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Eco-friendly synthesis of silver nanoparticles using leaf extract of *Grewia flaviscences* and study of their antimicrobial activity

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ABSTRACT

In this work, we report the eco-friendly synthesis of silver nanoparticles (AgNPs) by bio-reduction method. Aqueous extract of *Grewia flaviscences* plant leaf was used as stabilizing and reducing agent. Formation of AgNPs was initially identified by change in color from light yellow to dark brown. UV–visible spectra showed the characteristic surface Plasmon resonance peak of synthesized AgNPs between 380 and 460 nm. X-ray diffraction analysis revealed the crystalline nature of AgNPs, whereas Energy-dispersive X-ray spectroscopy confirms the metallic nature of silver. Field emission scanning electron microscopy and transmission electron microscopy images showed the spherical nature of the particles whereas size distribution was measured by Zetasizer, which revealed that size of AgNPs were in the range of 50–70 nm. Biosynthesized AgNPs exhibited antimicrobial activity against both gram positive and gram negative bacteria, *Bacillus* and *Pseudomonas aeruginosa*. Sps., respectively. The effects of leaf extract and silver nitrate concentrations on AgNPs have also been studied.

1. Introduction

Nanomaterials often show and considerably changed physical, chemical and biological properties compared to their macro scale counterparts. Nanomaterials may provide solutions to technological and environmental challenges in the areas of solar energy conversion, catalysis, medicine, water treatment etc. Based on size, distribution and morphology, nanomaterials exhibit completely new or improved advantageous properties. Nanomaterials often exhibit very interesting electrical, optical, magnetic and chemical properties. Synthesis and studying chemical and physical properties of silver nanoparticles (AgNPs) are currently of considerable interest because of their potential applications in mechanics [1], optics [2], sensors [3], drug delivery [4], DNA sequencing [5] and biomedical applications [6–8]. Synthesis of AgNPs can be achieved through various physical and chemical methods such as gamma radiation assisted [9], microwave assisted [10], laser ablation [11], polyol process [12], thermal decomposition [13], lithography [14] etc., methods. All of these methods have disadvantages like use of hazardous reagents, toxicity, high cost and high energy consumption. Biogenic synthesis of AgNPs has many advantages over physical and chemical methods, such as, low cost, rapid, eco-friendly and does not require any toxic substances. Various plant extracts including *Tithonia diversifolia*, *Cissus quadrangularis*, *Cassia angustifolia*, *Ocimum tenuiflorum*, *Coleus aromaticus*, and *Cinnamoum camphora* [15–20] were reported for biosynthesis of AgNPs. In the present study, rapid and eco-friendly synthesis of AgNPs from the aqueous leaf extract of *Grewia flavescens* was studied. The synthesized AgNPs were characterized and evaluated for their antimicrobial ability against gram positive bacteria *Bacillus* and gram negative bacteria *Pseudomonas aeruginosa*.

2. Materials and methods

Collection and preparation of plant extracts: Healthy leaves of *Grewia flaviscenes* were collected in our Yogi Vemana University campus, Kadapa, India. The leaves were washed with tap water and then with double distilled water. The leaves were shade dried for two weeks and grinded to fine powder. 5 g of leaf powder was added to 100 mL of distilled water in a 250 mL Erlenmeyer flask then boiled for 15 min and cooled to room temperature. Then, the leaf extract was filtered with Whatman No. 1 filter paper and collected filtrate was stored at 4 °C for further use.

Biosynthesis of AgNPs: Varied amounts (10, 20 and 30% wi th respect to $AgNO_3$) of aqueous leaf extracts were added to 1 mM $AgNO_3$ (Sigma-Aldrich) solution and kept in dark at room temperature for 6 h in a 250 mL Erlenmeyer flask. Bio-reduction was monitored by visual observation and spectrophotometrically. Silver nitrate concentration varied from 1 to 5 mM on keeping leaf extract concentration constant at 10% to $AgNO_3$.

