

Ecofriendly Synthesis and Characterization of Silver Nanoparticles from *Azadirachta indica* and *Ocimum sanctum* leaf extracts

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Abstract— The development of Nanotechnology and Nanoscience is creating the significance of researchers towards the synthesis of nanoparticles for the various applications like medicine, pharmaceutical and environmental management studies, etc. The advancement of simple, cost effective and reliable methods for the synthesis of nanomaterial is being emphasized worldwide. The major methods for the synthesis of nanoparticles are physical and chemical approaches that are often costly and potentially harmful to the environment. The present study is constant to the prospect of metal nanoparticle synthesis using plant extracts. Here silver nanoparticles were synthesized using *Azadirachta indica* and *Ocimum sanctum* leaf extract. The leaf extract acts as both reducing and capping agents. The synthesized silver nanoparticles were confirmed by the colour change after the addition of a leaf extract into the Silver nitrate solution. The synthesized silver nanoparticles were preliminarily characterized by using UV-Vis analysis and Fourier Transform Infrared Spectroscopy (FTIR), SEM image.

Key words: Capping agent, FTIR, *Azadirachta indica* and *Ocimum sanctum*

I. INTRODUCTION

Nanotechnology plays a very important role in modern research, it is the most capable technology that can be applied to almost all fields such as pharmaceutical, electronics, health care, food and feed, biomedical science, drug and gene delivery, chemical industry, energy science, cosmetics, environmental health, mechanics and space industries¹. The widespread practical application of metal nanoparticles (particles less than 100nm) is attributable to a number of their unique properties². Nanoparticles are synthesized by physical and chemical methods; these are suffering from drawbacks like expensive reagent, hazardous reaction condition, and longer time tedious process to isolate nanoparticles³. They can be synthesized by using different methods, including CO₂ laser pyrolysis⁵, Micro emulsion-based method⁶, Evaporation condensation, laser ablation⁷, Electrochemical method⁸, Chemical vapor synthesis method⁹. Green synthesis provides advancement over chemical and physical method as it is cost effective, environment friendly, easily scale up for large scale synthesis and in this method there is no need to use high pressure, energy, temperature and toxic chemicals⁴.

It has long been known that plants are able to reduce metal ions, both on their surface and in various organs and tissues remote from the ion penetration site. In this regard, plants (especially those which have very strong metal ion hyper accumulating and reductive capacity) have been used for extracting precious metals from land which would be economically unjustifiable to mine; an approach known as phytomining. The metals accumulated by the plants can be

recovered after harvesting via sintering and smelting methods. Interestingly, a study of the metal bioaccumulation process in plants has revealed that metals are usually deposited in the form of nanoparticles². Green synthesis offer better manipulation, control over crystal growth and their stabilization. This has motivated an upsurge in research on the synthetic routes that allows better control of shape and size for various Nano technological applications⁴. *Azadirachta indica* commonly known as Neem belongs to Meliaceae family, and is well known in India and its neighboring countries for more than 200 years as one of the most versatile medicinal plant having a wide spectrum of biological activity. Every part of the tree has been used as a traditional medicine for a household remedy against various human ailments, from antiquity¹⁰. *Azadirachta indica* leaf extract has also been used for the synthesis of silver, gold and bimetallic (silver and gold) nanoparticles. The major advantage of using the neem leaves is that it is a commonly available medicinal plant and the antibacterial activity of the biosynthesized silver nanoparticle might have been enhanced as it was capped with the neem leaf extract¹¹. *Ocimum sanctum*, a wild herbaceous plant is very common in all tropical countries, including India. The stems are and often reddish in colour, covered with yellowish bristly hairs, especially in the younger parts. The leaves are oppositely arranged, lanceolate and are usually greenish or reddish, underneath measuring about 5cm long. The stem and leaves produce white or milky juice when cut¹². The plant has been widely acknowledged for the treatment of cough, hay asthma, bronchial infections, bowel complaints, worm infestations, kidney stones in traditional medicine¹³. Hence the aim of the present study is to synthesize silver nanoparticles by using aqueous neem extract and characterize. Thereby improving the importance of plant source and involving green chemistry for the synthesis of other nanoparticles as future research.

II. MATERIALS AND METHODS

A. Materials:

All the reagents used in this experiment were obtained from Tamilnadu, India. Distilled water was utilized for this process. Filtration was done by using Whatman no.1 filter papers. Glassware used for the complete reaction was washed thoroughly and rinsed with distilled water and dried in hot air oven.

B. Collection of Plant samples:

Neem and Tulasi leaves were collected from the local region. They were washed with distilled water and cleaned with water absorbent paper. Then it was cut into small pieces.

