparticles were synthesized using soybean pod dry waste extract through a simple, low-energy green approach, followed by detailed physicochemical and antibacterial characterization. While numerous studies have reported plant-mediated and agro-waste-based synthesis of silver nanoparticles [1,2,4,7,8], and some investigations have explored soybean seeds or other leguminous sources as reducing agents [2,10], the use of soybean pod or husk waste remains largely unexplored. The present work addresses this gap by demonstrating an environmentally sustainable, scalable method for producing bioactive silver nanoparticles from soybean pod dry waste [6,9].

Materials and Methods

Materials

Soybean pod dry waste was collected from agricultural fields in Akola, Maharashtra, India. Analytical-grade silver nitrate ($AgNO_3$) was used as the silver precursor. Distilled water was used for preparation of all solutions. Standard nutrient agar and nutrient broth were employed for antibacterial studies. The bacterial strains *Escherichia coli* (Gram-negative) and *Staphylococcus aureus* (Gram-positive) were used as test organisms.

Preparation of Soybean Pod Dry Waste Extract

Dry soybean pods were cleaned to remove dirt and debris, then air-dried and ground to a fine powder using a domestic blender. Exactly 15 g of powder was boiled in 150 mL distilled water for 25 minutes to extract bioactive reducing agents. The extract was allowed to cool at room temperature, filtered sequentially through muslin cloth and Whatman No. 1 filter paper to remove excess biomolecules and impurities. The resulting clear filtrate was stored at 4 °C and used for nanoparticle synthesis.

Green Synthesis of AgNPs

A fresh 1 mM $AgNO_3$ solution was prepared in distilled water. Typically, 8 mL of soyabean pod extract was added dropwise to 40 mL $AgNO_3$ solution under magnetic stirring at room temperature (25± 2°C), with optional mild heating (40-50°C) to accelerate reduction. Formation of AgNPs was visually confirmed by a rapid color change from pale yellow to dark brown within 30-60 minutes, indicating the reduction of Ag^+ to

Ag^0. The synthesis conditions were optimized by maintaining an extract-to-$AgNO_3$ ratio of 1:10, adjusting the pH to 9 using NaOH at room temperature with a reaction time of 1 hr.

Purification of Nanoparticles

The reaction mixture was centrifuged at 12,000 rpm for 20 minutes to collect the nanoparticle pellet. The pellete was washed three times with distilled water and once with ethanol to remove excess biomolecules. The purified nanoparticles were dried and stored for further characterization and assays.

Characterization

UV–Visible Spectroscopy

The optical characterization of silver nanoparticles was carried out using UV–Visible spectroscopy. An aliquot of the colloidal nanoparticle solution was analyzed in the wavelength range of 300–700 nm using a UV–Visible spectrophotometer, with distilled water used as a blank. The spectra were recorded at room temperature to monitor the optical properties of the synthesized nanoparticles.

Scanning Electron Microscopy (SEM)

The surface morphology of the silver nanoparticles was examined using scanning electron microscopy. A small quantity of the dried nanoparticle sample was mounted on a conductive stub using carbon tape and coated with a thin conductive layer to prevent charging. The samples were then observed under SEM at appropriate accelerating voltage and magnification.

Energy Dispersive X-ray Analysis (EDAX)

Elemental analysis of the silver nanoparticles was conducted using energy dispersive X-ray spectroscopy coupled with the SEM instrument. The EDAX spectrum was recorded during SEM imaging to determine the elemental composition of the nanoparticle sample.

Fourier Transform Infrared (FTIR) Spectroscopy

Fourier transform infrared spectroscopy was used to analyze the functional groups associated with the silver nanoparticles. The dried nanoparticle sample was mixed with potassium bromide (KBr) and compressed into pellets. FTIR spectra were recorded in the range of 4000–400 cm^{-1} using an FTIR spectrophotometer.

Antibacterial activity

The antibacterial activity of the synthesized silver nanoparticles was assessed by the agar well diffusion method. *E. coli* and *S. aureus* were cultured in suitable growth media and standardized before use. Sterile nutrient agar plates were inoculated uniformly with the microbial cultures, and wells of uniform diameter were aseptically made using a sterile cork borer. Different concentrations of the silver nanoparticle suspension were added into the wells, while a standard antibiotic and the solvent served as positive and negative controls, respectively. The plates were incubated under appropriate conditions for a specified duration to evaluate antibacterial efficacy.

