

GREEN BIOSYNTHESIS OF SILVER NANOPARTICLES USING *MAYTENUS EMARGINATA* FRUIT EXTRACT

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ABSTRACT

In the present study, an attempt has been made to synthesis AgNPs using *Maytenus emarginata* fruit. Biosynthesis of AgNPs was achieved by a novel, ecofriendly, simple, pollutant free, green chemistry procedure using aqueous fruit extract of *M. emarginata* as a reducing and capping agent. Rapid formation of stable silver nanoparticles was observed on exposure of the aqueous fruit extract with solution of 1 mM silver nitrate. The successful synthesis of AgNPs formation was confirmed by various spectral analysis like UV-visible spectroscopy, Scanning Electron Microscopy (SEM), Zeta potential and Fourier

Transform Infra-Red spectroscopy (FTIR). The UV-visible spectrum showed maximum absorption band at 436 nm. The particle size as determined by zeta potential was - 24.33. The average size of the nanoparticles was found to be 0.244 nm. The results of various techniques confirmed the formation of silver nanoparticles.

KEYWORD: Silver nanoparticles, *Maytenus emarginata*, XRD, SEM, FTIR.

INTRODUCTION

The nanotechnology field is one of the most active areas of research in modern materials science. Nanotechnology is an attractive research field because the use of nanoparticles is increasing in various sciences. Nanoparticles exhibit completely new or improved properties based on special characteristics such as size, morphology and distribution. (Smith *et al.*, 2006). Their potential effects are used in both *in vivo* and *in vitro* biomedical applications and research (Singh *et al.* 2008). Silver nanoparticles have potential applications in industry,

agriculture, pharmaceutical and medicine (Kim *et al.*, 2004; Sperling *et al.*, 2008) agriculture (Park *et al.*, 2006) and cosmetics (Kokura *et al.*, 2010). Nanoparticles have important role in drug delivery, antimicrobial activities, diagnosis and tissue engineering (Malabadi *et al.*, 2012; Song and Kim, 2009) and also used in various applications including dental, medical therapeutics and diagnosis, food packaging, catheters, textiles, and coatings (Yoksan and Chirachanchai, 2010; Lim *et al.*, 2012) catalysis (Schmid, 1992) biosensing (Mirkin *et al.*, 1996), etc.

Route of synthesis of nanoparticles by physical and chemical methods may have considerable environmental defect because it requires high pressure, energy, temperature and toxic chemicals, technically laborious and economically approach for the synthesis. For example, silver ions are reduced by chemical, electrochemical, radiation, photochemical methods (Ahmad *et al.*, 2010). But, plant extracts based biosynthesis of silver nanoparticles has been found to be cost effective and environmental friendly. The use of environmentally benign materials such as plant leaf extract (Parashar *et al.*, 2009), bacteria (Ahmad *et al.*, 2003; Saifuddin *et al.*, 2009), fungi (Basavaraja *et al.*, 2008) enzymes (Willner *et al.*, 2007) algae (Vivek *et al.*, 2011) for the synthesis of nanoparticles suggest many benefits of eco friendliness and compatibility for pharmaceutical and other biomedical usages as they do not use toxic chemicals for the synthesis protocol.

Historically, silver is known to have a disinfecting outcome and has many applications in traditional medicines. It has been reported that silver nanoparticles (SNPs) are non-hazardous to humans and most effective against bacteria, virus and other eukaryotic micro-organism (Jeong *et al.*, 2005). Silver nanoparticles exhibiting very strong bactericidal activity against multiresistant strains including both Gram positive and Gram negative bacteria can be considered as potential antimicrobial agent (Morones *et al.*, 2005; Panacek *et al.*, 2006; Padalia *et al.*, 2014).

Maytenus emarginata (Willd.) belongs to the family Celastraceae and is an evergreen tree that tolerates various types of stresses of the desert. Traditionally it is used for fever, asthma, rheumatism, gastrointestinal disorders worldwide. *Maytenus emarginata* plants have reported various activity like antipyretic activity (Sandhya and Vineela, 2012), antioxidant activity (Gupta and Sharma, 2012).