C. Preparation of *Azadirachta indica* and *Ocimum sanctum* leaf extract:

20 grams of sterile Neem and Tulasi leaves were added to 100 ml of distilled water and allow boiling for 60 minutes¹⁴. The broth extract was filtered and kept at 4°C for further use.

D. Synthesis of Nanoparticles:

10 ml of leaf extract¹⁵ was added to 90 ml of 1mM AgNO₃ solution³⁰ for the reduction of silver ions. Within a particular time change in colour¹⁰ from Greenish yellow to Black colour obtained by nanoparticles synthesis. A control setup was also maintained without adding silver nitrate to the plant extract. The formation of silver nanoparticles was also confirmed by UV-Vis spectrophotometric determination, FTIR and SEM³⁰.

E. Ultraviolet-Vis spectroscopy Analysis:

UV-Vis spectral analysis was done by using UV-Visible Spectrophotometer. The reduction of pure Ag⁺ ions was monitored the UV-Vis spectrum of the reaction medium after diluting a small aliquot of the sample into deionized water. 100 µl of the sample was pipetted into a test tube and diluted with 10 ml of deionized water and subsequently analyzed at room temperature¹⁵.

F. Fourier Transform Infra-Red Spectroscopy (FTIR):

For FTIR measurements, the silver nanoparticle solution was centrifuged at 20,000 rpm for 20 minutes¹⁶. The pellet was washed three times with 20 ml of deionized water at least three times. It was analyzed with FTIR of finally dried resultant purified suspension after the ground KBr pellets¹⁷.

G. Scanning Electron Microscopy (SEM):

In order to visualize dispersing effects, SEM images of raw material of Ag nanoparticles from Neem and Tulasi leaf extracts were prepared. This preparation comprised of centrifuging the samples (13,000 rpm for 20 min), discarding the supernatant, drying the pellet overnight, and resuspending the pellet in ethanol. One drop of the obtained suspension was put on a glass slide²⁵. 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 min²⁶.

exhibit yellowish brown colour in water and this colour arise due to surface Plasmon vibration of metal nanoparticles^{18, 20}. The formation of silver nanoparticles was observed by UV-Vis spectra 200nm to 600nm where the peak was observed at 341nm for Neem and 252nm and 288nm for Tulasi. (Fig 3a & Fig 3b) shows the absorption spectra of the *Azadirachta indica* and *Ocimum sanctum* Leaf extracts and the peaks. This band was recognized as a surface Plasmon resonance band and ascribed to the excitation of free electrons in the nanoparticles. The result of UV-Vis spectroscopy was highly similar to Sajesh kumar et.al, 2015 and Yogeswari Rout et.al, 2011.



Fig. 1: AgNO₃ solution, Control Test Neem leaf extract and Tulasi leaf extract



Fig. 2a: AgNO₃ solution without adding the plant extract of Neem (Control), After the addition of AgNO₃ into the neem leaf extracts (Test)



Fig. 2b: AgNO₃ solution without adding the plant extract of Tulasi (Control), After the addition of AgNO₃ into the Tulasi leaf extracts (Test)

III. RESULTS AND DISCUSSION

A. UV-Vis spectroscopy analysis:

The comprehensive study of biosynthesis and characterization of silver nanoparticles using *Azadirachta indica* was carried out and is reported in this paper. An UV-Vis Spectroscopy is one of the important methods to study the formation of metal nanoparticles, provided Surface Plasmon Resonance exists for the metal. Fig.1 shows sample tubes containing AgNO₃ solution, *Azadirachta indica* leaf extract, *Ocimum sanctum* leaf extract and Fig.2 & Fig.3 shows the conical flask containing AgNO₃ solution after 10 mins of reaction time with the leaf extracts of Neem and Tulasi. It is experimental that the colour of silver nitrate solution changed from colorless to brown with increasing the intensity with the incubation time, its representing the formation of silver nanoparticles. It is well known that silver nanoparticles