Results and Discussion

Visual observation and UV–Vis analysis

A rapid color change was observed immediately after mixing the plant extract with $AgNO_3$ solution, confirming the formation of silver nanoparticles. UV–Visible spectroscopic analysis revealed a distinct surface plasmon resonance (SPR) band centered at 420 nm, further supporting nanoparticle formation.

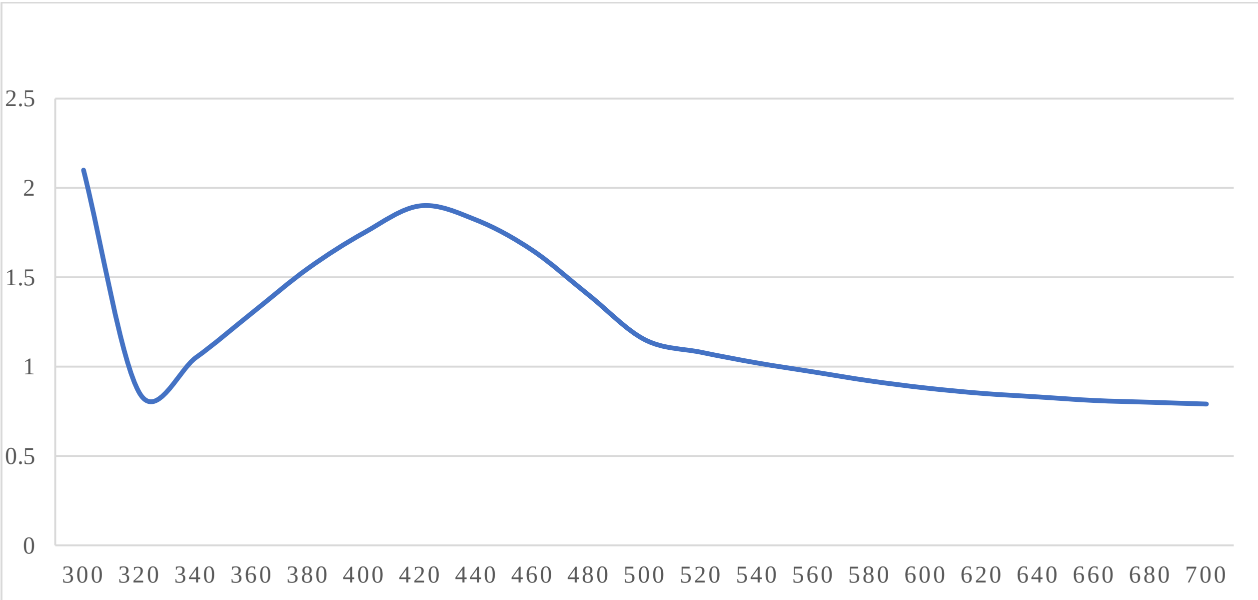

Figure 1. UV-Vis spectrum of AgNPs

SEM & Energy Dispersive X-ray Analysis (EDAX)

Area 1 **Area 2**

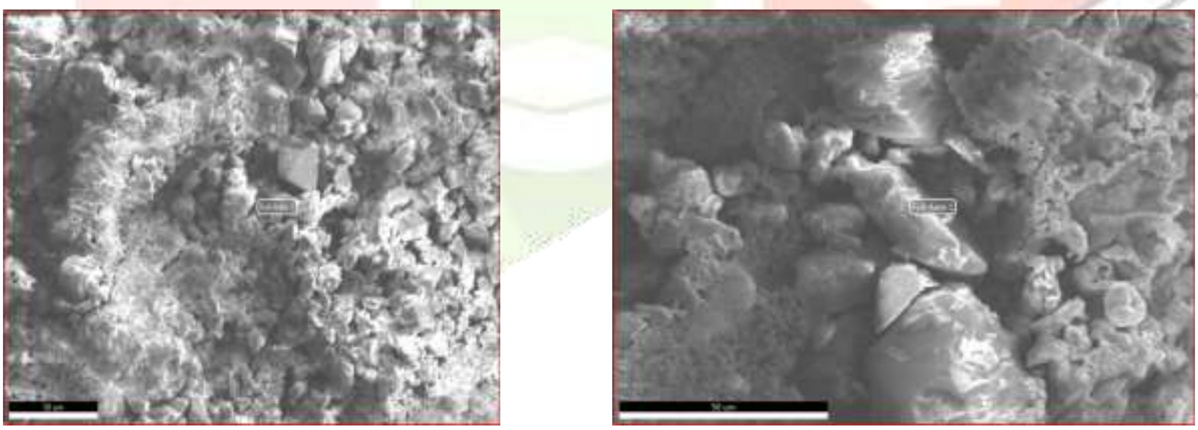

Fig. 2. (A) SEM images of spherical AgNPs .

