Asparagus racemosus mediated Green Synthesis of Polymer-Coated Silver Nanoparticles: Insights into Biomedical Efficacy

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Abstract

This study highlights the eco-friendly synthesis of polymer-encapsulated silver nanoparticles (AgNPs) using Asparagus racemosus extract as a natural reducing and stabilizing agent. This approach provides a sustainable and cost-effective alternative to conventional chemical methods, minimizing the use of toxic reagents and promoting green nanotechnology. This biogenic synthesis approach reduces environmental impact and leverages the therapeutic properties of Asparagus racemosus, known for its medicinal benefits. The synthesized AgNPs were encapsulated in a polymer matrix to improve stability and enable controlled release, enhancing their suitability for biomedical use. Characterization of the synthesized nanoparticles using UV-Vis spectroscopy, FTIR, XRD and TEM confirmed their successful formation, consistent morphology and nanoscale dimensions.

The polymer-encapsulated AgNPs demonstrated strong antimicrobial effects against various pathogenic bacteria, indicating broad-spectrum potential. This green synthesis approach not only ensures sustainable NPs production but also underscores the potential of these AgNPs as dual-functional agents for antioxidant and antimicrobial therapy. The findings suggest that Asparagus racemosus-mediated AgNPs could be developed into effective therapeutic agents with lower environmental impact and improved biomedical performance.

Keywords: Green synthesis, silver nanoparticles, *Asparagus racemosus*, Antimicrobial activity, Biomedical applications.

Introduction

Silver nanoparticles (AgNPs) have garnered significant attention in recent years due to their unique physicochemical properties, making them highly valuable for various applications, particularly in biomedicine. Their nanoscale size and high surface area-to-volume ratio enhance their reactivity, enabling them to exhibit potent antimicrobial, antiviral and anticancer activities even at low concentrations[17]. This characteristic is especially advantageous in the medical field as AgNPs can interact directly with biological cells and pathogens, enhancing therapeutic efficacy and reducing dosage requirements. Given these benefits, AgNPs have found applications in drug delivery systems, wound healing, medical device coatings and cancer therapies, underscoring their relevance and utility across biomedical domains[23].

The green synthesis of AgNPs offers a sustainable alternative to conventional chemical and physical methods, which typically involve toxic reagents and high energy consumption. This environmentally friendly strategy uses natural agents, such as plant extracts, bacteria, or fungi, to aid the reduction and stability of AgNPs without adding hazardous compounds[14]. Green synthesis not only minimizes environmental impact but also generates NPs that are often more biocompatible, as the natural capping agents from plant extracts provide stability and can further enhance biological activity[1]. By utilizing renewable resources, green synthesis aligns with the principles of sustainability and holds promise for scalable, eco-friendly NPs production[8].

Among the various plant based methods for green synthesis, *Asparagus racemosus* has emerged as a valuable agent due to its medicinal properties and rich phytochemical profile. Known for its antioxidant, anti-inflammatory and anticancer effects, *Asparagus racemosus* contains bioactive compounds like saponins, flavonoids and phenolic acids that facilitate the reduction of silver ions and stabilize AgNPs during synthesis[3]. The therapeutic properties of these compounds contribute to producing AgNPs with enhanced bioactivity, making the resulting NPs potentially more effective in medical applications[5]. The dual benefit of synthesizing AgNPs while harnessing the medicinal potential of *Asparagus racemosus* makes it a promising candidate for green synthesis.

A review of existing literature on AgNPs highlights their synthesis, functionalization and extensive use across various fields, with a strong emphasis on biomedical applications. Researchers have explored various synthesis methods aiming to optimize particle size, shape and surface properties as these factors influence their effectiveness in biological systems. The antimicrobial and anticancer properties of AgNPs are well-documented with studies investigating their interactions with pathogens and cancer cells to understand their mechanisms of action and enhance therapeutic outcomes[4].

The vast body of research underscores the versatility of AgNPs and their potential for continued development in targeted therapies, diagnostics and environmental

applications[16]. The biological activity of AgNPs, notably their antibacterial and anticancer properties, has been widely researched, demonstrating their potential to break microbial cell membranes, create reactive oxygen species (ROS) and trigger apoptosis in cancer cells[15]. Their selective cytotoxicity makes them valuable for treating infections and targeting cancer cells while sparing healthy cells in lower concentrations. These properties also enable their use in drug delivery systems, where AgNPs can be engineered to deliver drugs precisely to diseased cells or tissues[6,9,10,20].

