FACILE GREEN ROUTE SYNTHESIS OF SILVER NANOPARTICLES USING NATURAL POLYMER AND THEIR ANTIBACTERIAL ACTIVITY

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A facile, green route method has been developed for the synthesis of silver nanoparticles. A single step ecofriendly approach has been employed usinga natural polymer, okara juice (*Abelmoschus esculentus*) as a binding agent, along with mandelic acid as reducing agent. UV-visible absorption spectrum recorded for the solution has shown significant bands in the range of 200nm to 600nm.The morphology of silver nanoparticles was observed by FE-SEM and Transmission electron microscopy (TEM). The prepared silver nanoparticles showed unique anti-microbial activity against gram-negative bacteria, such as *Escherichia coli*, and gram-positive bacteria, such as *Staphylococcus aureus*.

Keywords: biopolymer binder, ecofriendly synthesis, silver nanoparticles, antibacterial activity

INTRODUCTION

Nanoparticles are of extreme research interest, as they bridge the gap between bulk materials and atomic or molecular structures. The synthesis of nanoparticles has been extensively studied mainly owing to their advantageous applications.[1-2] The properties of nanomaterials strongly depend upon the size and morphology of nanoparticles, theirinteractions with stabilizers and surrounding media.

A collection of chemical reduction systems have been enforced to synthesize balanced and assorted morphologies of silver nanoparticles in water by the use of different reducing agentslike sodium borohydride,[3] hydrazine,[4] ascorbic acid.[5] The morphology, size and the size distribution build upon the strong and weak type of organic substrates to reduce the silver salts. Several biosynthesis methods have been reported, using several microbial flora and fauna likefungi,[6] yeasts, bacteria,[7] plantsor plant extracts.[8-10]

The green route synthesis of nanoparticles opens a new gateway for the production of nano-particles, which are non-toxic and economically viable.

EXPERIMENTAL

Chemicals

$AgNO_3$ and mandelic acid (99%) were purchased from Merck. Deionized water was used as solvent for the preparation of stock solutions.

Synthesis and characterization of Ag nanoparticles

Reduced nano-Ag was prepared by the mandelic acid reduction of $AgNO_3$ in the presence of okra juice as a stabilizing and binding agent under UV lamp (500 KW) conditions.

Okra juice (150 g in 100mL deionized water) was taken in a beaker at room temperature under UV radiation and $AgNO_3$ (0.045 M, 10mL) was added to the mixture with vigorous stirring at 600 rpm. Ag(I) reduction started by the addition of 7mL (1%) mandelic acid. The reaction mixture was stirred for 150 min for appropriate mixing to take place, resulting in a dark brown coloured solution, indicating the formation of the nanoparticles.

The optical transmission/absorption spectrum of the nanoparticles was determined by a UV-VIS spectrophotometer (UV-1700 Pharma Spectrophotometer, Shimadzu). The X-ray diffraction pattern was recorded using an X-ray diffractometer (Bruker D8 Advance Powder XRD), using CuKα radiation of wavelength λ=0.15406 nm in the scan range 2Θ = 10°-60°. FTIR analysis was carried out using an FTIR-84005 Shimadzu Spectrophotometer within the range of 400-4000 cm^{-1} in a KBr matrix. A Field Emission Scanning Electron Microscope (FE-SEM), Hitachi (S 4800), was used for elemental analysis of the prepared nano-silver particles. Detailed morphologies of silver nanoparticles were observed using TEM (FEI, TECNAI G^2-20, ULTRA-TWIN).

RESULTS AND DISCUSSION

UV-Visible spectroscopy

Figure1 shows a broad absorption band at 440 nm, which is a characteristic absorption band of spherical silver nanoparticles due to surface Plasmon. The reduction rate of Ag^+ ions turned up rapidly, comparably to the conventional method.[11] Furthermore, the UV-visible spectrumrevealed that the reaction medium exhibited an absorption band around 260 nm, which is attributed to binder or biopolymer moiety.

X-ray diffraction study (XRD)

The XRD pattern of silver nanoparticles (Fig.2) shows peaks at 2θ values of 19° (200), 27.81° (220), 32.24°(311), 34.10°(222), 38.39°(111) and 46.03°(420), corresponding to six diffraction facets of silver nanoparticles. The broad of X-ray diffraction peaks indicate that the crystallite size was very small.