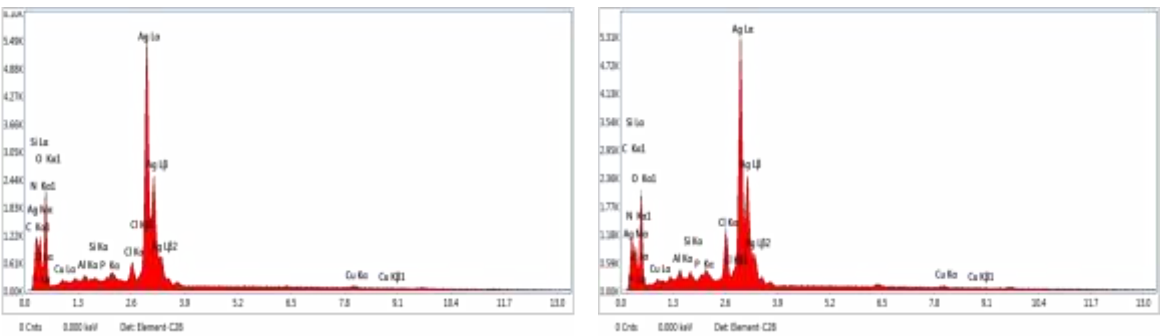

Fig. 2. (B) Size distribution

SEM analysis revealed that the synthesized silver nanoparticles were predominantly spherical in shape with a relatively uniform size distribution and slight agglomeration. EDAX analysis showed that silver (≈ 47 wt%)

was the major constituent of the synthesized nanoparticles, confirming the successful formation of silver nanoparticles using soybean pod dry waste extract. The presence of oxygen (32 wt%), nitrogen (12 wt%), and carbon (5 wt%) was attributed to plant-derived biomolecules such as proteins, amino acids, and polyphenolic compounds associated with the nanoparticle surface, indicating effective capping and stabilization. A minor copper signal (1.5 wt%) was observed, which originated from the copper grid used during SEM analysis.

FTIR

FTIR spectrum of Soyabean Pod Dry Waste Extract-mediated silver nanoparticles exhibited characteristic absorption bands confirming the role of plant-derived biomolecules in nanoparticle formation and stability. Peaks at 987.60 cm^{-1} and 940.34 cm^{-1} were attributed to C-O stretching vibrations from phenolic and flavonoid groups, indicative of their involvement in Ag^+ bioreduction. Bands around 851.61, 806.28, and 749.38 cm^{-1} corresponded to aromatic C-H out-of-plane bending, while lower wavenumber peaks (699-435 cm^{-1}) in the fingerprint region suggested interactions between nanoparticles and protein as capping agents from Soyabean Pod Dry Waste Extract, ensuring colloidal stability. The absence of precursor peaks further confirms successful nanoparticle formation without the utilization of harsh chemical reagents.

Antibacterial activity

Figure 3. Zone of inhibition by soybean pod AgNPs against (a) E. coli and (b) S. aureus.

The synthesized silver nanoparticles exhibited notable antibacterial activity against the *E. coli* and *S. aureus*, as evidenced by clear zones of inhibition of 1cm observed around the wells. The antibacterial effect was found to be concentration dependent, with higher concentrations of silver nanoparticles producing larger zones of inhibition compared to lower concentrations and the negative control. The activity of the nanoparticles was comparable to that of the standard antibiotic, demonstrating their effective antibacterial potential.

