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Research Article

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Ecofriendly synthesis and applications of silver nanoparticles

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ABSTRACT

Synthesis of nano materials using novel and ecofriendly methods is under expedition. A novel green route method has been adopted for the preparation of silver nanoparticles, using Abelmoschus esculentus (Okra) juice as a natural binder. XRD, FTIR, UV-Vis, FE-SEM and TEM have been employed for characterization. UV-Vis analysis of the nanomaterial gave a peak at 445nm indicating a successful green route synthesis. FTIR analysis showed the presence of bands at 446, 497, 599, 922, 1036, 1117, 1200, 1383,1600,1774,2928, 3296 and 3561 cm^{-1}. Field emission scanning electron microscopy and Transmission electron microscopy reported spherical shape of the prepared silver nanoparticles. Antibacterial studies have been carried out by diffusion (cup plate) method for Escherichia coli, Staphylococcus Aueus and Cadida, which showed positive results against these bacteria.

Keywords: Biomaterials; Electron microscopy; Fourier transform infrared spectroscopy (FTIR); Nanostructures; Polymers

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INTRODUCTION

Nano materials are now-a-days a subject of interest due their varied applications. With the growing demand of nano materials for diverse applications, adoption of ecofriendly techniques for synthesis is becoming a matter of utmost importance [1]. Most of the ecofriendly techniques are fast, involve less expertise and are economical too. These methods have been reviewed as better alternatives to conventional ones and easy to scale up. Many biological substances and products of plant origin have been tested for the preparation of nano materials. Research has been carried out for the synthesis of silver nanoparticles from fungi [2], bacteria [3] and also different plants such as *Coriandrum Sativum* Leaf [4] *Aloe vera* [5], *Olea europaea* leaves [6], *Capsicum annum* [7], *Ocimum sanctum* (Tulsi) leaf [8], *Mulberry leaves* [9], *Carica papaya* [10] etc.

The present investigation was carried out to synthesise silver nanoparticles, which find various applications such as catalysts in organic chemical reactions, in bio-labelling, as an intercalation substances for electrical batteries and also as antimicrobials [11-13], by an environmental friendly method utilising *Abelmoschus esculentus* (Okra) juice. The biosynthesis involved reducing the number of silver ions in the solution of silver nitrate with an aqueous vegetable extract of Abelmoschus esculentus. Further, these green route synthesized nanoparticles were studied for human pathogens such as *E.coli*, *S. Aureus* and *Fungi Candida*.

EXPERIMENTAL SECTION

Materials

All the reagents used in the experiment were of A.R. grade. Silver nitrate and Tartaric acid were procured from Aldrich chemical Ltd., USA).

Preparation of the Extract

An extract of Abelmoschus esculentus (Okra) commonly known as “Lady Finger” was prepared taking the vegetable, washing it thoroughly thrice in double distilled water and then cutting into fine pieces weighing 25 gms. These fine pieces were the transferred into a 250mL Erlenmeyer flask in which 100 mL of deionized water was added. This flask was then kept undisturbed for 3 hour at room temperature. Filtration was then carried out to remove the vegetable waste, finally giving the extract in the filtrate.

Synthesis of Silver Nanoparticles

Solutions of silver nitrate 0.045 M and 15 % tartaric acid were prepared in deionized water. 50 mL of the vegetable extract of Abelmoschus esculentus (Okra) was mixed with 10 mL of 0.045 M silver nitrate and 15 mL of 15% tartaric acid under photo light (500 KW). A faint orange to dark brown colour was observed which indicated the formation of nano particles and the absorbance was noted at a time interval of 30 minutes to 150 minutes, using UV-Visible spectrophotometer. The obtained solution was stored at room temperature for about 12 hours in an amber colour bottle for the complete settlement of nanoparticles. This solution was then centrifuged for 1 hour and the supernatant was eliminated. To this 5 mL deionized water was added and the prepared particles were washed using centrifugation. These particles were then dried on a watch glass and stored.

