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SYNTHESIS AND CHARACTERIZATION OF SILVER NANOPARTICLES USING *MORINGA OLEIFERA* LEAVES

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ABSTRACT

Moringa oleifera is a fast-growing, drought-resistant tree. It is commonly called as Drumstick tree from the appearance of the long, slender, triangular seed-pods. This plant grows in poor nutrient soil and it has tremendous potential for human use. In the present study, the aqueous leaves extract of *Moringa oleifera* was used to synthesize silver nanoparticles. 5 ml of aqueous leaf extract was added to 50 ml 1mM Silver nitrate. After 72 hours of incubation at room temperature, the formation of stable dark brown color indicated the synthesis of AgNPs. Synthesized silver nanoparticles were characterized using UV-VIS Spectroscopy, XRD, and TEM.

Key words: *AgNO₃*, *PDA*, *Bavistin*, *Moringa oleifera* leaves, and *Mercuric chloride* etc.

INTRODUCTION

Nanoparticles, generally considered as particles with a size up to 100 nm, exhibit completely new or improved properties as compared to the bulk material that they are collected based on particular characteristics such as size, distribution, and morphology [Wildenberg 2005]. Nanoparticles of noble metals, such as gold, silver and platinum are broadly applied in many fields and also directly come in contact with the human body, such as shampoos, soaps, detergents, shoes, cosmetic products, and tooth paste, besides medical and pharmaceutical applications [Parveen *et al.*, 2012]. In present days, nanoparticles based on their electrical, optical, magnetic, chemical and mechanical properties are used in various areas, such as the medical sector for diagnosis, antimicrobial, drug delivery and also they are also used in the electronic and optoelectronic industry [Phillips *et al.*, 2011, Raveendran and Guliants 2009] in the chemical sector for catalysis [Ju-Nam and Lead 2008] for environmental protection [Kim *et al.*, 2010] and energy conversion [Ravindra and Rajasab 2014]. Nanoparticle synthesis is generally carried out by physical and chemical methods, such as laser ablation, pyrolysis, chemical or physical vapour deposition, lithography electro deposition, sol gel etc., which are costliest, human hazardous, and not eco friendly. Because of the use of toxic and hazardous reagents emits toxic byproducts in environment. Compared to physical and chemical methods, the green synthesis is low cost, ecofriendly, competent, and fast method for producing nanoparticles. Currently, there is a growing need to develop environmentally benevolent nanoparticles synthesis processes that do not use toxic chemicals in the synthesis protocol. So the researchers in the field of nanoparticles synthesis and assembly have turned to biological inspiration [Song *et al.*, 2009].

In biosynthesis, many prokaryotic and eukaryotic micro organisms such as bacteria, fungi, yeast, and macro organisms like plants are using for nanomaterial synthesis either intra or extracellularly. Compare to micro organisms' plants are the rich source of nature and are easily available in nature and also their enzymatic activity is more. Grounding on these potential properties we selected plant extract for synthesis of Silver nanoparticles. In the present work, we used *Moringa oleifera* leaves extract for AgNPs synthesis. *Moringa oleifera* fast-growing, drought-resistant tree belongs to family moringaceae. Plant is widely grown in poor nutrient soil and it has tremendous potential for human use. Because of its ever green properties, easy availability and more metabolic rate we selected *Moringa oleifera* for silver nanoparticles synthesis.

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MATERIALS AND METHODS

Materials

Silver nitrate (AgNO_3), *Moringa oleifera* leaves, and Mercuric chloride

METHODOLOGY

Sample collection

Fresh leaves of *Moringa oleifera* were collected in sterilized polythene bag from Gulbarga University campus kalaburgi. And brought to Mycology and Plant pathology Laboratory and stored in laboratory conditions for further studies.