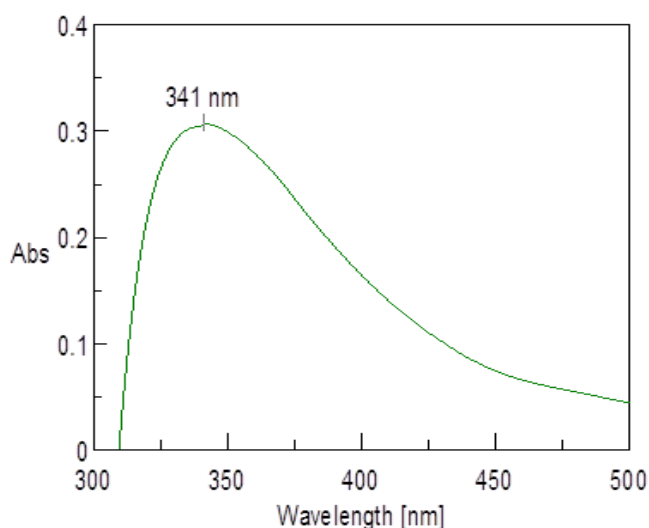


Fig. 3a): UV-Vis spectra of silver nanoparticles

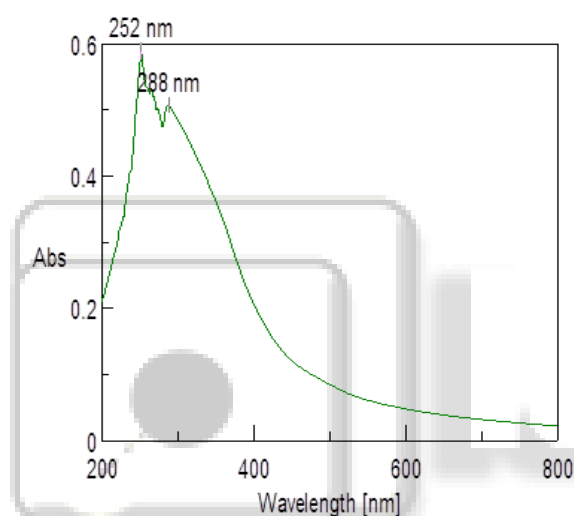


Fig 3b): UV-Vis spectra of silver nanoparticles from Neem from Tulasi

FTIR:

The infrared spectra are recorded on Fourier Transform Spectrophotometer in the mid-infrared region (MIR) within the range (400-4500 cm⁻¹). Due to the complex interaction of atoms within the molecule, IR absorption of the functional groups may vary over a wide range. However, it has been found that many functional groups give characteristic IR absorption at specific narrow frequency range. Multiple functional groups may absorb at one particular frequency range but a functional group often gives rise to several characteristic absorptions. Stretching and bending vibrations are varied after formulation can be observed. Thus, the spectral interpretations should not be confined to one or two bands only actually the whole spectrum should be examined²¹⁻²². The spectrum of silver nanoparticles from Neem shows (Fig: 4a) a peak formation at 3774.65 cm⁻¹ confirms the O-H stretch, a strong, sharp band for O-H stretch at 3411.69 cm⁻¹, medium stretch for =C-H at 2929.63 and 2867.62 cm⁻¹, primary amines have two bands, secondary have one band often very weak for N-H stretch at 1764.92 cm⁻¹, 1379.58 cm⁻¹ confirms the C-N stretch medium band and =C-H strong bending at 820 cm⁻¹ wave number which was compared with the control having a two broad strong

band at wave number of 3434.89 and 688.83cm⁻¹ and one sharp medium band at 1639.40cm⁻¹. The overall observation confirms the presence of alcohol, alkene and amines. The spectrum of silver nanoparticles from Tulasi (Fig: 4b) shows a broad medium band for O-H stretch H bonded and C=C stretch at 3438.16 cm⁻¹ and 1592.25 cm⁻¹ at 2366.73 cm⁻¹ the sharp medium band for O-H stretch, 1378.89 confirms the sharp band for C-F, 816.96 cm⁻¹ confirms the strong sharp band for =C-H bending and C-Cl stretch and C-Br stretch was at the wave number of 721.15 and 603.00 cm⁻¹. The similar FTIR results for Silver nanoparticles for Neem and Tulasi were already reported Silverstein et.al, 2002, Ashutosh Kar et.al, 2006, Pandia et.al, 2009.

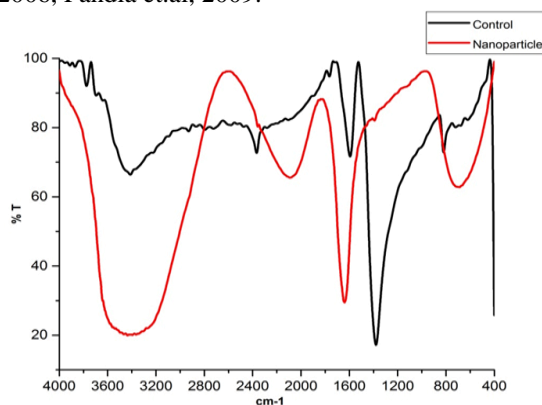


Fig 4a): Comparative FTIR spectra of synthesized silver nanoparticle from Neem leaf extract and Control (Neem leaf extract without adding AgNO₃ solution)

