



## Short communication

# Synthesis and characterization comparison of peanut shell extract silver nanoparticles with commercial silver nanoparticles and their antifungal activity



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## ABSTRACT

Peanut shell extract mediated synthesis of silver nanoparticles (AgNPs), their characterization compared to commercial AgNPs, and the antifungal activity of synthesized and commercial AgNPs were studied. Ultraviolet–Visible spectra (450 nm), X-ray diffraction peaks ( $2\theta = 39.26, 45.54, 65.12, \text{ and } 79.21$ ), and Fourier transform infrared spectroscopy confirmed that both AgNPs were similar. High resolution-transmission electron microscopy showed that both the nanoparticles were mostly spherical and oval in shape, with an average particle size of 10 to 50 nm. Both the AgNPs (30  $\mu\text{g}$ ) showed a 5 to 6 mm zone of inhibition against the agricultural fungal pathogens *Phytophthora infestans* and *Phytophthora capsici*.

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## Introduction

Among metal-based nanoparticles, silver nanoparticles (AgNPs) have been the most widely recognized due to their wide ranges of applications, including agricultural sciences, based on their specific characteristics, such as size, shape, and orientation [1]. Recent enhancements were realized using AgNPs with high antimicrobial activity, which can be obtained through physical, chemical, phytochemical, biological, and agricultural waste modes of synthesis [2–4]; the agricultural waste mode of AgNP synthesis has a special application because it's less toxic, more environmentally friendly, and cheaper than other modes of synthesis [5,6]. Lee et al. [5] and Ahmad et al. [6] suggested the probable mechanism of the reduction of  $\text{Ag}^+$  ions by NADH-dependent nitrate reductase.

Recently, fungal diseases like *Phytophthora infestans* is an oomycete that causes the serious potato disease known as late blight or potato blight and *Phytophthora capsici* is also belongs to oomycete plant pathogen that causes blight and fruit rot of

peppers and other important commercial crops will decreased plant yields, especially as said overhead fungi belonging to the oomycete class; oomycetes have become major pathogens of agriculture crops, causing several blights. These blights are due to the development of fungicide resistance, making it difficult to control the diseases [5]. In response, AgNPs are being researched as antifungal agents, as they are highly toxic to most fungi. Woo et al. [7] and He et al. [8] previously reported that AgNPs damage fungal hyphae, causing cytoplasm leakage, inhibited conidial development, and distorted conidiophores. Earlier Kim et al. [9] studied the fungicidal properties of chemical synthesized nano-size silver colloidal solution used as an agent for antifungal treatment of various plant pathogens. Lamsal et al. [10] used AgNPs instead of using commercial fungicide to control *Colletotrichum* sp responsible for pepper anthracnose in pepper plant. Patel et al. [23] explained about the use of nano fungicides based on silver nano emulsion tackle fungal pathogen problems in agriculture and hold promise in the future. Thus, the objectives of the present study were to synthesize AgNPs using peanut shell extract (PNse), an agricultural waste, and compare the structures and antifungal activity of synthesized AgNPs to commercially-available AgNPs (C-AgNPs).

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## Materials and methods

### Synthesis of AgNPs

Peanut shells (50 g) were washed thoroughly with distilled water, dried, blended, and boiled in 200 ml of sterile Nanopure water (conductivity =  $18 \mu\Omega/\text{m}$ , TOC < 3 ppb; Barnstead, Waltham, MA, USA) for 30 min in a heating mantle to obtain the extract, followed by filtration. Five ml of extract was added to 45 ml of  $10^{-3}$  M silver nitrate ( $\text{AgNO}_3$ ; Sigma-Aldrich, St. Louis, MO, USA). Sim et al. [11] reported the composition of peanut shell extract are as follows, total dietary fiber (58.8 to 78.2 g), protein (5.8 to 6.1 g), ash (6.6 to 21.7 g), total phenolic (175 to 431 mg), total saponins (10.8 to 23.4 mg) and phytic acid (108 to 159 mg) and alkaloid (5 to 23.8 g) per 100 g dry weight samples. Reduction of silver ions in the reaction mixture was monitored by the change in color of the reaction mixture from light yellow to dark brown with a yellow shade. To separate the reaction product (silver nanoparticles), it was subjected to centrifugation at 13,000 rpm for 15 min, followed by freeze drying.

### Characterizations of AgNPs

The nanoparticles obtained from PNse at ambient temperature were scanned after the color change using a UV–vis spectrophotometer (UV-1800, Shimadzu, Kyoto, Japan) within a working wavelength range of 300 to 700 nm using a dual beam operated at 1 nm resolution. Fourier transform infrared spectroscopy (FTIR) spectra of AgNPs and PNse were obtained with a Perkin-Elmer FTIR spectrophotometer (Norwalk, CT, USA) in the diffuse reflectance mode at a resolution of  $4 \text{ cm}^{-1}$  in KBr pellets. High resolution transmission electron microscopy (HRTEM; JEM-2010HR, JEOL, USA) was then used to examine the surface morphologies and sizes of the PNse synthesized AgNPs and commercially-available AgNPs (C-AgNPs) (silver nanopowder, <100 nm, 99.5% metals; Sigma-Aldrich, USA). X-ray powder diffraction of both the samples was obtained using a Rigaku X-ray diffractometer (XRD; Rigaku, Japan).

### Antifungal activity

Agricultural fungal pathogens *P. infestans* (KACC 40718) and *P. capsici* (KACC 40475) were procured from the Korean Agriculture Culture Collection, Suwon, South Korea, and the cultures were maintained on Potato Dextrose Agar. The antifungal activity of the PNse-AgNPs and C-AgNPs were determined according to the method of Lee et al. [5], using Nystatin as a control.