Preparation of leaf extract

Collected sample *Moringa oleifera* leaves were surface sterilized, and dried under shade. Dried leaves were cut into small pieces and grinded to powder. 10 gram of *Moringa oleifera* leaf powder boiled in 200 ml of distilled water for 10 minutes then filtered it with whatman No. 1 filter paper. The prepared plant extract solution was cooled at 4° C and stored in laboratory condition (Fig. 1) for further experimental work.

Green synthesis of silver nanoparticles

50 mL of 1 mM aqueous solution of silver nitrate (AgNO_3) was taken in 100 mL conical flask. Then the prepared leaf extract solution with various concentrations from 5, 10, and 15 mL was added separately and agitated at room temperature. Control treatment (without Silver nitrate, only plant extract and distilled water) was also run along with experimental flask.(Fig. 2) After 24, 48 and 72 hours of time interval culture filtrate and Silver nitrate solutions turned colourless to dark brown colour due to reduction of Silver nitrate to Silver ions.

Characterization of synthesized silver nanoparticles

UV- Visible spectroscopy

UV-Visible spectroscopy is simplest way to confirm the formation of nanoparticles. The reduction of Silver ions was confirmed by qualitative testing of supernatant by UV- Visible spectrophotometer. The UV –Visible spectroscopy measurements were performed on Elico spectral photometer as a resolution of 1nm from 200 to 800 nm with distilled water as blank reference.

XRD study

Powdered sample was used for X-ray diffraction; analysis for silver nanoparticles was performed by using monochromatic Cu $\text{K}\alpha$ radiation ($\lambda=1.5406 \text{ \AA}$) operated at 40 kV and 30 mA at 2θ angle pattern. The Coherently diffracting Crystallography domain size of the Silver nano particle was calculated from the width of the XRD peaks using scherrer formula.

TEM analysis

Samples were prepared for Transmission electron microscopic Analysis (IIT Mumbai) TEM Technique was employed to see the size and shape of the synthesized silver nanoparticles, the dilute drops of suspension were allowed to dry slowly on carbon-coated grids for TEM measurement

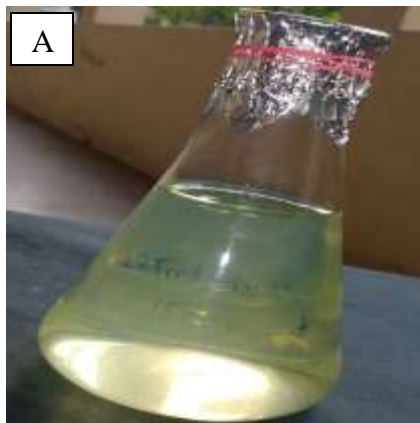
RESULTS AND DISCUSSION

It was observed that there is variation in the particle sizes around 30% of particles in 25 nm range and 25% in 30 nm range and 20% in 35 nm ranges. The particles range from 12 nm least to 75 nm high, the TEM image suggests that the particles are polydispersed and are rounding spherical in shape.

Three different concentrations that are 5 ml, 10 ml, and 15 ml of *Moringa oleifera* plant extracts screened for Biological synthesis of Silver nanoparticles. Plant extract was treated with 1 Mm Silver nitrate in 100 ml conical flask the reduction of silver ion into silver nanoparticles during exposure to plant extract was followed by changing color, colorless to dark brown. It is known that silver nanoparticles exhibits brown color in aqueous solution due to excitation of surface plasmon vibrations in Silver nanoparticles. Interestingly, 10 ml and 15 ml concentration plant extracts were changed the color within 24 hours from colorless to brown whereas 5 ml concentration plant extract changed the color within 72 hours the UV

