

OPTIMIZING THE GREEN SYNTHESIS OF SILVER NANOPARTICLES WITH POMEGRANATE EXTRACT FOR ANTIBACTERIAL PURPOSES: AN INNOVATIVE APPROACH

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INTRODUCTION

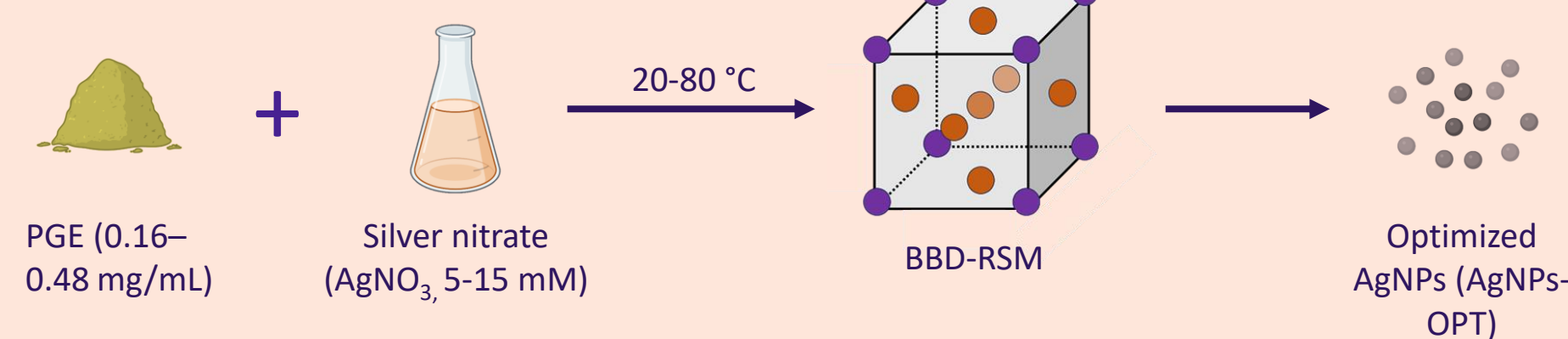
Silver nanoparticles (AgNPs) have attracted significant attention due to their remarkable antimicrobial and biocidal properties. Green synthesis, which utilizes plant extracts as reducing and stabilizing agents, offers an eco-friendly alternative to traditional methods, reducing toxicity and enhancing biocompatibility. Among natural sources, pomegranate (*Punica granatum*) peel extract (PGE) is particularly rich in antioxidant compounds such as ellagitannins and polyphenols, which not only facilitate nanoparticle synthesis but also enhance their biological activity.

This study focuses on optimizing the green synthesis of AgNPs using PGE through a Box–Behnken response surface methodology (BBD-RSM) to refine key parameters for enhanced reproducibility and efficiency. The synthesized AgNPs were fully characterized, and their antimicrobial properties were evaluated.



METHODS

AgNP synthesis:



PGE characterization:

- Phenolic Content Determination
- Total Flavonoids Quantification
- High-Performance Liquid Chromatography (HPLC)
- Antioxidant determination
- Protein Quantification
- Total Reducing Sugars' Quantification

AgNPs characterization:

- Scanning electron microscopy (FESEM)
- UV-Vis Spectrophotometry
- Dynamic Light Scattering (DLS)
- Fourier Transform Infrared (FTIR) Spectroscopy
- X-Ray Diffraction Analysis (XRD)
- Antibacterial Assays

