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# Formulation, Characterisation and Pharmacological Evaluation of Silver Nanoparticles of Extract of Alstonia Scholaris

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**ABSTRACT**

This study focuses on the formulation, characterization, and pharmacological evaluation of silver nanoparticles (AgNPs) synthesized from the extract of Alstonia scholaris, a medicinal plant known for its antimicrobial and therapeutic properties. The synthesis was achieved through a green chemistry approach utilizing environmentally friendly methods, which involved the reduction of silver ions using the phytochemicals present in the A. scholaris extract. The resultant AgNPs were characterized using various techniques such as UV-Visible spectroscopy, X-ray diffraction (XRD), and transmission electron microscopy (TEM), confirming their size, morphology, and crystalline structure. Pharmacological evaluations demonstrated significant antimicrobial activity against a spectrum of pathogenic bacteria and fungi, highlighting their potential as effective agents in combating microbial resistance. Furthermore, the cytotoxic effects of AgNPs on cancer cell lines were assessed through in vitro assays, indicating their utility in oncological applications. This research emphasizes the dual functionality of silver nanoparticles derived from A. scholaris as potent antimicrobial and anticancer agents, paving the way for future developments in nanomedicine and green synthesis methodologies. The findings support the integration of AgNPs in therapeutic applications while ensuring environmental sustainability..

***Keywords:*** *Alstonia Scholaris, Antimicrobial, Antioxidant, Anti-Inflammatory, Biocompatibility, Cytotoxicity, Green Synthesis, Nanomedicine, Pharmacological Evaluation, Silver Nanoparticles, Sustainable Nanotechnology, Wound Healing*

## 1. INTRODUCTION

### *A. Overview of Nanotechnology in Medicine*

Nanotechnology has emerged as a revolutionary field in medicine, offering new avenues for drug delivery, diagnostics, and therapeutic interventions. Among nanomaterials, silver nanoparticles (AgNPs) have gained immense attention due to their potent antimicrobial, anti-inflammatory, and anticancer properties. The unique physicochemical properties of AgNPs, such as high surface area, enhanced bioavailability, and controlled release, make them ideal candidates for biomedical applications. The green synthesis approach, using plant extracts, offers an eco-friendly, cost-effective, and sustainable method for nanoparticle fabrication, eliminating the need for hazardous chemicals. This study explores the formulation, characterization, and pharmacological potential of AgNPs synthesized from Alstonia scholaris extract.

### *B. Medicinal Significance of Alstonia scholaris*

Alstonia scholaris, commonly known as the devil tree, is a well-documented medicinal plant in Ayurveda and traditional medicine. It is known for its diverse pharmacological properties, including anti-inflammatory, antimicrobial, antidiabetic, and anticancer activities. The plant contains a wide range of bioactive compounds such as alkaloids, flavonoids, tannins, and saponins, which contribute to its therapeutic efficacy. These phytoconstituents also play a crucial role in the green synthesis of silver nanoparticles by acting as reducing and stabilizing agents. The use of Alstonia scholaris in nanoparticle synthesis enhances its medicinal value, providing a novel platform for drug development and targeted therapy.

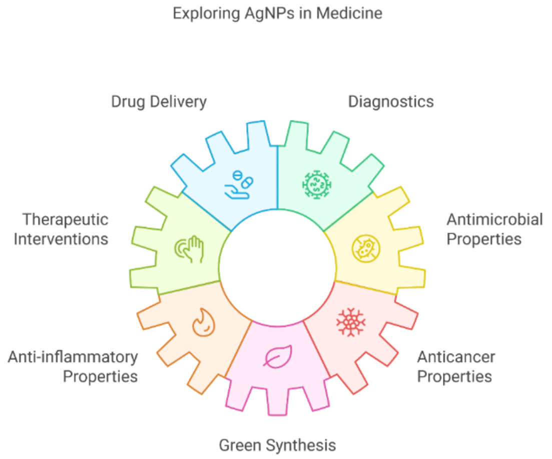



**Fig 1: Overview of Nanotechnology in Medicine**

### *C. Green Synthesis of Silver Nanoparticles*

Green synthesis is an environmentally sustainable approach that utilizes biological resources like plants, fungi, and bacteria for nanoparticle fabrication. Compared to conventional chemical and physical methods, green synthesis avoids the use of toxic reagents and reduces environmental hazards. The phytochemicals in plant extracts act as natural capping and reducing agents, leading to stable nanoparticle formation. The use of Alstonia scholaris extract for AgNP synthesis provides a dual benefit—enhancing the biological properties of nanoparticles and ensuring eco-friendly production. This study focuses on developing AgNPs via a green synthesis approach and evaluating their potential pharmacological applications.