Characterization

Silver nanoparticles prepared as above were characterized by different analytical techniques. Optical transmission / absorption spectrum was determined by a UV-Vis spectrophotometer (UV-1700 Pharma Spectrophotometer, Shimadzu). FTIR analysis was carried out using FTIR-84005, Shimadzu, Spectrophotometer with range of 400-4000 cm^{-1} in a KBr matrix. 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°. Field Emission Scanning Electron Microscope (FE-SEM), (JEOL-JSM-6360A) has been used for elemental analysis of the prepared nano-silver particles. Detailed morphologies of nano-silver particles were observed using TEM (FEI, TECNAI G^2-20, ULTRA-TWIN).

RESULTS AND DISCUSSION

UV-Visible Spectroscopic analysis

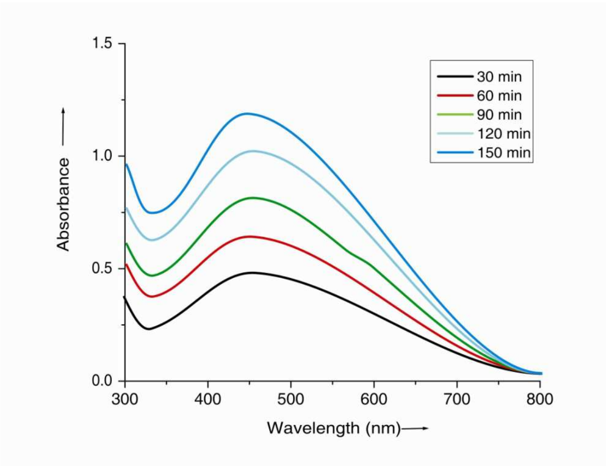

Fig. 1 UV-visible spectra of silver nanoparticles after time interval of 30 minutes

The addition of *Abelmoschus esculentus* (okra) extract to silver nitrate ($AgNO_3$) solution resulted in a colour change of the solution from faint orange to dark brown due to production of silver nanoparticles. This colour change was mainly due to the excitation of surface Plasmon vibration with the silver nanoparticles. UV-Vis absorbance spectra of the reaction mixture was taken from 30 to 150 minutes (Figure 1) at room temperature under photolight, giving a peak at 445 nm indicating a rapid green route synthesis of silver nanoparticles [14].

Fourier Transform Infrared spectroscopy (FTIR) Studies

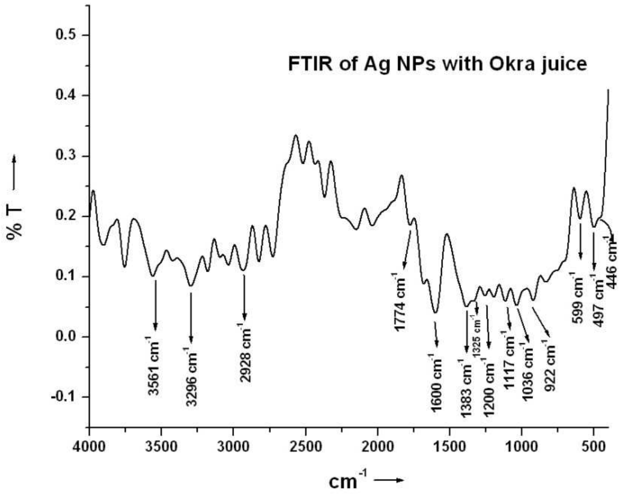

Fig. 2 FTIR absorption spectra of silver nanoparticles binded with abelmoschus esculentus (okra) extract

FTIR measurements of Ag-nanoparticles with *Abelmoschus esculentus* (Okra) showed the presence of bands at 446, 497, 599, 922, 1036, 1117, 1200, 1383,1600,1774,2928, 3296 and 3561 cm^{-1}(figure 2). The bands at 1600 and 1383 cm^{-1} corresponded to C-C and C-N stretching respectively indicating the presence of ligands similar to those found in biopolymers (proteins, carbohydrates) [15]. The region of 3600-3100 cm^{-1} showed band is corresponding to –OH stretching vibration. C-H stretching vibration due to the presence of CH and CH_2 was observed at 2928 cm^{-1}. C=O stretching vibration was well depicted at 1724 cm^{-1}. Anti-symmetrical deformation of C-O-C resulted in a peak at 1162 cm^{-1}. Furthermore the absorption at 600 cm^{-1} corresponded to C-OH bending [16].