Conclusion

The present study successfully demonstrated the green synthesis of silver nanoparticles using soybean pod dry waste extract, as confirmed by visual observation and comprehensive physicochemical characterization. The rapid color change and a characteristic surface plasmon resonance band at 420 nm in UV–Visible spectroscopy indicated effective nanoparticle formation. SEM analysis revealed predominantly spherical nanoparticles with relatively uniform size distribution, while EDAX confirmed silver as the major elemental component along with the presence of plant-derived biomolecules that contributed to effective capping and stabilization. The FTIR analysis confirms that phenolic, flavonoid, and proteinaceous biomolecules present in soyabean pod dry waste extract effectively mediated the reduction. Additionally, the synthesized silver nanoparticles exhibited significant, concentration-dependent antibacterial activity against the tested microorganisms, highlighting their potential as eco-friendly antibacterial agents. Overall, the findings validate soybean pod dry waste extract as an efficient, sustainable reducing and stabilizing agent for the synthesis of biologically active silver nanoparticles.

References

1. Ahmed, S., Ahmad, M., Swami, B. L., & Ikram, S. (2016). A review on plant extract–mediated synthesis of silver nanoparticles for antibacterial applications. *Journal of Advanced Research, 7*(1), 17–28.
2. Gómez-García, R., Campos, D. A., & Aguilar, C. N. (2021). Agro-industrial residues as sources of bioactive compounds for nanotechnology applications. *Food Research International, 140*, 109855.
3. Iravani, S. (2011). Green synthesis of metal nanoparticles using plants. *Green Chemistry, 13*(10), 2638–2650.
4. Kumari, R. M., Thapa, N., Gupta, N., Kumar, A., & Nimesh, S. (2016). Antibacterial and antioxidant activity of silver nanoparticles synthesized using *Potentilla fulgens* root extract. *Journal of Radiation Research and Applied Sciences, 9*(1), 79–90.
5. Mittal, A. K., Chisti, Y., & Banerjee, U. C. (2013). Synthesis of metallic nanoparticles using plant extracts. *Biotechnology Advances, 31*(2), 346–356.
6. Pathak, P. D., Mandavgane, S. A., & Kulkarni, B. D. (2017). Valorization of agricultural waste into value-added products. *Waste and Biomass Valorization, 8*(4), 1059–1074.
7. Rai, M., Yadav, A., & Gade, A. (2009). Silver nanoparticles as a new generation of antibacterials. *Biotechnology Advances, 27*(1), 76–83.
8. Sharma, V. K., Yngard, R. A., & Lin, Y. (2009). Silver nanoparticles: Green synthesis and their antibacterial activities. *Advances in Colloid and Interface Science, 145*(1–2), 83–96.
9. Chandran, N., Ramesh, S., & Shanmugam, R. (2024). Synthesis of Silver Nanoparticles Using Azadirachta indica and Syzygium aromaticum Extract and Its Antibacterial Action Against Enterococcus faecalis: An In Vitro Study. *Cureus*, *16*(7), e65044.
10. Zhang, X.-F., Liu, Z.-G., Shen, W., & Gurunathan, S. (2016). Silver nanoparticles: Synthesis, characterization, properties, applications, and therapeutic approaches. *International Journal of Molecular Sciences, 17*(9), 1534.
11. Hosamani, G.B., Patil, R.R., Benagi, V.I., Chandrashekhar, S., & Nandihali, B.S. (2019). Synthesis of Green Silver Nanoparticles from Soybean Seed and its Bioefficacy on Spodoptera litura (F.). *International Journal of Current Microbiology and Applied Sciences*.
12. Ch. Prasad, Ponneri Venkateswarlu (2014). Soybean seeds extract based green synthesis of silver nanoparticles. *Indian Journal of Advances in Chemical Science 2*(3), 208-211.
13. Nande, A., Raut, S., Michalska-Domanska, M., & Dhoble, S. J. (2021). Green Synthesis of Nanomaterials Using Plant Extract: A Review. *Current pharmaceutical biotechnology*, *22*(13), 1794–1811.
14. Dangi Sandeep and Biradarpatil NK (2018). Biosynthesis of silver nanoparticles using soybean seed extract. *J Pharmacogn Phytochem* 7(4):2676-2680
15. Alabdallah, N. M., & Hasan, M. M. (2021). Plant-based green synthesis of silver nanoparticles and its effective role in abiotic stress tolerance in crop plants. *Saudi journal of biological sciences*, *28*(10), 5631–5639.
16. Darbin Kumar Poudel, Purushottam Niraula, Himal Aryal, Biplab Budhathoki, Sitaram Phuyal, Rishab Marahatha, Kiran Subedi (2022). Plant-mediated green synthesis of Ag NPs and their possible applications: A critical review. *Journal of Nanotechnology, 2022*, Article 2779237.