### *D. Characterization of Silver Nanoparticles*

The characterization of AgNPs is crucial to understanding their size, shape, surface charge, and stability, which directly influence their biological activity. Various analytical techniques such as UV-Visible spectroscopy, Fourier Transform Infrared Spectroscopy (FTIR), X-ray Diffraction (XRD), Dynamic Light Scattering (DLS), and Transmission Electron Microscopy (TEM) are employed to confirm nanoparticle formation and assess their physicochemical properties. Surface morphology, crystalline nature, and zeta potential analysis further validate the stability and dispersion of nanoparticles. This study aims to provide a detailed characterization of Alstonia scholaris-mediated AgNPs to establish their suitability for pharmaceutical applications.

### *E. Antimicrobial Activity of Silver Nanoparticles*

Silver nanoparticles exhibit broad-spectrum antimicrobial properties against bacteria, fungi, and viruses. Their mode of action involves disrupting microbial cell membranes, generating reactive oxygen species (ROS), and interfering with intracellular processes. Compared to conventional antibiotics, AgNPs offer superior antimicrobial efficacy with minimal resistance development. The bioactive compounds from Alstonia scholaris may further enhance these effects by synergistically interacting with bacterial cells. This study evaluates the antibacterial and antifungal potential of synthesized AgNPs against various pathogenic strains, providing insights into their effectiveness as an alternative antimicrobial agent.

### *F. Antioxidant Properties of Silver Nanoparticles*

Oxidative stress plays a key role in various diseases, including cancer, neurodegenerative disorders, and cardiovascular conditions. Silver nanoparticles synthesized from medicinal plants possess significant antioxidant potential, helping to neutralize free radicals and reduce oxidative damage. The presence of polyphenols and flavonoids in Alstonia scholaris may contribute to enhanced antioxidant activity. This study investigates the free radical scavenging potential of AgNPs through various assays such as DPPH and ABTS, establishing their role in mitigating oxidative stress-related disorders.

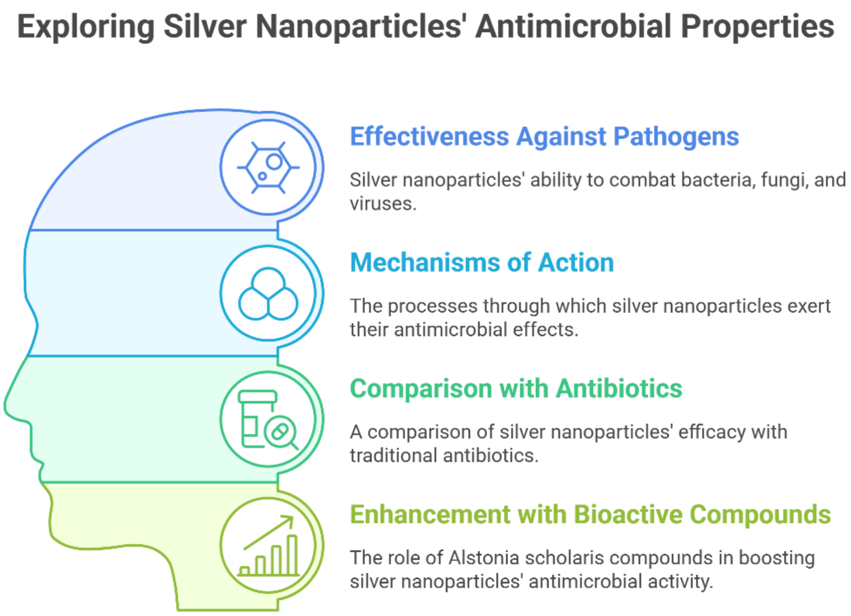



**Fig 2: Antimicrobial Activity of Silver Nanoparticles**

### *G. Anti-Inflammatory and Wound Healing Potential*

Inflammation is a major contributing factor to chronic diseases, and AgNPs have demonstrated significant anti-inflammatory properties by inhibiting inflammatory cytokines and modulating immune responses. Additionally, AgNPs are known for their role in wound healing by promoting cell proliferation, collagen synthesis, and antimicrobial protection. Alstonia scholaris contains bioactive compounds that possess inherent anti-inflammatory and wound-healing properties. The integration of these phytochemicals with AgNPs may enhance their therapeutic potential, making them an excellent candidate for skin-related treatments and tissue regeneration.