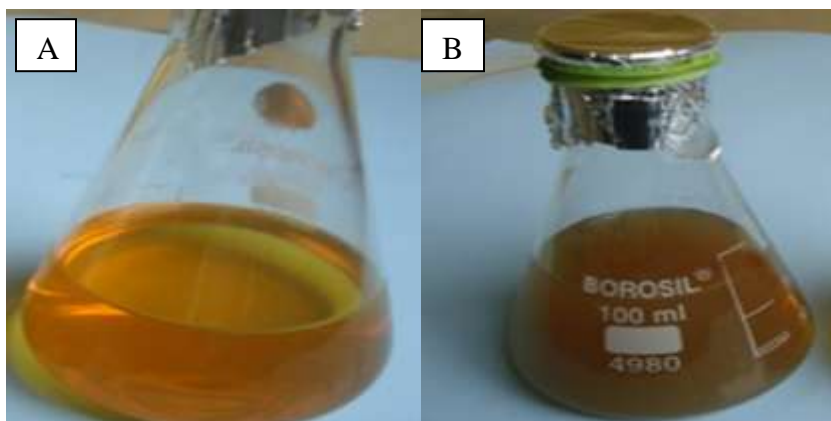
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VIS-Spectroscopy of the synthesized silver nanoparticles were in the range of 420,425, and 430 respectively(Fig. 3).



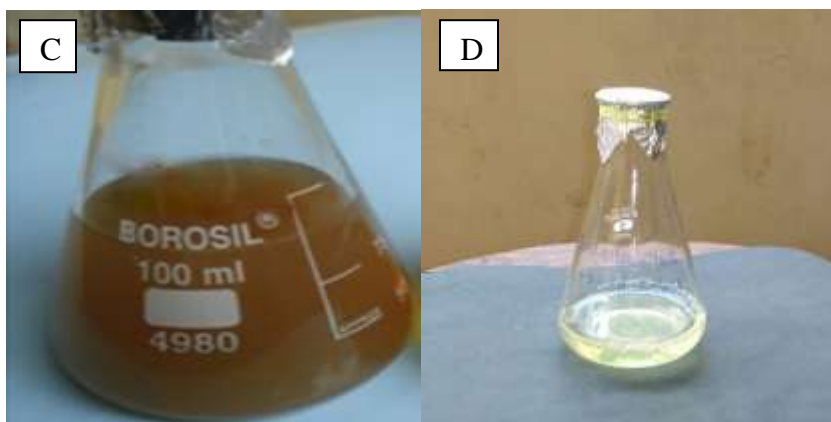
(A) 250ml plant extract

Fig 1: Aqueous plant extract of *Moringa oleifera*



(A) 50 ml $AgNO_3$ +5ml plantextract

(B) 50 ml $AgNO_3$ +10ml plantextract



(C) 50 ml $AgNO_3$ +15ml plantextract

(D) 50 ml DW+15ml plantextract

Fig 2: Biosynthesis of silver nanoparticles-color change reaction: conical flask containing the aqueous plant extract of *Moringa oleifera*

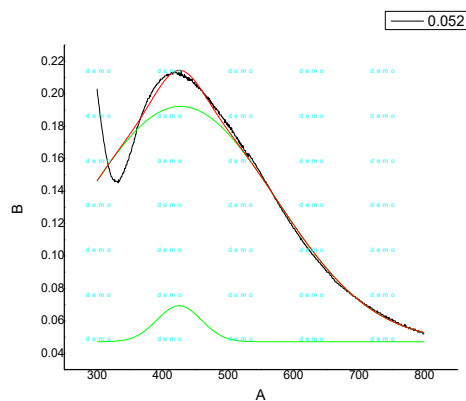


Fig 3: UV-Vis spectrum of silver nanoparticles synthesized using *Moringa oleifera* plant extract. UV-Vis spectra recorded as function of time of reaction of an aqueous solution of 1mM silver nitrate solution with the plant filtrate. The time of reaction is indicated next to the respective curves.

