Josip Juraj Strossmayer University of Osijek
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ODJEL ZA KEMIJU

PHYTO-MEDIATED SYNTHESIS OF AMORPHOUS SELENIUM NANOPARTICLES USING WALNUT (*Juglans regia* L.) SEPTUM EXTRACT

Valentina Pavić[1], Zorana Katanić[1], Liljana Krstin[1], Lovro Mihajlović[1], Anamarija Perić[2], Stjepan Šarić[2], Martina Šrajer Gajdošik[2], Elvira Kovač-Andrić[2], Vlatka Gvozdić[2]

[1]Department of Biology, Josip Juraj Strossmayer University of Osijek, Ulica cara Hadrijana 8/A, 31000 Osijek, Croatia

[2]Department of Chemistry, Josip Juraj Strossmayer University of Osijek, Ulica cara Hadrijana 8/A, 31000 Osijek, Croatia

INTRODUCTION

Selenium nanoparticles (SeNPs) have attracted increasing attention due to their biological activity, biocompatibility, and lower toxicity compared with some inorganic selenium forms. Amorphous selenium is particularly interesting due to its potentially higher biological availability. Green synthesis provides a sustainable approach to SeNP production by using plant-derived compounds as natural reducing and stabilizing agents. Walnut (*Juglans regia* L.) fruit septum is an agro-industrial by-product rich in bioactive compounds suitable for nanoparticle synthesis. This study aimed to synthesize and characterize SeNPs using walnut fruit septum extract and evaluate their antibacterial activity against selected bacterial strains.

MATERIALS AND METHODS

Walnut (*Juglans regia* L.) fruit septum extract was used as a natural reducing and stabilizing agent for selenium nanoparticle synthesis. Sodium selenite (Na_2SeO_3) served as the selenium precursor. The obtained SeNPs were characterized by UV–Vis spectroscopy, FT-IR spectroscopy, and PXRD analysis.Antibacterial activity was evaluated against four standard bacterial strains: Gram-positive *Bacillus subtilis subsp. spizizenii* ATCC 6633 and *Staphylococcus aureus* ATCC 6538, and Gram-negative *Escherichia coli* ATCC 35218 and *Pseudomonas aeruginosa* ATCC 27853. Minimum inhibitory concentrations (MICs) of SeNPs and walnut septum extract were determined by a broth microdilution method, with amikacin used as a positive control.

RESULTS

The formation of SeNPs was confirmed by UV–Vis, FT-IR, and PXRD analyses. The UV–Vis spectrum showed a characteristic absorption maximum at approximately 259 nm, supporting successful SeNP formation (Figure 1). FT-IR analysis revealed a broad band at 3000–3500 cm^{-1} and changes in the fingerprint region, indicating the involvement of functional groups from walnut septum extract in selenium reduction and nanoparticle stabilization (Figure 2). PXRD analysis showed a broad diffraction halo in the 20–30° 2θ region without distinct crystalline peaks, confirming the predominantly amorphous nature of the synthesized SeNPs (Figure 3).

The synthesized SeNPs showed pronounced antibacterial activity, particularly against *Bacillus subtilis* (MIC 0.0078 mg mL^{-1}) and *Staphylococcus aureus* (MIC 0.1250 mg mL^{-1}), followed by *Escherichia coli* (MIC 0.5000 mg mL^{-1}). No inhibition of *Pseudomonas aeruginosa* was observed at the highest tested SeNP concentration (1 mg mL^{-1}). In comparison, the walnut septum extract showed substantially weaker antibacterial activity, with MIC values of 3.7500 and 7.5000 mg mL^{-1} against *S. aureus* and *B. subtilis*, respectively, and no inhibition of *E. coli* or *P. aeruginosa* at 30 mg mL^{-1} (Table 1).

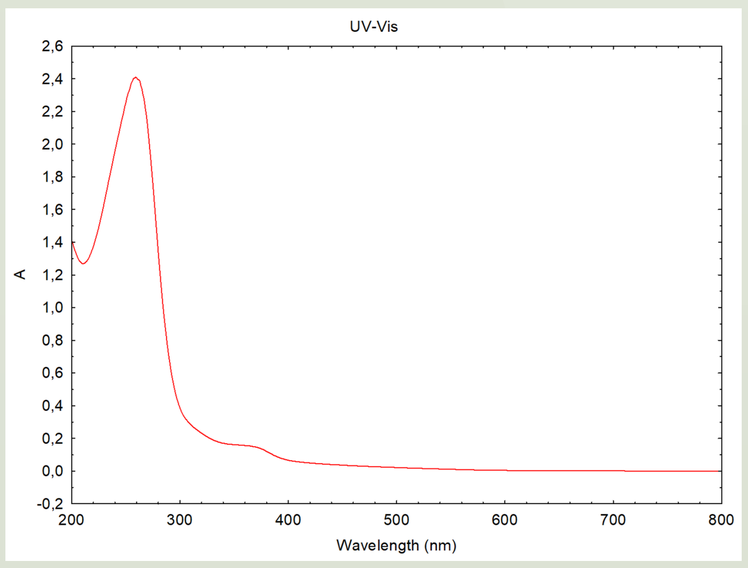

Figure 1. UV–Vis absorption spectrum of selenium nanoparticles synthesized using walnut fruit septum extract.