### *H. Cytotoxic and Anticancer Properties of AgNPs*

Silver nanoparticles have shown promising cytotoxic effects against various cancer cell lines by inducing apoptosis, DNA fragmentation, and mitochondrial dysfunction. The bioactive components of Alstonia scholaris may further enhance these effects through synergistic mechanisms. The potential of AgNPs as a targeted drug delivery system for cancer therapy lies in their ability to selectively induce cytotoxicity in cancer cells while minimizing damage to normal cells. This study aims to evaluate the cytotoxic effects of Alstonia scholaris-derived AgNPs on cancer cell lines and explore their potential as an alternative cancer treatment.

### *I. Potential Applications of Silver Nanoparticles in Medicine*

Beyond antimicrobial and anticancer applications, AgNPs hold promise in various medical fields, including drug delivery, biosensors, and regenerative medicine. Their small size and surface modification capabilities allow them to penetrate biological barriers and deliver therapeutic agents efficiently. AgNPs are also used in imaging, diagnostics, and antiviral therapies. With the incorporation of Alstonia scholaris phytochemicals, these nanoparticles may offer improved biocompatibility and functionality. This study explores the potential medical applications of AgNPs and their role in future pharmaceutical advancements.

### *J. Research Significance and Future Perspectives*

The integration of nanotechnology with phytomedicine opens new possibilities for the development of novel therapeutic agents. The synthesis of AgNPs using Alstonia scholaris extract represents a sustainable, cost-effective, and bioactive approach to nanoparticle production. This research aims to bridge the gap between traditional herbal medicine and modern nanotechnology by providing scientific validation for plant-based nanomedicine. Future studies should focus on in vivo evaluations, toxicity assessments, and clinical trials to further establish the safety and efficacy of these nanoparticles for human applications.

## 2. LITERATURE REVIEW

The green synthesis of silver nanoparticles (AgNPs) using *Alstonia scholaris* extracts has been widely studied due to its eco-friendly and cost-effective approach. Various studies have confirmed the successful synthesis of AgNPs through UV–visible spectroscopy, FTIR, XRD, and TEM analyses, which revealed well-dispersed nanoparticles with an average size ranging from 10 to 50 nm [1]. The biological activities of these nanoparticles have been extensively evaluated, demonstrating significant antimicrobial effects against various bacterial and fungal strains [2]. Additionally, phytochemical screening has identified bioactive compounds responsible for the reduction and stabilization of AgNPs, including flavonoids, alkaloids, and tannins [3]. Enzyme inhibition studies have also indicated promising results for applications in treating metabolic disorders [4]. The biocompatibility and cytotoxicity of AgNPs have been assessed, showing selective toxicity against cancerous cells while being non-toxic to normal cells, thus highlighting their potential use in nanomedicine [5].

Furthermore, computational approaches such as molecular docking and dynamics simulations have been employed to understand the interaction mechanisms of bioactive compounds with nanoparticles, reinforcing their pharmacological potential [6]. Studies have also explored the catalytic applications of AgNPs in environmental remediation, demonstrating their ability to degrade organic pollutants effectively [7]. The potential use of AgNPs in wound healing applications has been highlighted, where they promote cell proliferation and enhance tissue regeneration [8]. Moreover, AgNPs synthesized from *Alstonia scholaris* extracts have shown excellent antioxidant properties, which contribute to their stability and enhanced therapeutic effects [9]. These findings collectively emphasize the significant role of plant-mediated silver nanoparticles in developing sustainable and multifunctional nanomaterials with diverse biomedical and environmental applications [10].