Fourier transform infrared spectroscopy (FTIR)

FTIR measurements (Fig. 3) of the Ag-nanoparticles with *Abelmoschus esculentus* show the presence of bands at 501, 600, 922, 1036, 1117, 1200, 1383,1600,1724,2928, 3296 and 3561 cm^{-1}.The bands at 1600 and 1383 cm^{-1} developed for C-C and C-N stretching, respectively,which are commonly found in biopolymers (proteins, carbohydrates). This indicates the presence of a ligand for the silver nanoparticles, and thus confirms their synthesis. An absorption band of –OH stretching vibration corresponds to the region 3600-3100 cm^{-1}. The C-H stretching vibration from CH and CH_2 corresponds to the band at 2928 cm^{-1}. The peak 1724 cm^{-1} belongs to the carbonyl C=O stretching vibration. The peak at 1383 cm^{-1} and the little shoulder at 1320 cm^{-1} in the spectrum indicate the bending vibration of C-H and C-O groups of the aromatic ring in aromatic compounds. An absorption peak at 1036 cm^{-1} is attributed to the C-O and O-H stretching vibration, indicating the presence of a polysaccharide in the cellulose of the binder. The absorbance peak at 1162 cm^{-1} is due to an anti-symmetrical deformation of the C-O-C band. Furthermore, the absorption at 600 cm^{-1} corresponds to C-OH bending.[12]

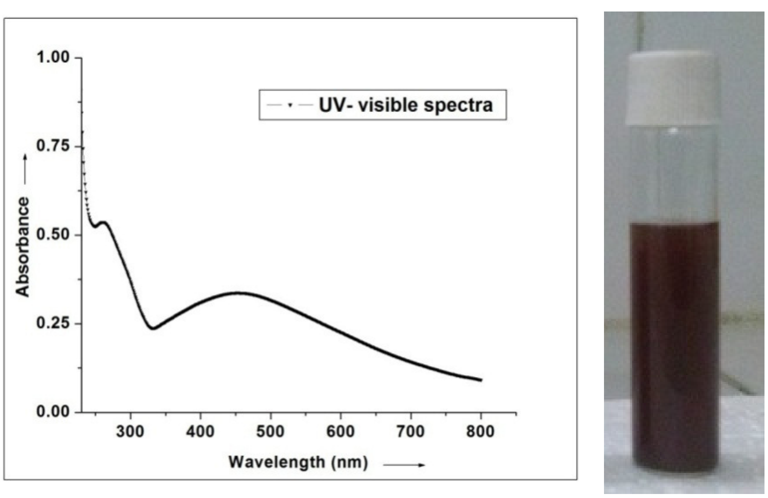


Figure 1: UV-visible spectra of *Abelmoschus esculentus* containing silver nanoparticles