This study focuses on synthesizing polymer-encapsulated AgNPs using *Asparagus racemosus* extract to create NPs with improved stability, bioavailability and efficacy for antimicrobial and anticancer applications. Polymer encapsulation is employed to enhance stability, control AgNPs release and increase biocompatibility, facilitating safer and more effective therapeutic use. By integrating green synthesis with polymer encapsulation, this research introduces a novel approach to producing AgNPs that harness the medicinal properties of *Asparagus racemosus* and enable controlled therapeutic delivery, contributing to advancements in NPs based treatments.

Material and Methods

Materials and reagents: The materials utilized in this research include *Asparagus racemosus* root extract (Pinnacle Herbals Pvt. Ltd., Bengaluru, India; Pharmaceutical grade), silver nitrate ($AgNO_3$) (Loba Chemie Pvt. Ltd., Mumbai, India; AR grade, 99.9% purity) and polyvinylpyrrolidone (PVP) (Central Drug House (CDH) Pvt. Ltd., New Delhi, India; USP grade). Distilled water was obtained from the Department of Chemistry, Gokul Global University, Siddhapur, India. DPPH (2,2-diphenyl-1-picrylhydrazyl) (Himedia Laboratories Pvt. Ltd., Mumbai, India; AR grade) and Methanol (Finar Limited, Ahmedabad, India; HPLC grade, ≥99.8% purity) were used in the study. Ascorbic acid (Thomas Baker Chemicals Pvt. Ltd., Mumbai, India; AR grade, ≥99% purity) was utilized as a reference antioxidant. For microbiological studies, nutrient agar medium (Titan Biotech Ltd., New Delhi, India; Laboratory grade) and Chloramphenicol (Sisco Research Laboratories (SRL) Pvt. Ltd., Mumbai, India; Pharmaceutical grade, ≥98% purity) were employed.

Preparing an aqueous extract of *Asparagus Racemosus*: The aqueous extract of *Asparagus racemosus* (Shatavari) was made by collecting fresh roots and carefully washing them with distilled water to eliminate contaminants. The cleaned roots were air-dried at room temperature until completely dehydrated, then crushed to a fine powder. Approximately 10 grams of this powdered root material were combined with 100 mL of distilled water at a 1:10 ratio. To efficiently extract the bioactive components, the mixture had been heated in a water bath at 60-70°C for 2-3 hours, with intermittent stirring. After cooling to room temperature, the extract was passed through Whatmann filter paper to produce a clear solution. The extract was either utilized right away or kept at 4°C in a dark, airtight container to retain stability and avoid deterioration until further usage[10].

Green synthesis of AgNPs using *Asparagus racemosus* extract: The green synthesis of AgNPs using *Asparagus racemosus* extract begins with the manufacture of an aqueous extract from plant roots, acting as a natural reducing and stabilizing agent. The silver ion precursor was produced as a 1 mM solution of silver nitrate ($AgNO_3$). The *Asparagus racemosus* extract was added dropwise to the $AgNO_3$ solution, stirring continuously at room temperature. After 2-3 hours at ambient conditions, the reaction mixture gradually changed color to yellowish-brown, indicating the reduction of Ag^+ ions and production of AgNPs via surface plasmon resonance.

The AgNPs were purified by centrifuging the mixture at 10,000 rpm for 20 minutes to separate the NPs, then wash with distilled water to remove any remaining extract or unreacted ions. The purified AgNPs were then dried and kept for further characterization and applications[18].

Synthesis of Polymer Encapsulated PVP-AgNPs: The synthesis of PVP-AgNPs involves using PVP as both a stabilizing and capping agent to enhance NPs stability and dispersion. 1 mM $AgNO_3$ solution was first prepared by dissolving silver nitrate in distilled water. Separately, PVP is dissolved in water to create a uniform solution, aiding in controlling NPs size and providing a stabilizing matrix. The $AgNO_3$ solution is progressively added to the PVP solution while continuously stirring to ensure uniform dispersion of Ag^+ ions. Then, an aqueous extract of *Asparagus racemosus*, prepared as a natural reducing agent, is added dropwise to this mixture.