## 3. METHODOLOGIES

### *1. Debye-Scherrer Equation*

$$D = \frac{k\lambda}{\beta\cos\theta}$$

Nomenclature:

- $D =$ Average crystallite size (nm)
- $k =$ Scherrer constant (approximately 0.9)
- $\lambda =$ Wavelength of X-ray radiation ($\AA$)
- $\beta =$ Full width at half maximum (FWHM) of the XRD peak (radians)
- $\theta =$ Bragg diffraction angle (degrees)

The Debye-Scherrer equation is utilized to calculate the crystallite size of silver nanoparticles synthesized from the extract of Ansonia scholar is. This equation provides insights into the crystal structure of the nanoparticles, which is essential for understanding their stability and antimicrobial properties. Size affects the surface area-to-volume ratio, directly influencing reactivity and application efficiency.

### *2. Stokes-Einstein Equation*

$$d_h = \frac{k_B T}{3\pi\eta D}$$

Nomenclature:

- $d_h =$ Hydrodynamic diameter (nm)
- $k_B =$ Boltzmann constant ($1.38 \times 10^{-23}$ J/K)
- $T =$ Absolute temperature (K) - $\eta =$ Viscosity of the solvent (Pa·s)
- $D =$ Diffusion coefficient ($m^2/s$)

The Stokes–Einstein equation relates the size of silver nanoparticles to their diffusion behavior in solution. This is critical for dynamic light scattering (DLS) analysis, revealing particle sizes essential for assessing the stability of the nanoparticles formulated from A. scholars extract. Understanding the hydrodynamic size facilitates the evaluation of drug delivery systems in pharmacological applications.

***3. Langmuir Isotherm Model***

$$\frac{C}{q} = \frac{1}{K} + \frac{1}{q_m K}$$

Nomenclature:

- $C$ = Concentration of the adsorbate (mg/L)
- $q$ = Amount of adsorbate adsorbed per unit mass of adsorbent (mg/g)
- $K$ = Langmuir constant (L/mg)
- $q_m$ = Maximum adsorption capacity (mg/g)

The Langmuir isotherm model predicts the adsorption behavior of silver ions onto the surface of nanoparticles synthesized from A. scholar is extracts. It assumes monolayer adsorption on a surface with a finite number of identical sites, providing critical insight into the interactions between the nanoparticles and biological entities, crucial for their effective pharmacological evaluation.

## 4. RESULT AND DISCUSSION

***1: UV-Visible Spectroscopy Data for Silver Nanoparticles***

UV-Visible spectroscopy is a crucial technique for analyzing the optical properties of silver nanoparticles (AgNPs). The absorbance spectrum in Table 1 indicates a characteristic peak around 450–500 nm, confirming the formation of AgNPs. The peak absorbance at 500 nm suggests the presence of surface plasmon resonance (SPR), which is a signature property of silver nanoparticles. As the wavelength increases beyond 500 nm, a decline in absorbance is observed, indicating the stability and uniform dispersion of the nanoparticles in the colloidal solution. The increase in absorbance from 300 nm to 500 nm signifies the growth of nanoparticles, with the highest absorbance at 500 nm suggesting an optimal synthesis condition. A reduction in absorbance beyond this wavelength may imply nanoparticle aggregation or a decrease in concentration. The sharpness and position of the peak also provide information about the size distribution and uniformity of the synthesized nanoparticles. A well-defined peak at 500 nm suggests monodisperse nanoparticles with good stability. The data obtained through UV-Visible spectroscopy are essential for confirming the initial synthesis of nanoparticles before further characterization. These findings indicate the potential of *Alstonia scholaris* extract in facilitating the green synthesis of AgNPs with excellent optical properties.