### Statistical analyses

Statistical analysis was done using “SPSS Statistics 20.0” software (IBM Corporation, Armonk, NY, USA). Experiments were conducted as duplets and repeated five times. Each experiment was performed in triplicate and the final values were presented as the mean  $\pm$  the standard deviation (SD).

## Results and discussion

The reaction mixture turned from light yellow to dark brown, indicating  $\text{AgNO}_3$  reduction, within 10 min, due to the excitation of surface plasmon vibrations within the synthesized AgNPs [3,6]. The absorbance band of the reduced silver sample showed a peak at 450 nm (Supplementary Fig. 1) and its associated materials. The UV absorbance at 450 nm was in accordance with a previous report [12–14].

The FTIR spectra of PNse-AgNPs and C-AgNPs are shown in Fig. 1a and b. Ossman et al. [15] explained that the presence of

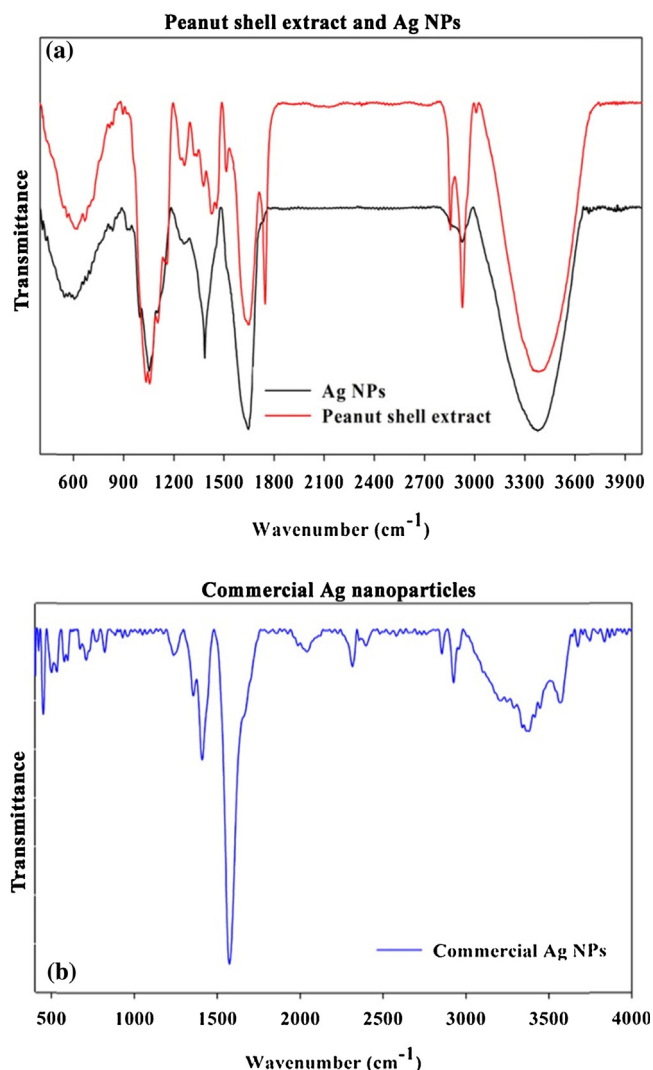


Fig. 1. FTIR spectra of PNse, PNse-AgNPs, and C-AgNPs.

various peaks in PNse shows its complex nature (Fig. 1a). The peak at  $3388 \text{ cm}^{-1}$  is attributed to the surface hydroxyl groups ( $-\text{OH}$ ), due to the presence of alcoholic, phenolic, amino, and carboxylic derivatives. The peaks at  $2920 \text{ cm}^{-1}$  and  $2138 \text{ cm}^{-1}$  are assigned to the C–H asymmetrical stretching bands of aromatics, aliphatics, and olefins and the C–C stretching band of the alkyne group. The peaks at  $1629 \text{ cm}^{-1}$  indicate C=O carboxyl compound, C=C ascribed to ketones, aldehydes alkenes, ester, and aromatic groups. The spectral results of PNse-AgNPs were shown at the  $3410$  and  $2900 \text{ cm}^{-1}$  peaks, which are assigned to  $-\text{OH}$  and  $-\text{CH}$  stretching, respectively. The other peak at  $1600 \text{ cm}^{-1}$  was due to an amide group,  $1382 \text{ cm}^{-1}$  was assigned to  $-\text{CN}-$  stretching amines, and  $1020 \text{ cm}^{-1}$  was assigned to the C–N stretching vibration of the amine group. Fig. 1b shows the C-AgNP's peak at  $1658 \text{ cm}^{-1}$ , indicating that the C=O bond may occur due to bond weakening as a result of the partial bond formation with the surfaces of AgNPs. Moreover, the peaks at  $1013$  and  $1074 \text{ cm}^{-1}$  are due to a C–N bond red shift to  $1024$ – $1043 \text{ cm}^{-1}$  and  $1074$ – $1120 \text{ cm}^{-1}$  for the N–OH complex. The peaks at  $1052 \text{ cm}^{-1}$  result from the coordination of Ag nanopowder with N and O atoms of C=O and C–N bonds. TEM was employed to visualize the size and shape of both the AgNPs. TEM images of PNse-AgNPs and their particle size distribution, fringe pattern, and SAED are shown in Fig. 2a–d, and for C-AgNPs in Fig. 2e–h, respectively. Both the NPs were circular and oval, with smooth edges (Fig. 2a and e), and the size of the particles ranged

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