The reduction of Ag^+ ions by the extract leads to the formation of AgNPs within the PVP matrix, indicated by a color change to yellowish-brown. The mixture is continuously stirred at room temperature until the color change confirms the synthesis of PVP-AgNPs. The NPs are collected by centrifugation at 10,000 rpm for 20 minutes, then washed with distilled water to remove excess PVP and unreacted compounds. The purified PVP-AgNPs are then dried and stored for subsequent analysis and potential applications in biomedical or antimicrobial fields[22].

Characterization: The synthesized PVP-AgNPs were characterized using various analytical methods to understand their optical, morphological and structural properties. UV-visible spectroscopy (Shimadzu-1800) was used to assess optical characteristics over a wavelength range of 300 to 800 nm, identifying specific absorbance peaks indicative of NPs formation. Transmission electron microscopy (TEM) was used using a JEM1010-JEOL model to undertake morphological analysis, including size and shape. A tiny droplet of re-dispersed PVP-AgNPs in water was put on a carbon-coated copper grid and air-dried to ensure imaging stability. Crystallinity was determined by X-

ray diffraction (XRD) on a Miniflex Rigaku-600 at 30 kV and 2 mA, scanning at 10° per minute across a 2θ range of 3° to 90°.

Using Cu-Kα radiation and a graphite monochromator revealed good diffraction patterns, confirming the crystalline structure. FT-IR spectroscopy (Shimadzu-8400) was utilized to identify functional groups on Gymnema sylvestre extract and the surface of PVP-AgNPs. The spectra ranged from 4000 to 500 cm^{-1}, providing insights into molecular interactions and NP stabilization.

Antimicrobial activity assay: Overnight bacterial cultures incubated at 37°C were used to evaluate antibacterial activity. Nutrient agar medium was prepared, distributed into a 100 mL conical flask and sterilized at 121°C for 15 minutes in an autoclave. After sterilization, the medium was put onto Petri dishes. Chloramphenicol was utilized as the positive control. The antibacterial activity of the test extracts was determined using the agar well diffusion technique. Bacterial inoculums were evenly distributed on the agar surface using a sterile glass spreader and four wells were created using a sterile cork borer. The test extracts were applied to the wells in concentrations of 40, 60 and 80 mg/mL. To determine antibacterial efficiency, the width of the inhibition zone surrounding each well was measured after incubating the plates at 37°C for 24 hours[22].

Antioxidant activity test: The antioxidant activity was measured with the DPPH test. For this, 3 mL of the test extract was combined with 1 mL of a 0.1 mmol/L DPPH solution in methanol. The combination was incubated at 37°C for 30 minutes before being measured at 517 nm with a spectrophotometer, using ascorbic acid as a reference. The radical scavenging activity, given as the percentage of inhibition (I), was estimated by comparing the absorbance of the test sample (At) with that of the control (Ac)[22].

$$I = \frac{At}{Ac} \times 100 \quad (1)$$

Results and Discussion

UV-Visible Spectroscopy: The UV-Visible spectrum shows in figure 1 displayed distinct absorbance peaks for the *Asparagus racemosus* extract, AgNPs and PVP-AgNPs while PVP itself exhibited no significant absorption in the visible region. Each component's spectrum offers insight into its optical properties and interactions in the composite structure. The plant extract shows a characteristic absorbance peak around 420 nm, attributed to the presence of phenolic compounds and flavonoids, which have π-π transitions due to their conjugated systems. These compounds are responsible for the plant extract's reducing capability, allowing it to convert Ag^{+} ions into AgNPs in a green synthesis process.

The PVP spectrum shows negligible absorption within the 300-800 nm range, which is expected as PVP lacks chromophores that absorb visible light. PVP does not contain conjugated double bonds or aromatic rings that would enable π-π or n-π transitions. This absence of absorption makes PVP an excellent stabilizing and capping agent, as it does not interfere with the optical properties of the synthesized NPs, allowing clear observation of the NPs absorbance peaks. The AgNPs exhibit a strong absorbance peak around 440 nm a feature attributed to the SPR effect. This peak occurs due to the collective oscillation of electrons on the NPs surface when exposed to light.

The exact position of the SPR peak can vary slightly depending on particle size, shape and surrounding medium, but it typically lies within the 400-450 nm range for small AgNPs. This peak confirms the successful synthesis of AgNPs through reduction by the plant extract[12].