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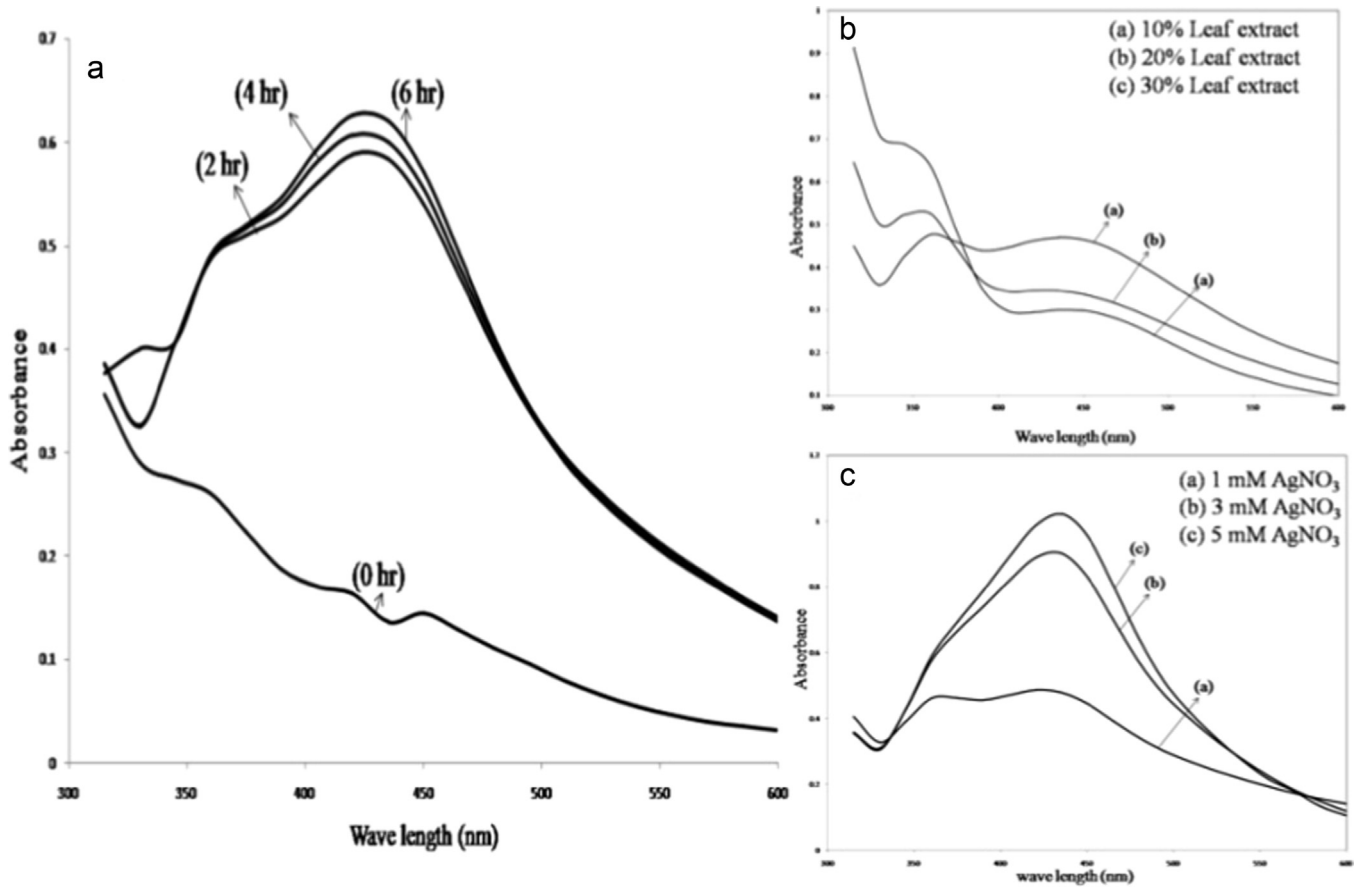

Fig. 1. (a) UV–vis spectra of AgNPs formation at different time intervals (b) UV–vis spectra with varying leaf extract concentration and (c) UV–vis spectra with varying silver nitrate concentration.