X-ray diffraction

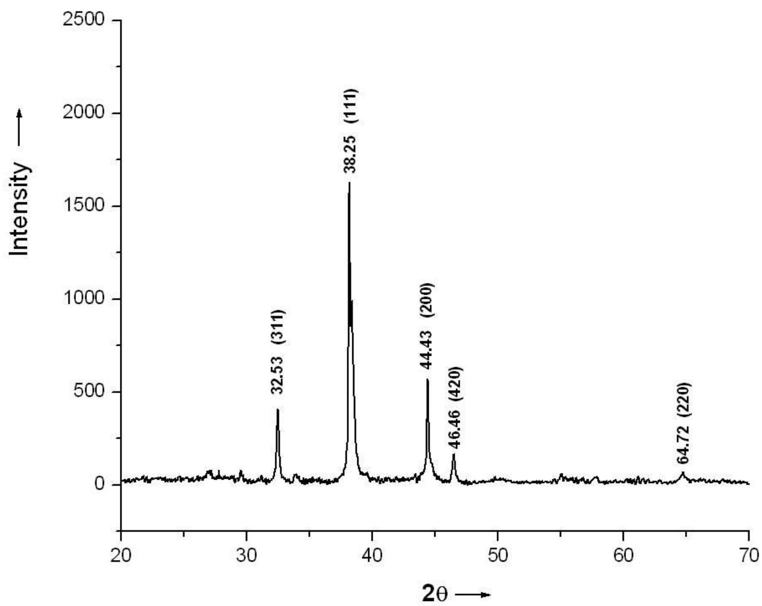

Fig. 3 X-ray diffractogram of silver nanoparticles synthesized from Abelmoschus esculentus (okra) extract

XRD spectrum of silver nanoparticles (Figure 3), as evident from the peaks at 2θ values of 32.53, 38.25, 44.43, 46.46 and 64.72°, corresponded to [311], [111], [200], [420], [222] planes for silver, respectively [17]. The broadening of the XRD peaks indicated the nanoparticles size of silver particles. The average crystalline size was determined using Scherrer's equation (Eqn. 1) as below:

$$\mathbf{D} = \frac{0.9\lambda}{\beta(\cos\theta)} \qquad \text{----- (1)}$$

Where λ is the wavelength β is the line broadening at half the maximum intensity (FWHM) and θ is the Bragg angle. The average crystalline size of silver nanoparticles as calculated from the XRD broadening was found to be within 30 nm [18].

First Emission Scanning Electron Microscopy (FE-SEM)