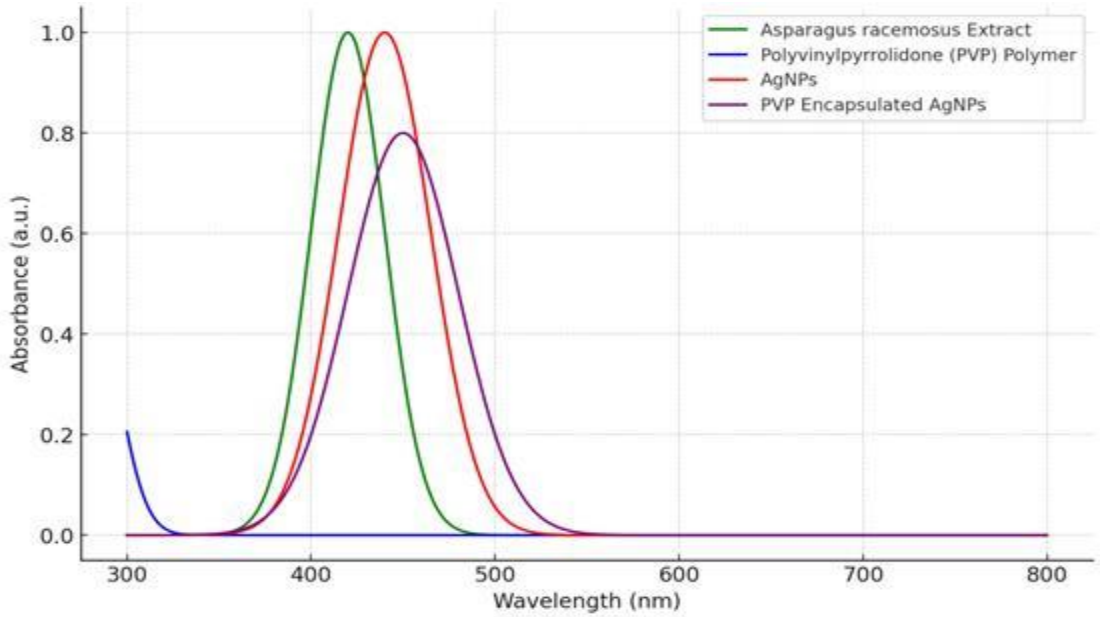

Figure 1: UV-Visible Spectra of *Asparagus Racemosus* Extract, PVP Polymer, AgNPs and PVP-AgNPs

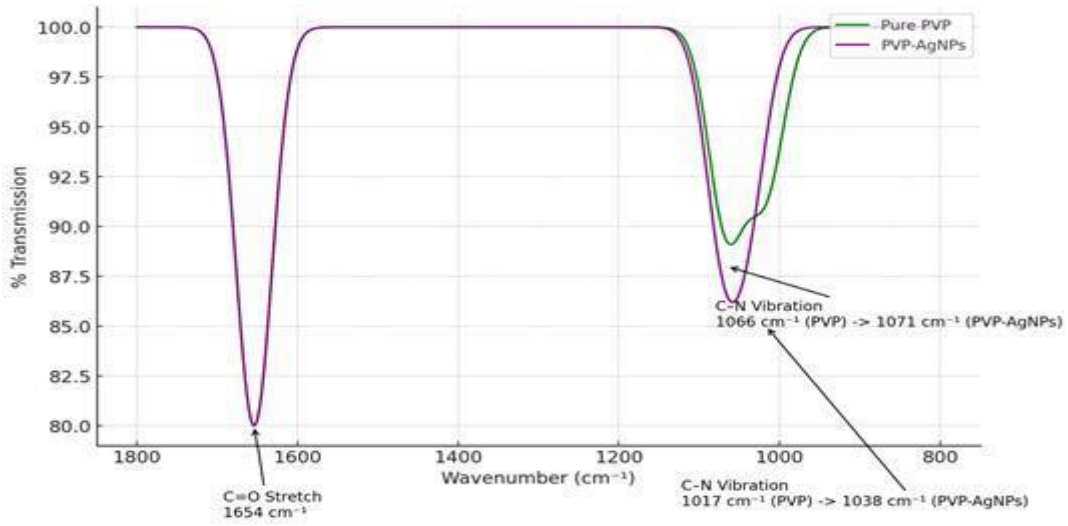

Figure 2: FTIR Spectrum of Pure PVP and PVP-AgNPs

For PVP-AgNPs, the spectrum shows a similar SPR peak at around 450 nm, slightly red-shifted compared to bare AgNPs. This shift occurs due to the surrounding PVP polymer matrix, which alters the dielectric environment around the NPs. Encapsulation with PVP can stabilize the AgNPs, preventing aggregation and offering enhanced control over their optical properties which are useful for biomedical and optical applications[4].