In the present work, an attempt has been made to synthesize silver nanoparticles using aqueous Fruits extract of *Maytenus emarginata*. The characterization was done using several spectral analysis. UV-Vis spectroscopy, Zeta Potential Measurement, FTIR analysis, SEM analysis

MATERIAL AND METHOD

Chemicals

Fresh fruits of *Maytenus emarginata* were purchased from local market of Rajkot Gujarat, India. All the chemicals were obtained from Hi Media Laboratories and Sisco research Laboratories Pvt. Limited, Mumbai, India. Ultra purified water was used for experiment.

Preparation of the extract for synthesis of silver nanoparticles

Fresh fruits were thoroughly washed with tap water, followed by double distilled water and cut into small pieces. 5 g of cut fruits were boiled for 10 min in 100 ml ultra pure water and filtered through Whatmann No. 1 filter paper. The filtered fruits extract was used for the synthesis of silver nanoparticles.

Synthesis of silver nanoparticles: Aqueous solution (1mM) of silver nitrate (AgNO_3) was prepared and used for the synthesis of silver nanoparticles. 3ml of extract was added to 40 ml of 1 mM AgNO_3 solution for the reduction of Ag^+ ions. The synthesis of silver nanoparticles was carried out at room temperature ($25^\circ \text{C} \pm 2^\circ \text{C}$) for 24 h in dark. The silver nanoparticles solution thus obtained was purified by repeated centrifugation at 10000 rpm for 10 min followed by redispersion of the pellet of silver nanoparticles into Acetone. After air drying of the purified silver particles, stored at 4°C for further analysis.

Characterization of the synthesized silver nanoparticles: Synthesis of silver nanoparticles solution is easily observed by ultraviolet – visible (UV-Vis) spectroscopy. The reduction of the Ag^+ ions in solution was monitored by periodic sampling of aqueous component and measuring the UV-Vis spectra of the solution. UV-Vis spectra of these aliquots were monitored as a function of time of reaction on a spectrophotometer (Shimadzu UV-1601) in 400-700 nm range operated at a resolution of 10 nm.

Zeta Potential Measurement: Zeta potential is an essential parameter for the characterization of stability in aqueous nano suspensions. The zeta potential measurement was performed using a Microtra (Zetara Instruments).

FTIR analysis: Possible functional groups involved in the synthesis and stabilization of silver nanoparticles was studied by FTIR spectroscopy. The FTIR was recorded in the range of 400-4000 cm^{-1} Nicolet IS10 (Thermo Scientific, USA) The various modes of vibrations were identified and assigned to determine the different functional groups present in the *Maytenus emarginata* fruits extract

XRD measurement: The structure and composition of purified silver nanoparticles were analyzed by XRD. The silver nanoparticles was collected for the determination of the formation of Ag nanoparticles by an X'Pert Pro x-ray diffractometer (PAN analytical BV) operated at a voltage of 40 kV and a current of 30mA with Cu $K\alpha$ radiation in θ - 2θ configurations. The crystallite domain size was calculated from the width of the XRD peaks, assuming that they are free from non-uniform strains, using the Scherrer formula. $D = 0.94 \lambda / \beta \cos \theta$ where D is the average crystallite domain size perpendicular to the reflecting planes, λ is the X-ray wavelength, β is the full width at half maximum (FWHM), and θ is the diffraction angle.

SEM analysis: Scanning Electron Microscopic (SEM) analysis was done using Hitachi S-4500 SEM machine. Thin films of the sample were prepared on a carbon coated copper grid by just dropping a very small amount of the sample on the grid, extra solution was removed using a blotting paper and then the film on the SEM grid were allowed to dry by putting it under a mercury lamp for 5 min.

RESULTS AND DISCUSSION

Formation of silver nanoparticles: Synthesis of silver nanoparticles using plants extracts is getting more popular. Silver nanoparticles were synthesized within 1 h of incubation period using fruits extracts of *M. emarginata* in 1 mM of silver nitrate solution. After overnight incubation at room temperature, the colorless reaction mixture turned into a dark brown color solution which indicates the AgNPs synthesis. Color change in the reaction mixture is due to active molecules present in the fruits extracts which are responsible for possible bioreduction of silver metal ions into silver nanoparticles. The intensity of color change was increased directly proportion to the incubation period of nanoparticles synthesis (Mulvaney, 1996).