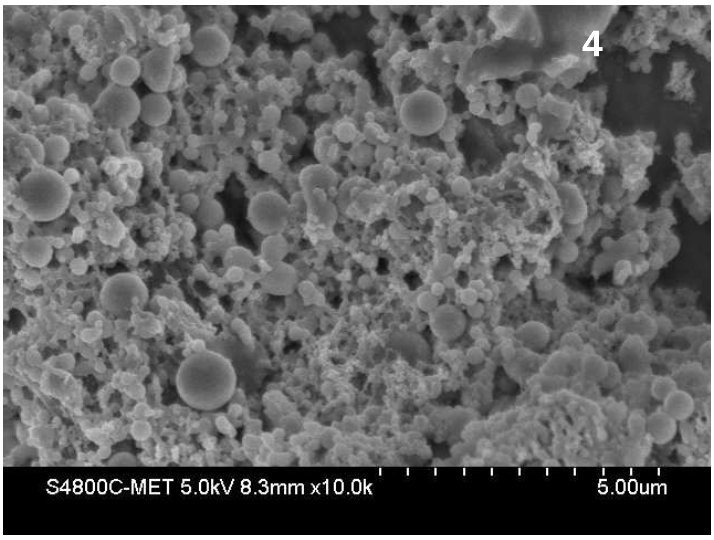

Fig. 4 FE-SEM image of spherical shaped silver nanoparticles at 5 µm

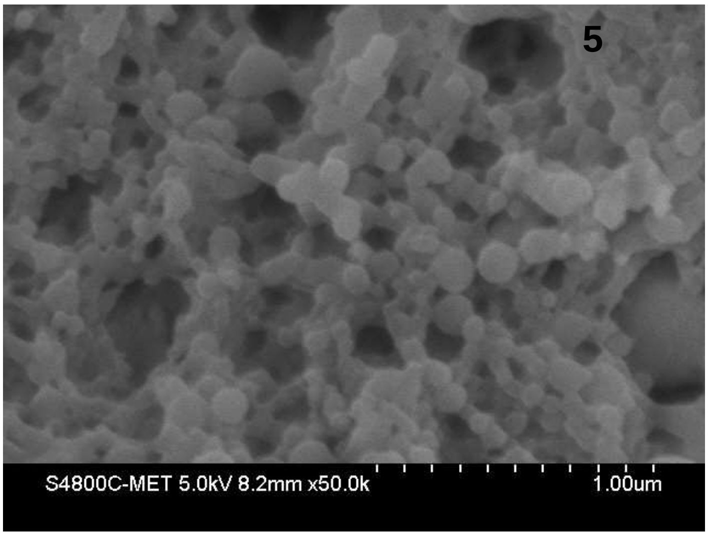

Fig. 5 FE-SEM image of spherical shaped silver nanoparticles at 1 µm

The surface morphology of the silver nanoparticles was characterized using FE-Scanning Electron Microscopy (Figure 4, 5 and 6). The appearance of the particles, as per the analysis, was highly crystalline and spherical in shape. The particles sizes of the sample estimated from the SEM images were greater than that obtained from XRD and TEM data. The SEM images symbolized the size of polycrystalline particles [19]. An investigation of some larger nanoparticles attributed to the fact that silver nanoparticles had the ability to agglomerate due to their high surface energy. In nanometer scale, some of the silver nanoparticles contributed to grow into twinned particles, most of them being fcc.

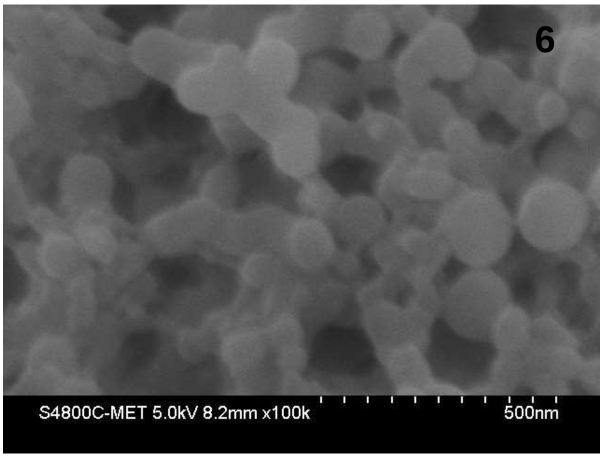

Fig. 6 FE-SEM image of spherical shaped silver nanoparticles at 500 nm

Transmission Electron Microscopy (TEM)

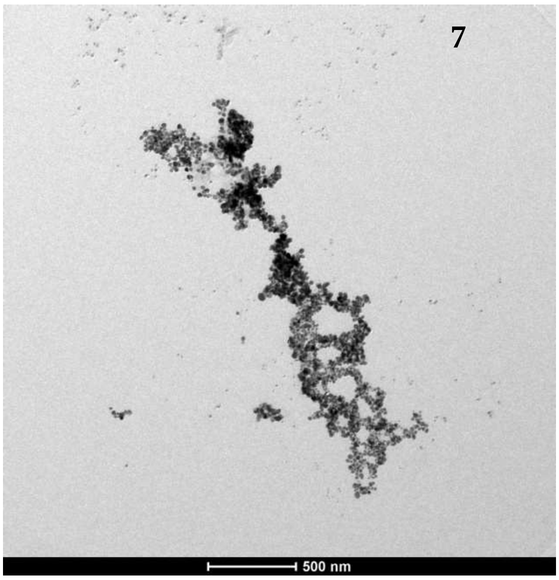

Fig. 7 TEM micrograph of the green route synthesized silver nanoparticles at 500 nm