The UV-visible spectrum highlights the optical behavior of each component and confirms successful synthesis and encapsulation. The distinct SPR peak for AgNPs and the slight red-shift for PVP-AgNPs illustrate the influence of PVP encapsulation, while the absence of PVP absorption indicates minimal interference with NPs analysis. This spectrum validates the green synthesis approach, confirming both NPs formation and successful encapsulation in the biocompatible PVP matrix.

Fourier transforms infrared spectroscopy (FTIR): The FTIR analysis of pure PVP and PVP-AgNPs reveals significant insights into the interactions between PVP and AgNPs shows in figure 2. In the FTIR spectrum of pure PVP, a prominent absorption peak is observed at 1654 cm^{-1}, which corresponds to the C=O stretching vibration in the pyrrolidone group. This peak is characteristic of PVP and indicates the presence of carbonyl functional groups. Additionally, two smaller peaks associated with C–N vibrations in the pyrrolidone structure appear at 1066 cm^{-1} and 1017 cm^{-1}. When PVP is used to stabilize PVP-AgNPs, slight shifts in the FTIR spectrum indicate interactions between the polymer and the NPs.

Specifically, the C–N vibration peaks at 1066 cm^{-1} and 1017 cm^{-1} in pure PVP shift to 1071 cm^{-1} and 1038 cm^{-1} in the presence of AgNPs. This red shift is attributed to the involvement of nitrogen electrons from the pyrrolidone ring in the coordination with AgNPs, suggesting a binding interaction[20]. Such interactions can stabilize the NPs by reducing their tendency to agglomerate, enhancing their dispersion in the solution. This stabilization effect is a result of the nitrogen atoms in the pyrrolidone ring coordinating with the silver surface, which is commonly observed when polymers are used as capping agents in NPs synthesis[12].

These spectral changes indicate that PVP effectively stabilizes AgNPs by forming a coordination complex. The interaction between PVP and AgNPs likely involves electron donation from nitrogen, facilitating strong interactions that maintain NPs stability. This finding is consistent with previous studies that highlight the role of nitrogen-containing groups in stabilizing metal NPs, making PVP a suitable stabilizer in AgNPs synthesis. The observed FTIR shifts provide strong evidence of the structural modifications in PVP upon AgNPs incorporation, affirming its role as a capping and stabilizing agent for silver colloids[12].

X-ray Diffraction Spectroscopy (XRD): The XRD spectrum of PVP-AgNPs, as shown in figure 3, confirms their crystalline nature through distinct Bragg reflection peaks observed at 2θ values of 38.49°, 44.62°, 64.85° and 77.68°. These peaks correspond to the (111), (200), (220) and (311) lattice planes of a face-centered cubic (FCC) silver crystal structure, matching the JCPDS Card No. 04-0783. The highest intensity of the (111) plane indicates it as the dominant orientation, a common feature in AgNPs synthesis due to the plane's inherent stability[22].

Notably, the encapsulation of AgNPs with PVP appears to introduce slight peak broadening, which is likely due to the reduced particle size and increased lattice strain associated with polymer encapsulation. The PVP layer surrounding the NPs can slightly hinder crystalline perfection, resulting in broader and somewhat less intense peaks, particularly in the lower-intensity reflections.

This broadening effect also supports the role of PVP as a stabilizing agent, where the polymer's interaction with the AgNPs surface may introduce slight structural modifications without altering the overall FCC crystal structure of silver[18].

The broadening and minor reduction in peak intensity indicate that PVP effectively encapsulates the AgNPs, contributing to their stability and preventing aggregation. This encapsulation likely leverages the nitrogen and oxygen atoms in PVP, coordinating with the silver surface to create a stable colloidal suspension. Thus, the XRD analysis confirms that PVP encapsulation preserves the crystalline integrity of AgNPs while potentially impacting the particle size and stability through slight structural modification, which is favorable for applications requiring long-term stability of AgNPs in solution[22].