RESULTS

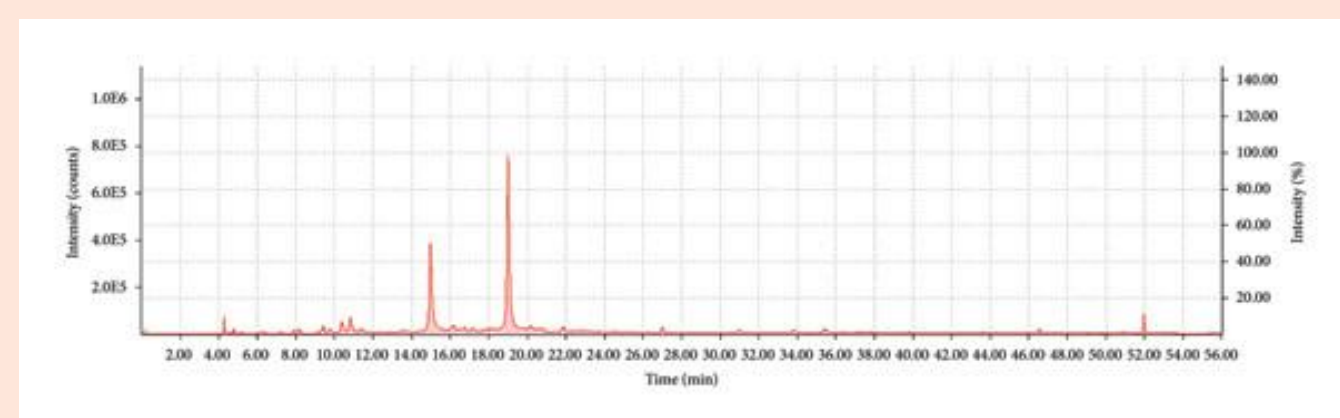


Figure 1. PGE (1 mg/mL) chromatogram obtained by HPLC. Punicalagin represents $29.45 \pm 1.76\%$ w/w of total dry weight (dw).

Table 1. Biochemical composition and antioxidant activity of the PGE employed to synthesize the AgNPs.

Polyphenolic content	29.94 ± 0.36 g GA eqs./100g dw
Flavonoid content	5.15 ± 0.33 g quercetin eqs./100g dw
Antioxidant capacity	1379 ± 40 mmol TROLOX eqs./100g dw
Total reducing sugars	57.2 g glucose eqs./100g dw
Protein content	0.84 ± 0.07 g BSA eqs./100 g dw

AgNPs were synthesized using BBD-RSM to optimize three responses simultaneously: hydrodynamic diameter (HDD), polydispersity index (PDI) and Z-potential).

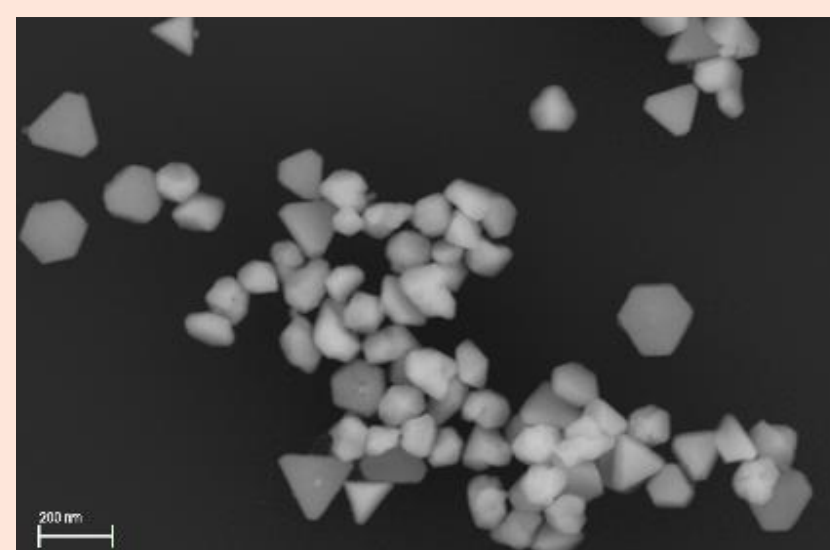


Figure 2. FESEM images of AgNPs-OPT. Scale bar: 200 nm.

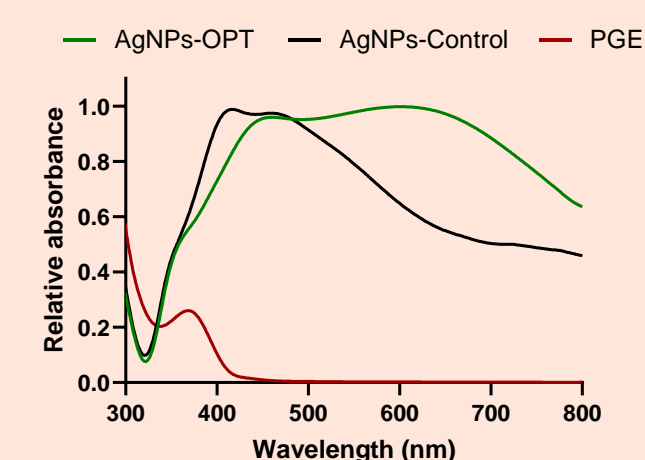


Figure 3. UV-vis spectra showing a surface plasmon resonance (SPR) band around 450 nm.