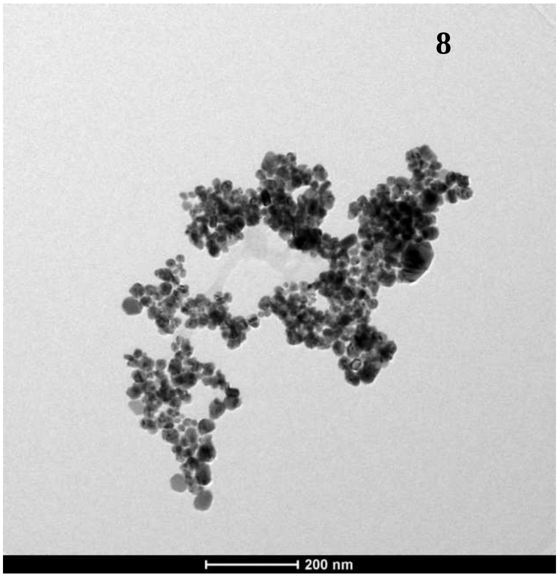

Fig. 8 TEM micrograph of the green route synthesized silver nanoparticles at 200 nm

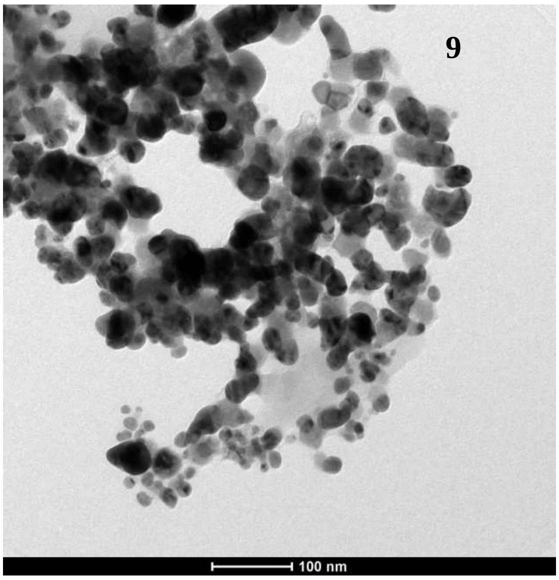

Fig. 9 TEM micrograph of the green route synthesized silver nanoparticles at 100 nm

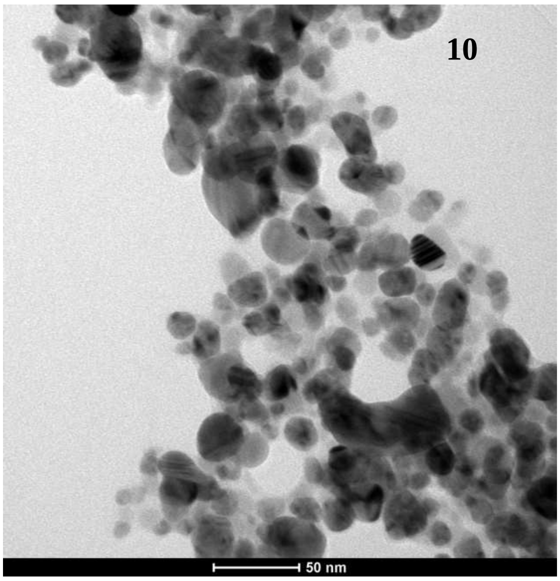

Fig. 10 TEM micrograph of the green route synthesized silver nanoparticles at 50 nm

TEM analysis has showed uniformly distributed, spherical silver nanoparticles with size ranging from 10 to 35 nm (Figure 7-12). Diffraction rings showed the nanocrystalline nature and corresponded to the interplanar distances of 2.35, 2.03, 1.44, 1.23 Å, in good agreement with the planes (111), (200), (220) and (311) respectively, for spherical silver nanoparticles[20-22].