Transmission Electron Microscopy (TEM): The TEM analysis of PVP-AgNPs revealed valuable insights into the morphology, distribution and average size of the NPs. As shown in the figure 4, the NPs were well-dispersed and predominantly spherical, indicative of uniform synthesis. The analysis confirmed an average particle size of approximately 33 nm, as reflected in the histogram depicting the size distribution. This relatively consistent particle size supports the successful stabilization and encapsulation of AgNPs by the PVP matrix, preventing significant aggregation and ensuring a homogeneous dispersion within the sample.

The images further emphasized the effective role of PVP in maintaining the colloidal stability of AgNPs. PVP, as a capping agent, facilitated the dispersion by binding to the NPs surfaces, thus reducing surface energy and minimizing the likelihood of particle aggregation. The uniformity of the NPs observed in the TEM micrographs aligns with the Gaussian distribution seen in the size analysis, suggesting that the synthesis process was controlled and reproducible.

This characteristic is essential for applications that demand consistent NPs properties such as biomedical and optical fields[11]. TEM results highlight the efficiency of PVP as a stabilizing agent in synthesizing AgNPs as 33 nm in size on average. The encapsulation by PVP ensured a uniform size distribution and prevented the aggregation of NPs, which is crucial for maintaining their functional properties. The observed morphology and size distribution suggest that the PVP-AgNPs are suitable for applications where stable and dispersed NPs are needed. The uniform particle size, confirmed through TEM analysis, corroborates the findings from UV-Visible and FTIR spectroscopy, providing a comprehensive understanding of the synthesized AgNPs' characteristics.

Antimicrobial Activity: 3% DMSO was used as a negative control to assess the antimicrobial activity of the produced AgNPs; it did not exhibit any inhibitory zones, nor did the plant extract. The study showed that AgNPs' antimicrobial activity rises with concentration. This dose-dependent activity is attributed to enhanced interactions between AgNPs and sulfur-containing proteins in bacterial cell walls disrupting cellular functions and leading to cell death.

Table 1 and figure 5 represent the inhibition zones observed for various AgNPs concentrations. At 1 mg/mL, AgNPs showed significant antibacterial effects with inhibition zones of 14 mm against *E. coli* (a Gram-negative bacterium) and 13 mm against *E. faecalis* (a Gram-positive bacterium). The antibacterial mechanism of AgNPs can be attributed to three main factors. First, there may be an electrostatic interaction between negatively charged AgNPs (stabilized by carboxylate groups) and positively charged bacterial membrane proteins, facilitating attachment. Second, AgNPs may induce physicochemical alterations in the bacterial cell wall, potentially causing leakage of intracellular contents and cell death. Third, AgNPs can penetrate bacterial membranes, hindering essential cellular processes such as DNA replication and respiration, which collectively lead to bacterial death[7].