UV-Vis spectral analysis: In order to monitor the formation of silver nanoparticles, the absorption spectra of the synthesized silver nanoparticles were recorded from the fruits

extract of *M. emarginata* reaction at 15 min time is presented in Fig. 1. The spectrum showed maximum absorption band at 436 nm that is surface plasmon resonance of silver nanoparticles. The intensity steadily increased as a function of reaction time. The colourless solution turned brown indicating the nanoparticles formation of silver. It is well known that the absorbance of Ag NPs was mainly depends upon the size and shape (Mock *et al.*, 2002). Mason *et al.*, (2012) and Christensen *et al.*, (2011) reported maximum absorption peak of silver nanoparticles at 435 nm from *Panicum virgatum* extract and *Murraya koenigii* leaves extract respectively.

Zeta potential measurement: Stability of AgNPs is determined by zeta potential measurement. Zeta potential value ± 30 mV is considered as stable nano suspension. The zeta potential was -24.33 mV (Fig. 2). This suggested that the surface of the nanoparticles was negatively charged that dispersed in the medium.

FTIR analysis: FTIR measurements were carried out to identify the possible biomolecules responsible for reduction, capping and efficient stabilization of the Ag nanoparticles. The FTIR spectra silver nanoparticles were shown in Fig. 3. The peak at 3522.13 cm^{-1} , 3398.89 cm^{-1} , 3184 cm^{-1} and 3034.12 cm^{-1} corresponds to O-H stretching strong alcohol, phenol, N-H stretching medium 1,2 amines and amides, O-H stretching carboxylic acid and C-H stretching aromatics respectively. The assignment at 1616.40 cm^{-1} , 1375.29 cm^{-1} , 808.20 cm^{-1} and 538.16 cm^{-1} corresponds to N-H bend medium 1° amines, C-H alkanes, C-H stretching aromatics and C-Br stretching medium alkyl halides respectively. It is used to identify the biomolecules on the surface of the AgNPs which are responsible for capping and efficient stabilization of the metal nanoparticles.

SEM analysis: SEM technique was employed to visualize the size and shape of silver nanoparticles. In Fig. 4. SEM images were obtained with *M. emarginata* fruits extracts. SEM grid which were prepared by placing a small amount of sample powder on a copper coated grid and drying under lamp. The size of silver nanoparticles was in the range of 0.095 to 0.149 nm; the average size of the nanoparticles was found to be 0.244 nm (Fig.4.8A and B). The shape of the biosynthesized nanoparticles was spherical and irregular in shape with moderate variation in size. Ponarulselvam *et al.*, (2012) reported spherical silver nanoparticles from *Catharanthus roseus* leaf extract with an average size ranging from 35 to 55 nm.

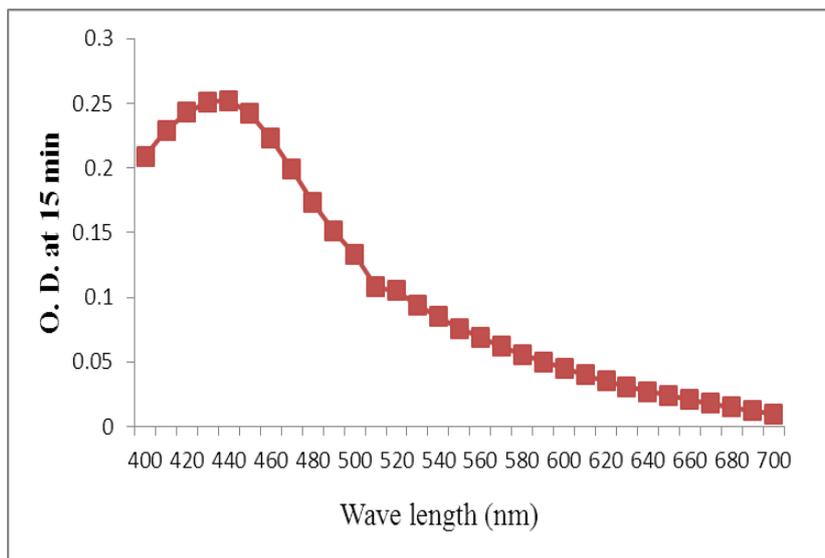


Fig.1 UV-visible spectrum of biosynthesized AgNPs

Mobility	-1.90u/s/V/cm
Zeta Potential	-24.33 mv
Charge	-0.18236 fC
Polarity	Negative
Conductivity	15 uS/cm