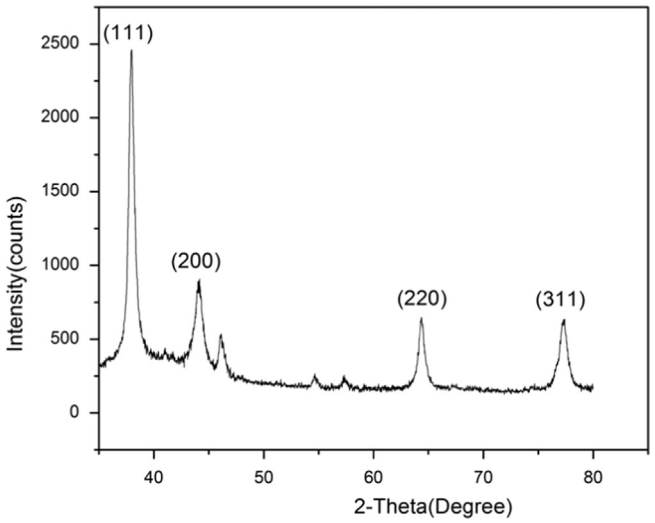

Fig. 2. XRD pattern of the AgNPs synthesized from the leaf extract *Grewia flaviscenes.*

Characterization of AgNPs: The reduction of silver ions into silver particles was monitored by UV–visible spectroscopy (SHIMADZU MODEL UV 1800, Japan). An aliquot of 100 μL of the sample was diluted with 1 mL distilled water and spectra were recorded between 200 and 700 nm. X-ray diffractometer (RIGAKU, SMARTLAB) operated at 30 kV 100 mA with Cu *K*β as a radiation source was used to record XRD patterns of dried AgNPs to confirm the crystalline nature. Field Emission Scanning Electron Microscopy (FESEM) image was recorded on SUPRA^{TM} 55 with co-relatively microscope at 20 kV along with Energy-dispersive X-ray (EDX) spectroscopy spectrum. Sample was prepared by evaporating few drops of AgNPs dispersed in distilled water on a clean glass slide. Transmission electron microscopy image was taken using a JEOL 3010 at 20 kV microscopy. TEM samples was prepared by pacing a drop of aqueous dispersion of AgNPs in distilled water on 200 mesh carbon coated copper grids and dried at ambient conditions for 10 to 12 h. Particle size distribution of the synthesized AgNps was carried out on Zetasizer S-90, Malvern, UK by taking dilute dispersion solution of AgNPs in Mill *Q* water.

Antimicrobial activity of AgNPs: Antimicrobial activity of AgNPs against *Bacillus,* and *P. aeruginosa* sps were carried out using Disc diffusion method [21]. Five cavities of wells were made in a each plate and well no. 1, 2 and 3 was filled with 10, 20 and 30 μL aqueous silver nanoparticles, well no. 4 was filled with leaf extract and well no. 5 was filled with 30 μL of 1% of *streptomycin* (antibiotic) solution. The plates were incubated in an incubator at 37 °C overnight, after incubation period zone of inhibition (ZoI) around the wells were measured.

3. Results and discussion

The reduction of silver ions into silver nanoparticles in the presence of aqueous leaf extract of *Grewia flaviscenes* was very rapid. The particles were formed with in 2 h as observed by change in color of the solution from light yellow to dark brown. However, the reaction was continued up to 6 h. It was also monitored by measuring UV–vis spectra of the reaction mixture at different intervals of time and the results are presented in Fig. 1(a). A strong absorption peak between 380 and 460 nm was observed and