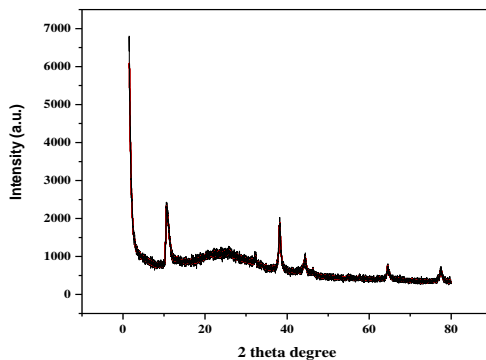


Fig 4: XRD analysis, peaks assigned to the corresponding diffraction signals (111), (200), (220), and (311) facets of Silver.

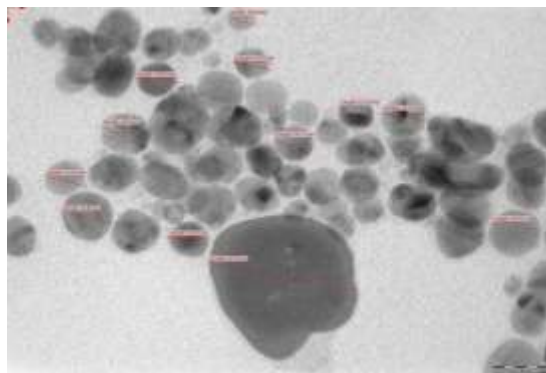


Fig 5: Transmission electron microscopic photographs of synthesized silver nanoparticles from *Moringa oleifera* leave extract.

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Table 1: UV-VIS Spectrum analysis shows time interval for changing color of plant extracts.

Plant extracts & concentration	Time taken for reduction	Uv – peaks in nm	Colour
<i>Moringa oleifera</i> 5 ml	72 hours	430-490	Colorless- Brown
<i>Moringa oleifera</i> 10 ml	24 hours	420-470	Colorless- Brown
<i>Moringa oleifera</i> 15 ml	24 hours	420-470	Colorless- Brown

XRD study

Obtained Silver nanoparticles were purified by repeated centrifugation at 3000 rpm for 40 minutes by redispersing silver nanoparticles pellet into 10 ml double distilled water. After drying silver nanoparticles in room temperature structure and composition analysis was carried out by XRD (Fig. 4) The crystallite domain size was calculated by the width of the XRD peaks using Scherer formula $D=0.96 \lambda/\beta \cos \theta$, where D is crystalline domain size perpendicular to reflecting planes, λ is the x-ray wavelength, β is the full width at half maximum and θ is the diffraction angle.

The average particle size was 30-35 nm. XRD analysis, peaks assigned to the corresponding diffraction signals (111), (200), (220), and (311) facets of Silver. The mean particle diameter of silver nanoparticles was calculated from the XRD pattern according to the line width of the (111) plane.

TEM Analysis

Sample was prepared for Transmission electron microscopic Analysis (IIT Mumbai) TEM Technique was employed to see the size and shape of the synthesized silver nanoparticles; it was observed that there is variation in the particle sizes around 30% of particles in 25 nm range and 25% in 30 nm range and 20% in 35 nm ranges. The particles range from 12 nm least to 75 nm high, the TEM image suggests that the particles are polydispersed (Fig. 5) and are rounding spherical in shape.

Conclusion

In the present study Silver nanoparticles were Green synthesized using *Moringa oleifera* plant extract. The plant extract in different concentration i.e. 5 ml 10 ml and 15 ml are challenged with 1mM Silver nitrate; change of mixture from color less to dark brown indicates the synthesis of Silver nanoparticles in the reaction mixture. And the crystallite domain size of synthesized silver nano particles was measured 30-35 nm by XRD analysis, shape and size of the silver nanoparticles was studied by TEM analysis. Results conclude that citrus limon plant extract is potential producer of Silver nano particles

ACKNOWLEDGEMENT

Authors wish to thank to Gulbarga University Gulbarga, Karnataka, India. And also thankful to USIC and Physics departments (G.U.G) for UV spectrum and XRD analysis and IIT Mumbai for TEM analysis

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