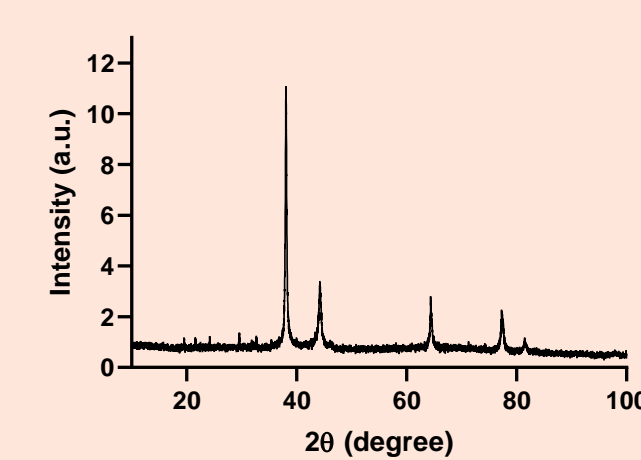


Figure 4. XRD pattern of AgNPs-OPT, which confirmed their crystalline nature.

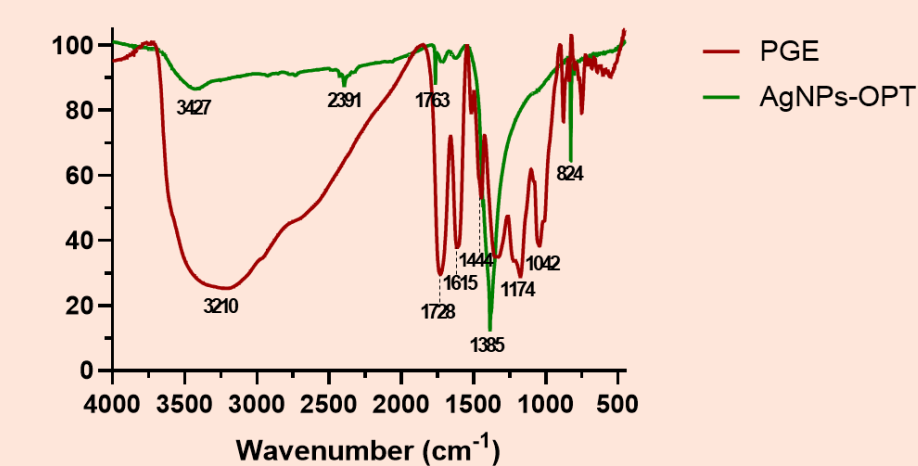


Figure 5. FTIR spectra showing functional groups of the biomolecules involved in the capping and stabilization of the AgNPs-OPT

Finally, antibacterial activity was evaluated against *Escherichia coli* and *Staphylococcus aureus*, and the minimum bactericidal concentrations (MIC) were 5.0 and 2.5 $\mu\text{g/mL}$, respectively.

CONCLUSIONS & FUTURE WORK

- Green synthesis of AgNPs was successfully optimized using BBD-RSM.
- AgNPs optimized HDD, PDI and ZP values simultaneously.
- Characterization confirmed correct synthesis and a good fit of the BBD-RSM model.
- AgNPs exhibited high antibacterial activity against *E. coli* and *S. aureus*.
- Future research will focus on understanding how AgNP characteristics influence their antibacterial mechanism and refining synthesis parameters to enhance antimicrobial effectiveness.

REFERENCES

Díaz-Puertas, R., et al, An Innovative Approach Based on the Green Synthesis of Silver Nanoparticles Using Pomegranate Peel Extract for Antibacterial Purposes, *Bioinorg. Chem. Appl.*, 2025, 2009069, 16.

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