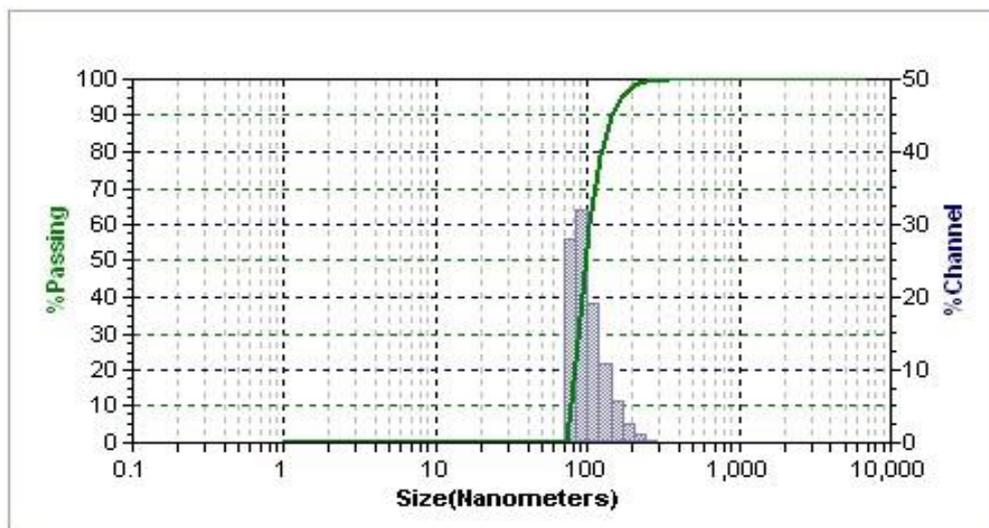


Fig.2 Zeta potential measurements of biosynthesized AgNPs

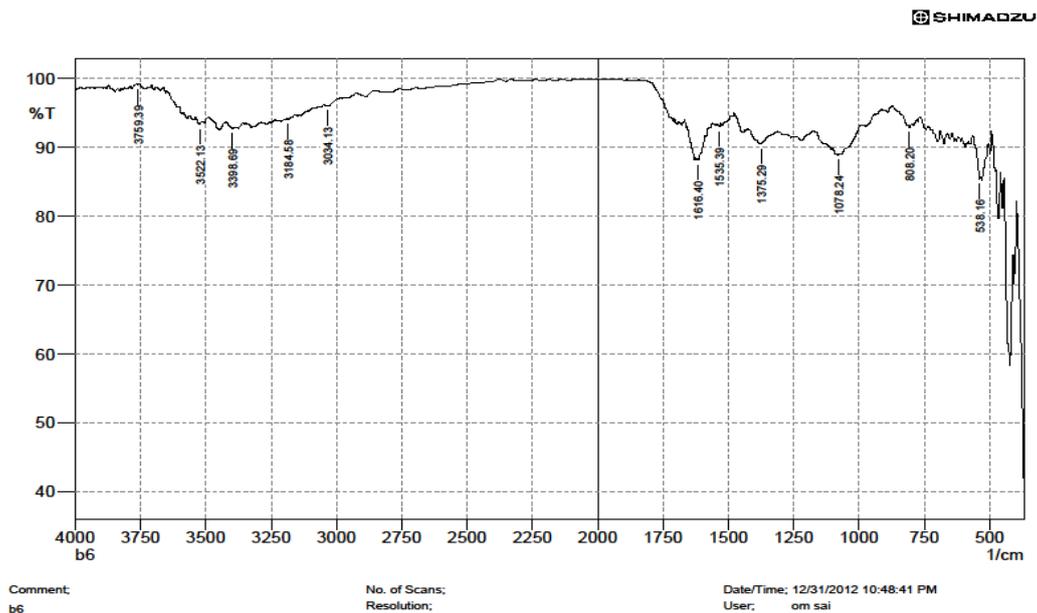


Fig. 3 FTIR spectrum of biosynthesized AgNPs.