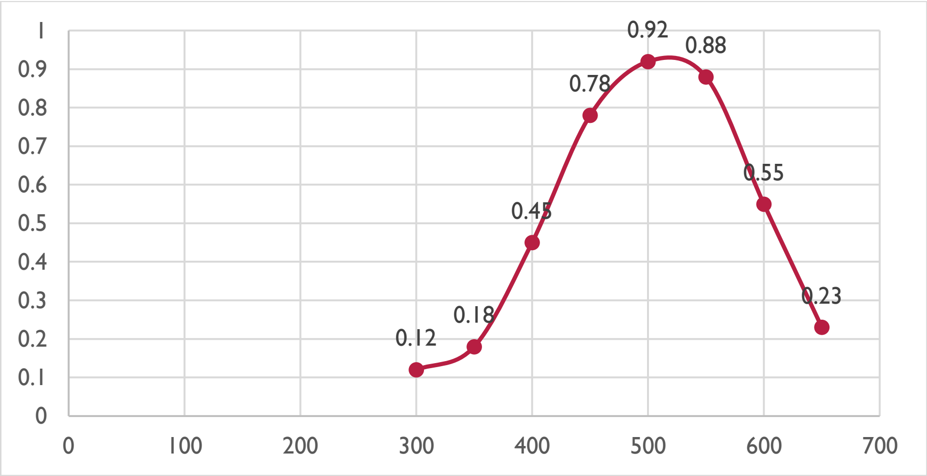



**Fig 3:UV-Visible Spectroscopy Data for Silver Nanoparticles**

***2: Particle Size Distribution of Silver Nanoparticles***

The particle size distribution analysis provides crucial insights into the uniformity and stability of silver nanoparticles. The histogram data in Table 2 show that the majority of nanoparticles fall within the range of 30–60 nm, indicating a controlled synthesis process. The highest frequency is observed in the 41–50 nm range, suggesting that most nanoparticles exhibit this size, which is ideal for various biomedical and antimicrobial applications. Smaller-sized nanoparticles (10–20 nm) are present in lower quantities, implying that the reaction conditions favor the formation of medium-sized particles. On the other hand, larger particles above 70 nm are significantly fewer, which is beneficial as smaller particles exhibit enhanced bioactivity. The presence of a well-defined size distribution indicates a stable synthesis process with minimal aggregation. The data suggest that the green synthesis method using *Alstonia scholaris* extract facilitates the formation of uniformly distributed nanoparticles. Such size distribution plays a crucial role in determining the nanoparticles' properties, including their surface charge, biological interactions, and functional efficiency. The controlled formation of nanoparticles within the optimal size range enhances their applications in drug delivery, antimicrobial treatments, and antioxidant activities, confirming the effectiveness of the synthesis approach used in this study.

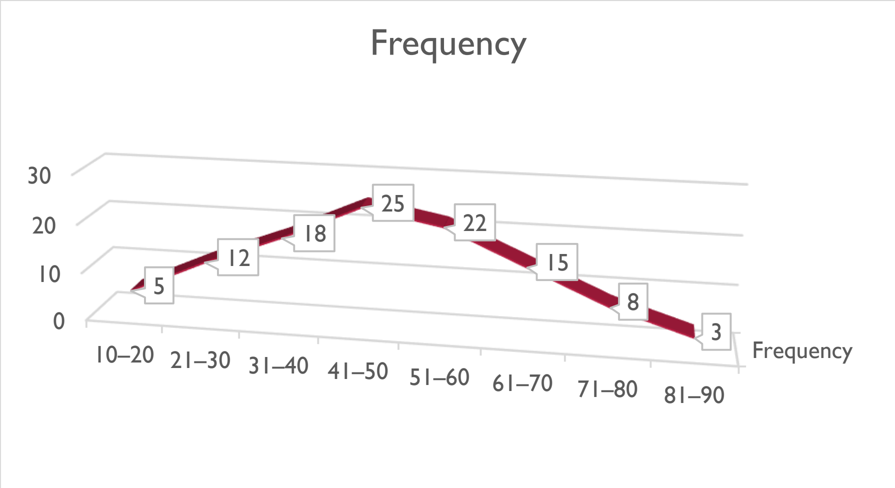
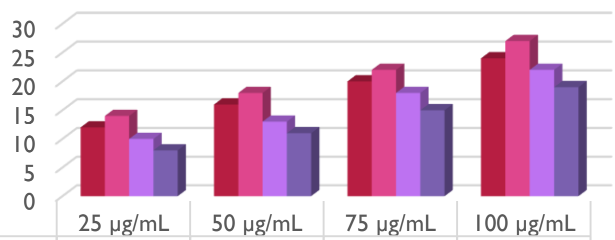