Antibacterial Activity

The Antibacterial activity of the silver nanoparticles was determined by well diffusion (Cup plate) method. Clinically isolated samples of *Staphylococcus aureus* (Gram positive), *Escherichia coli* (Gram negative) and *Candida* (Fungi) were used for study.

Table 1 Measure of zone of inhabitation

S. No.	Bacteria	Zone of inhibition (mm)
1	*E.Coli*	17
2	*S.Aureus*	11
3	*Candida*	30

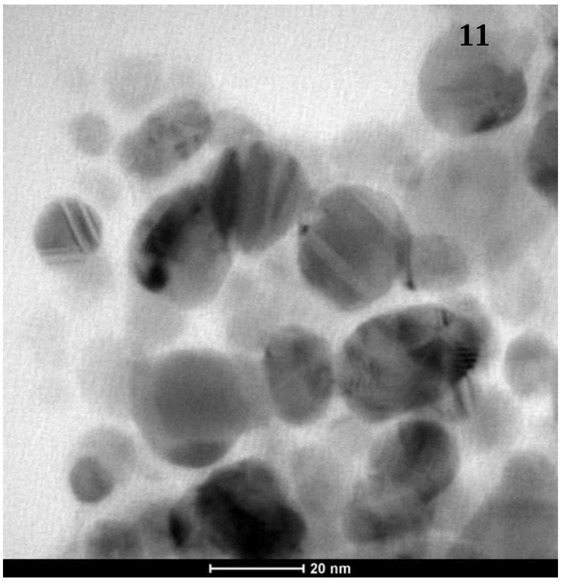

Fig. 11 TEM micrograph of the green route synthesized silver nanoparticles at 20 nm

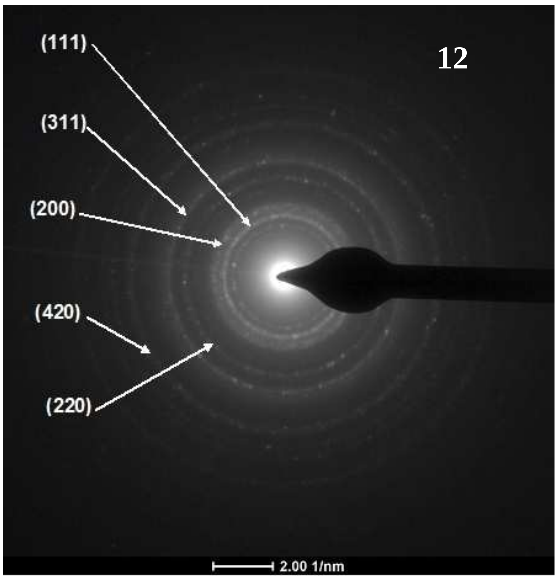

Fig. 12 SAED of silver nanoparticles

HIMEDIA NM 011 Nutrient Agar and sodium hydroxide (pH = 7.4) were used as the medium for culturing organisms. Silver nanoparticle solutions with concentrations 10 μL, 25 μL and 30μL were added for testing the activity of *Staphylococcus aureus*, *Escherichia coli* and *Candida* respectively, using a micropipette. Plates containing the bacteria were incubated at 37ºC for 24 hours [23-24] after which clear zones of inhabitation were observed (Table 1).

CONCLUSION

Abelmoschus esculentus, a natural binder can thus be applied as an ecofriendly and cheap, bio-polymer for the synthesis of silver nanoparticles yielding spherical silver nanoparticles via green route method. Silver nanoparticles prepared were crystalline in nature, sizes being approximately 30 nm, as confirmed by XRD, FE-SEM and TEM. Synthesized nano silver material by green route method showed good antibacterial activity against *Esherichia coli*, *Staphylococcus aureus* and *Candida*.

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