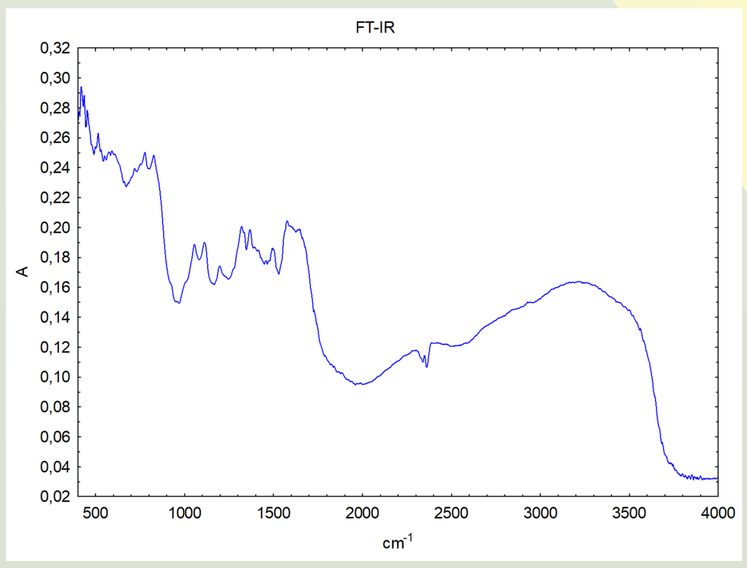

Figure 2. FT-IR spectrum of selenium nanoparticles synthesized using walnut fruit septum extract.

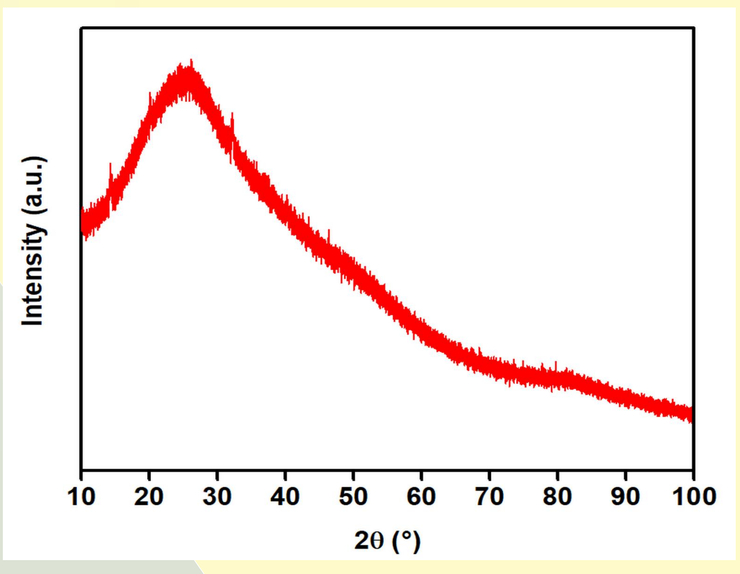

Figure 3. PXRD pattern of the synthesized selenium nanoparticles.

Table 1. Minimum inhibitory concentrations (MICs) of SeNPs, walnut septum extract, and amikacin.

	MIC mg mL^{-1}			
	Escherichia coli	*Pseudomonas aeruginosa*	*Staphylococcus aureus*	*Bacillus subtilis*
SeNPs	0.5000	> 1	0.1250	0.0078
Walnut septum extract	> 30	> 30	3.7500	7.5000
Amikacin	0.0039	0.0078	0.0020	0.0010

>1 and >30 indicate that no growth inhibition was observed at the highest tested concentration of SeNPs (1 mg mL^{-1}) and extract (30 mg mL^{-1}), respectively.

CONCLUSION

Walnut fruit septum extract was successfully used as a natural reducing and stabilizing agent for the green synthesis of predominantly amorphous SeNPs. The synthesized SeNPs exhibited marked antibacterial activity, particularly against *B. subtilis* and *S. aureus*, and were considerably more active than the walnut septum extract. These findings demonstrate the potential of walnut fruit septum as a sustainable source for producing biologically active SeNPs and support further investigation of their antimicrobial applications.