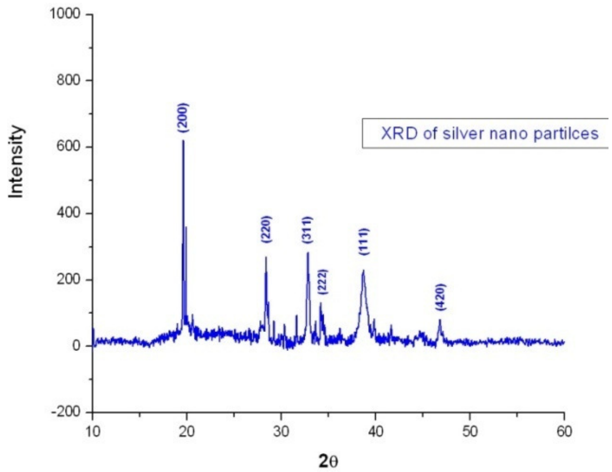


Figure 2: XRD pattern of silver nanoparticles

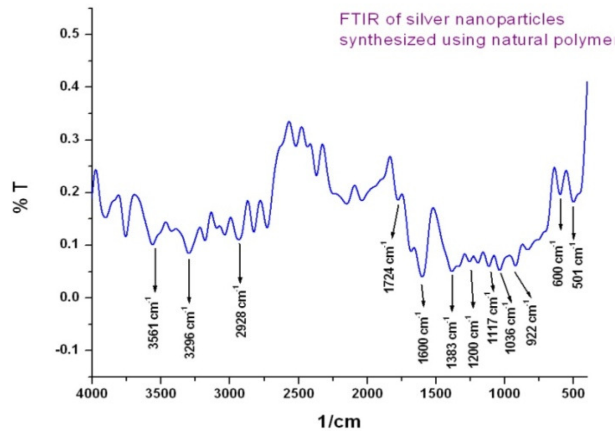
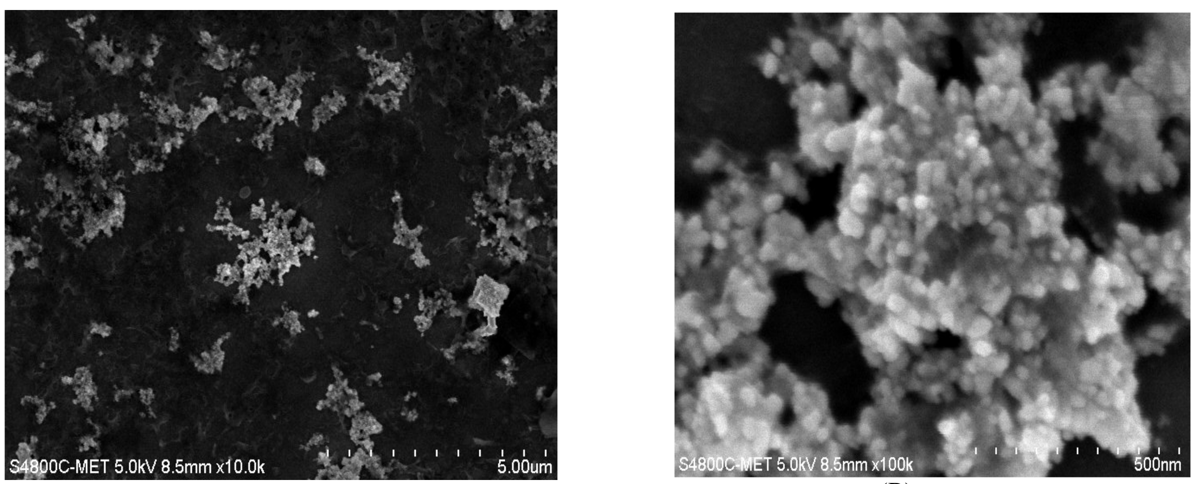


Figure 3: FTIR absorption spectra of silver nanoparticles bound with *Abelmoschus esculentus* juice



(A) (B)

Figure 4: FE-Scanning electron micrographs of silver nanoparticles quandary by *Abelmoschus esculentus* natural polymer at (A) 5µm and (B) 500nm,with spherical morphology

Field Emission Scanning Electron Microscopy (FE-SEM)

Scanning electron microscopy (Fig. 4) contributed more information on the morphology and size of the synthesized nanoparticles.The synthesized silver nanoparticles were found to possess a spherical shape and the particle size was found to range between 10 and 30 nm.

Transmission Electron Microscopy(TEM)

TEM micrographs (Fig. 5) with different magnificationsshowed a size distribution from 10 to 30 nm with spherical shape. The obtained radius values were in good agreement with theresults obtained from the FE-SEM analysis.

Antibacterial activity

Antibacterial activities of the silver nanoparticles were determined by the "Well diffusion method." The antibacterial activity was evaluated by measuring the zone of inhibition (ZOI) (Fig. 6).

The Ciprofloxacin antibiotic at 10 µg/mL was used in the test system as a positive control. The green route synthesized silver nanoparticles displayed appreciable antibacterial activity against the pathogens *Escherichia coli* (gram-negative) and *Staphylococcus aureus* (gram-positive). This study clearly indicated that the green route synthesized silver nanoparticles had a good antibacterial action against gram-negative bacteria and gram-positive bacteria.

The possible mechanism of bacterial lysis has been diagrammatically shown in Figure 7. Figure 8 gives a graphical representation of the results obtained for the minimum inhibitory concentration and zone of inhibition.