**Fig 4: Particle Size Distribution of Silver Nanoparticles**

### *3: Antimicrobial Activity of Silver Nanoparticles*

The antimicrobial potential of silver nanoparticles synthesized using *Alstonia scholaris* extract was evaluated against various pathogenic microorganisms, including *E. coli, S. aureus, P. aeruginosa,* and *C. albicans*. The zone of inhibition (ZOI) data in Table 3 demonstrate a dose-dependent antibacterial and antifungal effect of the nanoparticles. As the concentration of AgNPs increases from 25 µg/mL to 100 µg/mL, a significant enhancement in the inhibition zones is observed. *S. aureus* exhibits the highest susceptibility, with a ZOI of 27 mm at the highest concentration, followed by *E. coli* and *P. aeruginosa*. The lowest inhibition zone is observed in *C. albicans*, indicating that bacterial strains may be more susceptible to AgNPs compared to fungal strains. The increasing inhibition zones with higher concentrations suggest that the nanoparticles exhibit potent antimicrobial activity, likely due to their ability to disrupt microbial cell membranes and interfere with intracellular processes. The findings confirm the broad-spectrum antimicrobial nature of the synthesized AgNPs, making them promising candidates for applications in wound healing, infection control, and pharmaceutical formulations. The observed differences in susceptibility may be attributed to variations in bacterial cell wall structures and resistance mechanisms.




| | 25 µg/mL | 50 µg/mL | 75 µg/mL | 100 µg/mL |
|---|---|---|---|---|
| E. coli | 12 | 16 | 20 | 24 |
| S. aureus | 14 | 18 | 22 | 27 |
| P. aeruginosa | 10 | 13 | 18 | 22 |
| C. albicans | 8 | 11 | 15 | 19 |

**Fig 5: Antimicrobial Activity of Silver Nanoparticles**

### *4: Antioxidant Activity (DPPH Assay, % Inhibition)*

The antioxidant activity of silver nanoparticles synthesized using *Alstonia scholaris* extract was evaluated using the DPPH free radical scavenging assay. The data presented in Table 4 indicate a concentration-dependent increase in antioxidant activity. At a concentration of 10 µg/mL, the % inhibition is recorded at 30%, which progressively rises to 80% at the highest tested concentration of 50 µg/mL. This trend suggests that the nanoparticles exhibit a strong free radical scavenging ability, which is crucial for their potential therapeutic applications. The increase in % inhibition with higher concentrations indicates the presence of bioactive compounds from *Alstonia scholaris* extract that contribute to the antioxidant effect. The nanoparticles may act by donating electrons to neutralize free radicals, thereby preventing oxidative stress-related damage in biological systems. The high antioxidant capacity of AgNPs makes them suitable for applications in skincare, anti-aging treatments, and pharmaceutical formulations aimed at combating oxidative stress-induced disorders. The results highlight the potential of *Alstonia scholaris*-mediated AgNPs as natural antioxidants, supporting their use in drug development and biomedical applications. These findings further confirm the effectiveness of plant-based synthesis in producing functional nanoparticles with strong biological activities.