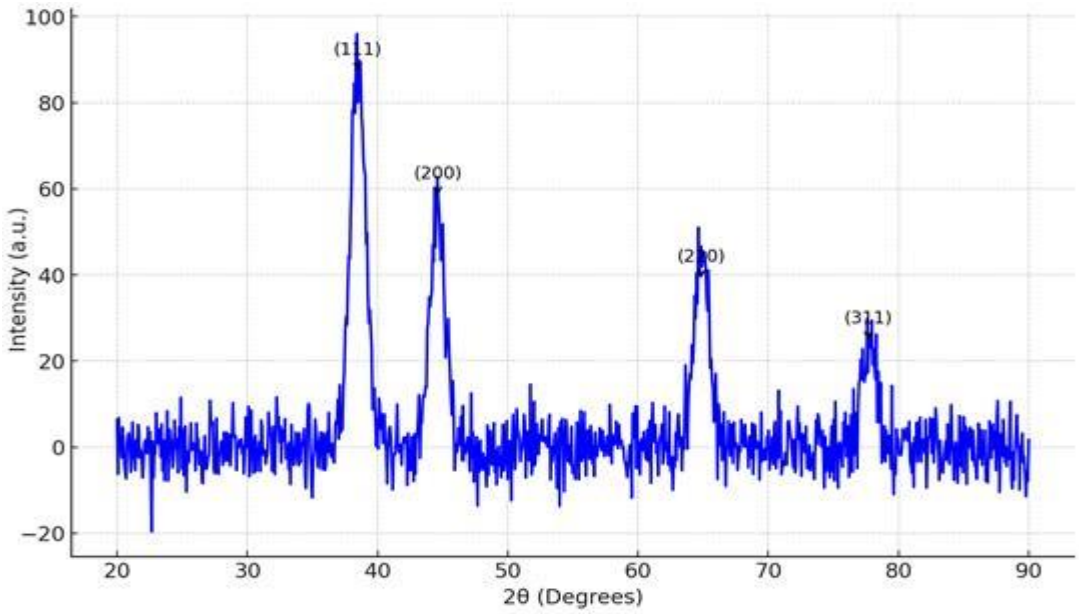

Figure 3: XRD Spectrum of PVP-AgNPs

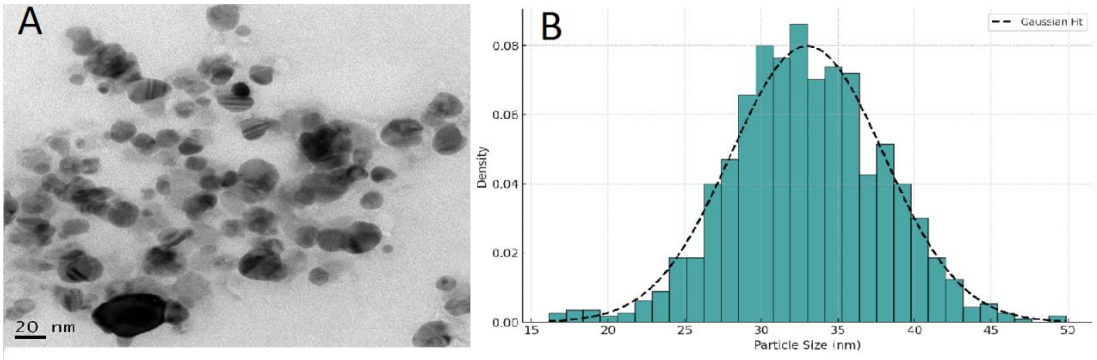

Figure 4: TEM Images of (A) PVP-AgNPs and (B) Particle Size Distribution Curve

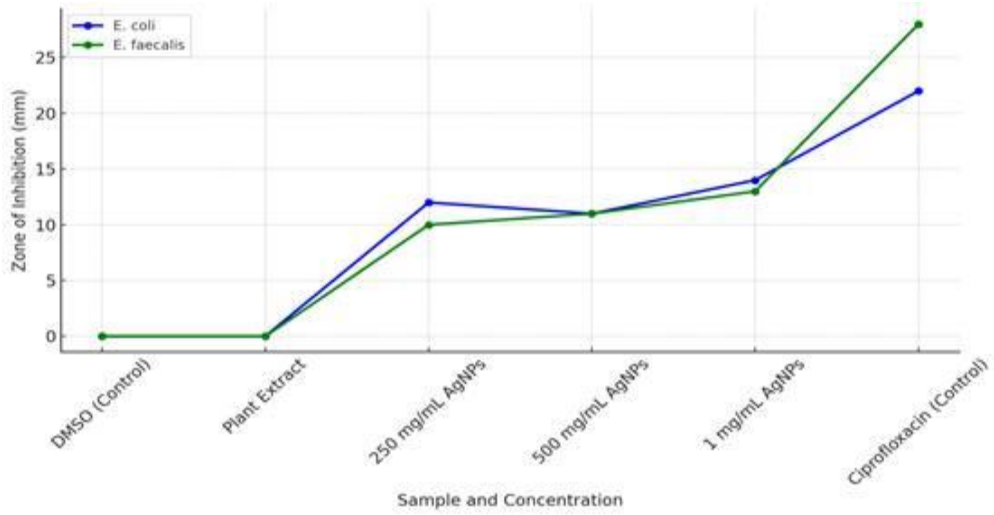

Figure 5: Plot of Antibacterial Activity of PVP-AgNPs for Various Conc. V/S Zone of Inhibition