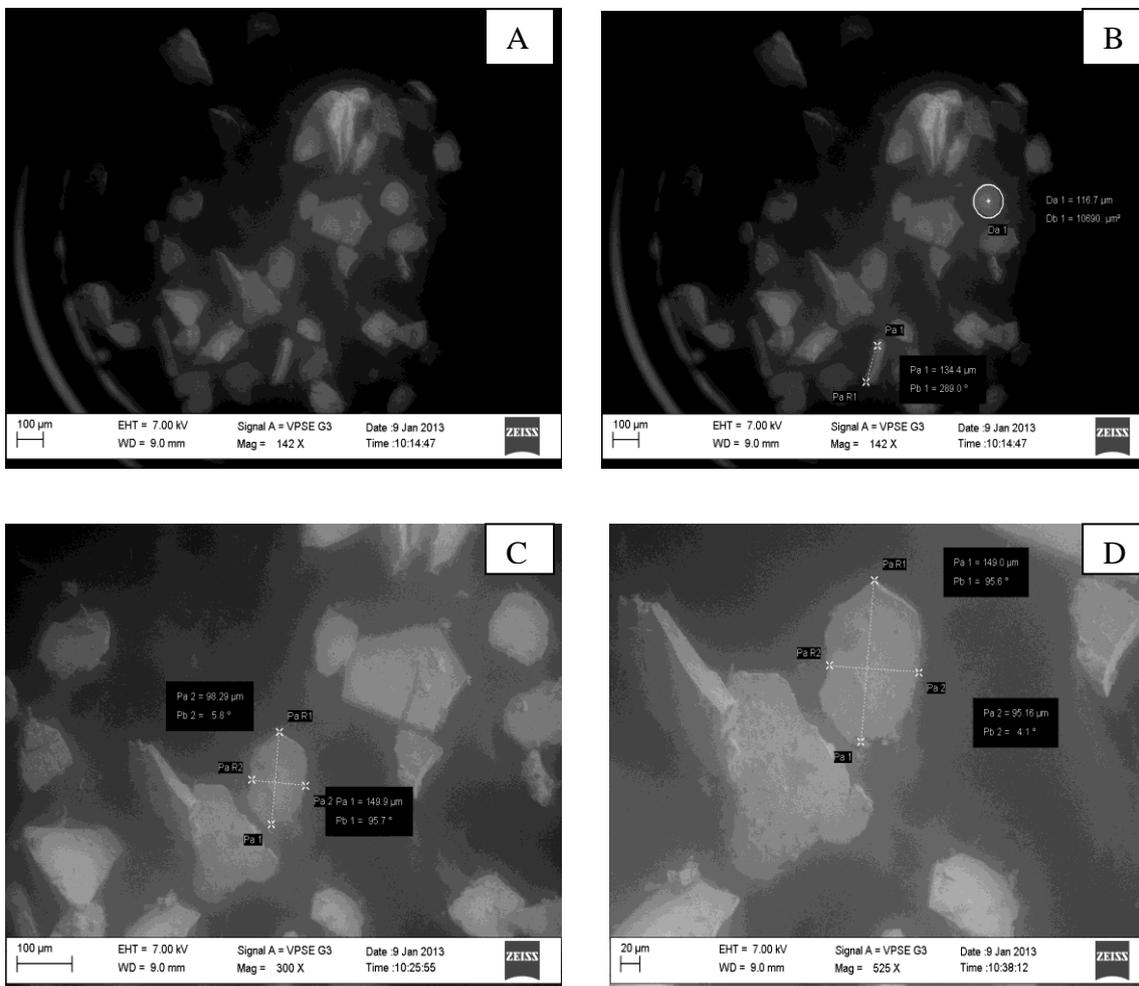


Fig.4 SEM images (A, B, C, D) of AgNPs in low and high magnification

CONCLUSION

In present study, silver nanoparticles were synthesized using *Maytenus emarginata* fruit. Green synthesis method is economical, environmental friendly, inexpensive and involves non-toxic materials. Silver nanoparticles were characterized by means of UV-Vis spectroscopy, FTIR analysis, Zeta potential, SEM analysis. Several functional groups present in the fruit extracts are likely to be responsible for the formation of silver nanoparticles. The silver nanoparticles were spherical and irregular in shape.

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