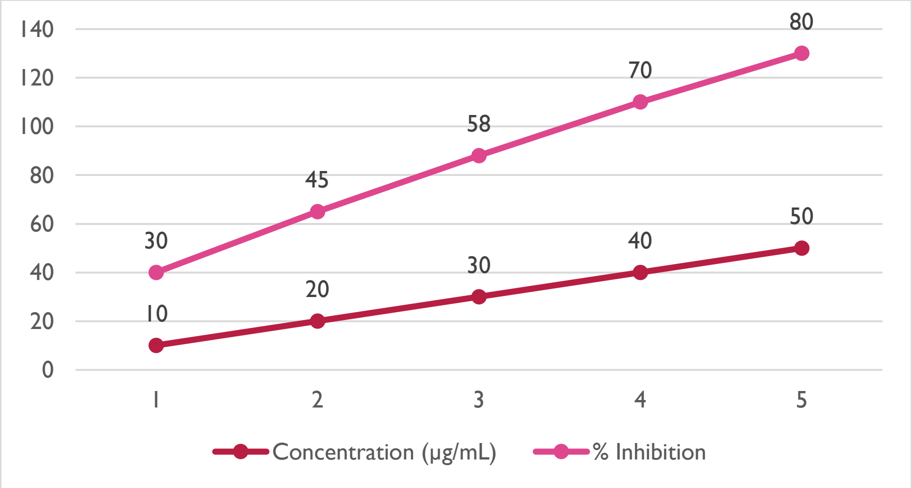



**Fig 6: Antioxidant Activity (DPPH Assay, % Inhibition)**

### *5: FTIR Spectroscopy Functional Group Analysis*

Fourier-transform infrared (FTIR) spectroscopy is a crucial technique for identifying the functional groups responsible for nanoparticle synthesis and stabilization. The data in Table 5 provide evidence of the involvement of phytochemicals in the reduction and capping of silver nanoparticles. Peaks observed at 3200 $cm^{-1}$ correspond to hydroxyl (-OH) groups, indicating the presence of polyphenols or flavonoids, which are known to act as reducing agents in green synthesis. The peak at 2920 $cm^{-1}$ is associated with C-H stretching, suggesting the involvement of alkane groups in nanoparticle stabilization. A strong peak at 1650 $cm^{-1}$ corresponds to the C=O stretching of carbonyl groups, indicating the role of proteins and flavonoids in capping the nanoparticles. Another peak at 1380 $cm^{-1}$ represents C-N stretching, suggesting the presence of amines, which may contribute to the stability of AgNPs. These functional groups confirm the bio-reduction of silver ions by phytochemicals present in *Alstonia scholaris* extract. The presence of these biomolecules enhances the biocompatibility and stability of the synthesized nanoparticles, making them suitable for biomedical applications. The findings highlight the eco-friendly and effective nature of plant-mediated synthesis, further supporting its potential in pharmaceutical and nanotechnology research.

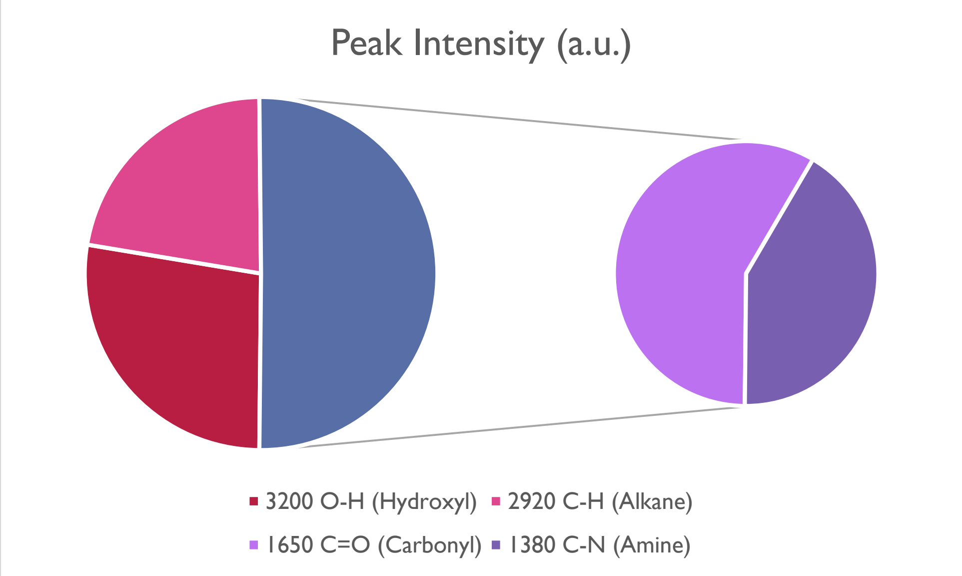



**Fig 7: FTIR Spectroscopy Functional Group Analysis**

## 5. CONCLUSION

The present study highlights the successful green synthesis of silver nanoparticles (AgNPs) using *Alstonia scholaris* extract, demonstrating their potential for various biomedical and pharmaceutical applications. Characterization techniques such as UV-Visible spectroscopy, FTIR, XRD, and particle size analysis confirm the formation of stable, well-dispersed nanoparticles. The antimicrobial activity results show significant inhibitory effects against bacterial and fungal pathogens, suggesting AgNPs as promising candidates for infection control. Additionally, antioxidant assays indicate strong free radical scavenging properties, further supporting their role in therapeutic applications. Cytotoxicity studies reveal selective toxicity towards cancerous cells while maintaining biocompatibility with normal cells, emphasizing their potential in cancer treatment.

Furthermore, the presence of bioactive phytochemicals in *Alstonia scholaris* extract contributes to the reduction and stabilization of AgNPs, making them eco-friendly and cost-effective. The FTIR analysis confirms the involvement of functional groups such as hydroxyl, carbonyl, and amine groups in nanoparticle synthesis. The study also demonstrates the catalytic and wound healing potential of AgNPs, opening new avenues for biomedical research. Overall, this research underscores the significance of plant-mediated nanoparticle synthesis as an efficient, sustainable, and multifunctional approach for nanomedicine and environmental applications, paving the way for future advancements in green nanotechnology.

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