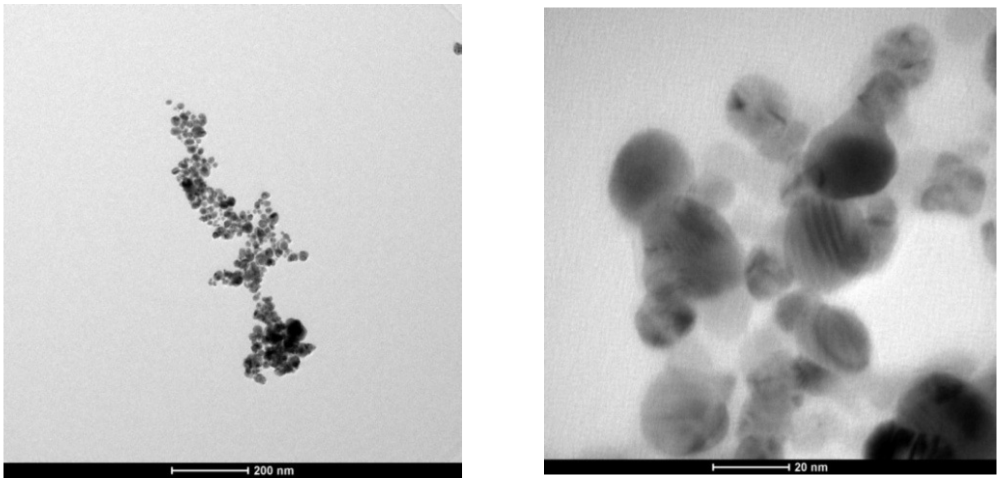


Figure 5: TEM micrographs of silver nanoparticles at (A) 200nm and (B) 20 nm with spherical shape

Figure 6: Comparison of the inhibition zone test between gram-positive (*S. aureus*) and gram-negative (*E. coli*) bacteria

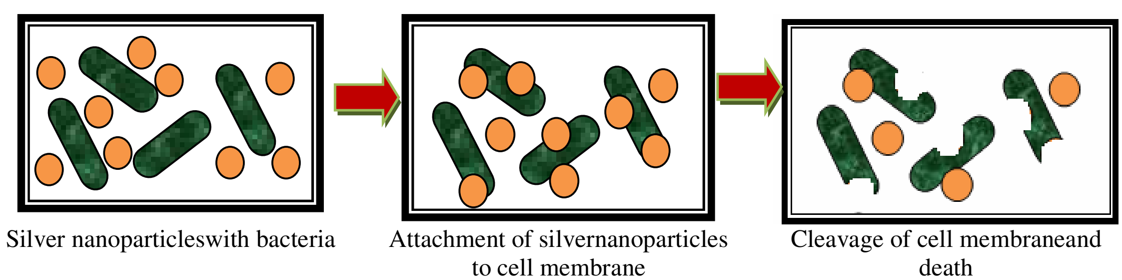
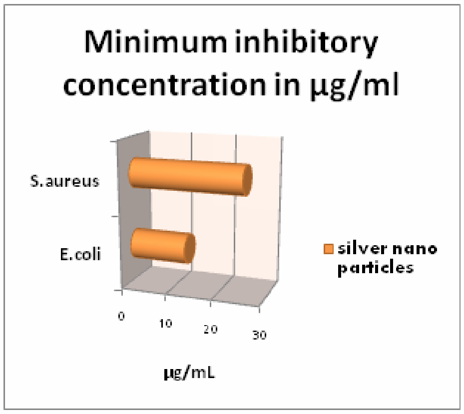


Figure 7: Possible mechanism of cell lysis of the tested bacterial strains



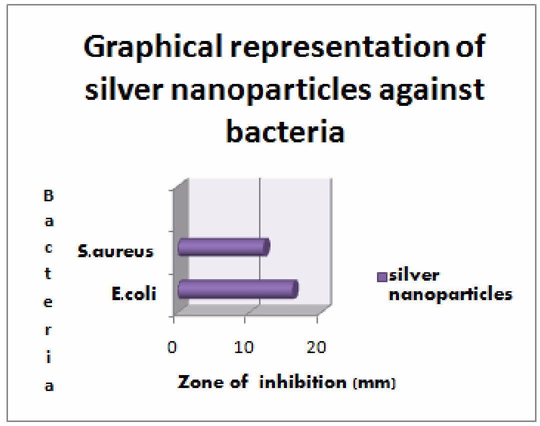


Figure 8: Graphical representation for antibacterial activity

CONCLUSION

Green route synthesis of nano sized silver particles using *Abelmoschus esculentus* as natural binder yielded spherically shaped silver nanoparticles. The synthesized nano silver material showed good bacterial activity against *Escherichia coli* and *Staphylococcus aureus*. Thus it can be concluded that *Abelmoschus esculentus* can be applied as an ecofriendly, cheap biopolymer for the synthesis of silver nanoparticles.

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