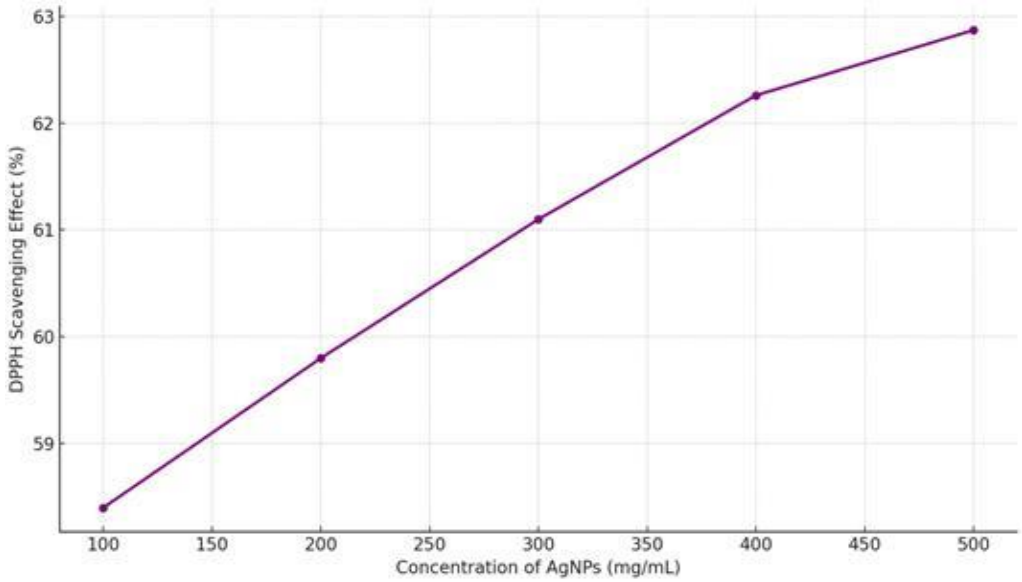

Figure 6: Plot of Antioxidant Efficacy of Various Concentrations of PVP-AgNPs against DPPH

Table 1
Zones of Inhibition for Antibacterial Activity of PVP-AgNPs

Bacteria	DMSO (Negative Control)	Plant Extract	1 mg/mL AgNPs	250 mg/mL AgNPs	500 mg/mL AgNPs	Ciprofloxacin (Positive Control)
E. coli	0	0	14	12	11	22
E. faecalis	0	0	13	10	11	28

Table 2
Antioxidant Efficacy of Various Concentrations of PVP-AgNPs against DPPH

Sample Number	Concentration of AgNPs (mg/mL)	DPPH Scavenging Effect (%)
1	100	58.40 ± 0.01
2	200	59.80 ± 0.02
3	300	61.10 ± 0.04
4	400	62.26 ± 0.03
5	500	62.87 ± 0.02

Antioxidant Activity: The antioxidant activity of synthesized PVP-AgNPs was evaluated using the DPPH assay where PVP-AgNPs served as radical scavengers and DPPH provided a radical source. Upon addition of AgNPs, the DPPH solution color shifted from deep violet to pale yellow, indicating a reduction in DPPH radicals. This visual change, along with a gradual decrease in absorbance at 517 nm, confirms the free radical scavenging ability of AgNPs and suggests a dose-dependent antioxidant effect. This graph illustrates the antioxidant activity of AgNPs as measured by the DPPH scavenging effect shows in figure 6.

As the concentration of AgNPs increases, there is a corresponding increase in the DPPH scavenging percentage, reaching a maximum of 62.87% at 500 mg/mL. This trend confirms the dose-dependent nature of AgNPs' antioxidant activity, highlighting their potential for free radical scavenging. As illustrated in table 2, the antioxidant efficacy of AgNPs increased with concentration. Specifically, at a 500 mg/mL concentration, the DPPH scavenging effect reached 62.87 ± 0.02%, indicating strong antioxidant activity at higher doses. This data supports the dose-dependent antioxidant potential of AgNPs, with an evident increase in DPPH scavenging at antioxidants, particularly at elevated doses[2].

Conclusion

In conclusion, the successful green synthesis of silver nanoparticles (AgNPs) using *Asparagus racemosus* root extract and their encapsulation with polyvinylpyrrolidone (PVP) were confirmed through multiple analytical techniques. UV-Visible spectroscopy identified characteristic peaks, with AgNPs showing a surface plasmon resonance at 440 nm, shifting to 450 nm upon PVP encapsulation, indicating enhanced stability. FTIR analysis revealed interactions between PVP and AgNPs, preventing aggregation, while XRD confirmed a face-centered cubic (FCC) structure with high crystallinity.

The synthesized AgNPs exhibited significant antibacterial activity against *E. coli* and *E. faecalis*, demonstrating their potential as antimicrobial agents. Additionally, DPPH assay results showed strong antioxidant activity, with a dose-dependent free radical scavenging effect. These findings highlight the potential of PVP-encapsulated AgNPs synthesized from *Asparagus racemosus* for biomedical applications, particularly in antimicrobial and antioxidant therapies.

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