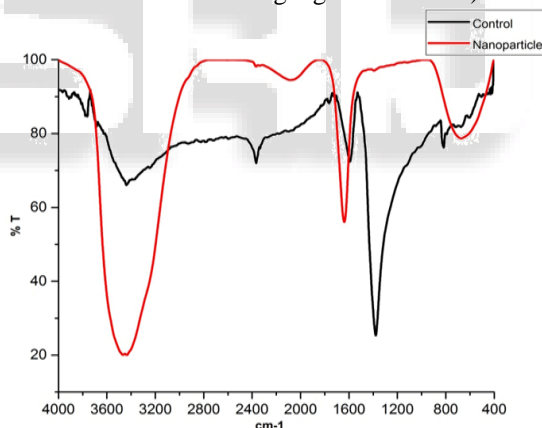


Fig 4b): Comparative FTIR spectra of synthesized silver nanoparticle from Tulasi leaf extract and Control (Tulasi leaf extract without adding AgNO₃ solution)

B. SEM:

The scanning electron microscopic was done using ZEISS model SEM machine. The magnification was done at 20μm, 10μm, 2μm. From the image of SEM micrographs we conclude that the nanoparticles are clustered. The particles are more (or) less spherical and ranges from 20-50 nm in size²⁵. The larger silver nanoparticles may be due to the aggregation of the smaller ones, due to the SEM measurements. The report shows that the silver nanoparticles synthesis from Neem and Tulasi are mostly spherical and cubic in shape and different sizes (Figures 6 & 7). Similar phenomenon was reported by Sajesh Kumar²⁶ et.al, 2015,

Akl M Awwad28 et.al, 2013, Yogeswari Rout27 et.al, 2012, Mallikarjuna29 et.al, 2011.

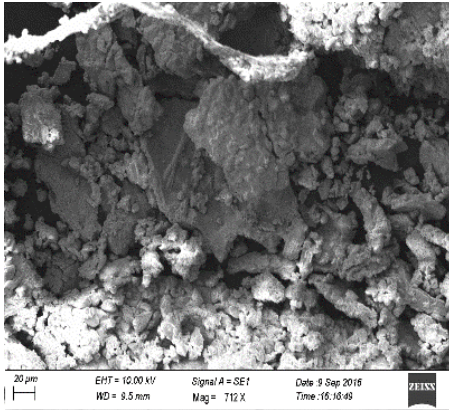


Fig.6a: SEM image of silver nanoparticles from Neem at 20µm

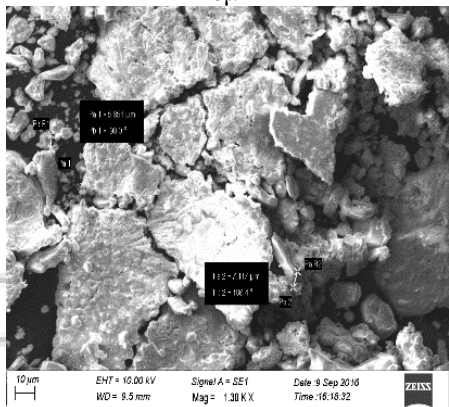


Fig 6b: SEM image of silver nanoparticles from Neem at 10µm

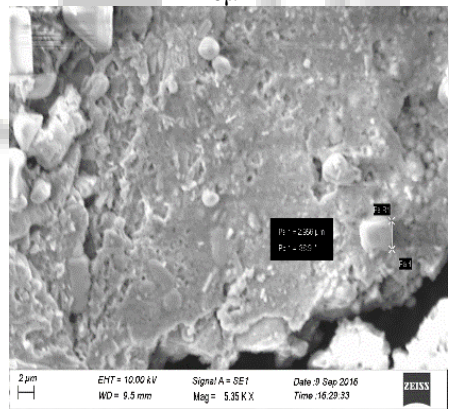


Fig. 6c: SEM image of silver nanoparticles from Neem at 2µm

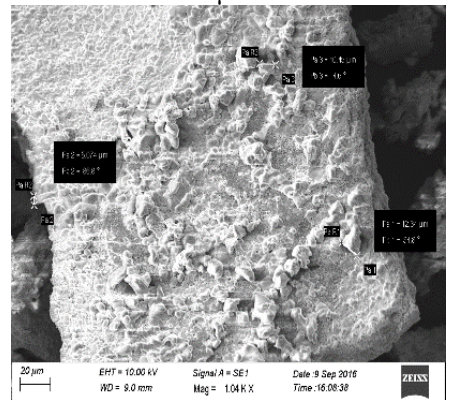


Fig7a: EM image of silver nanoparticles from Tulasi at 20µm

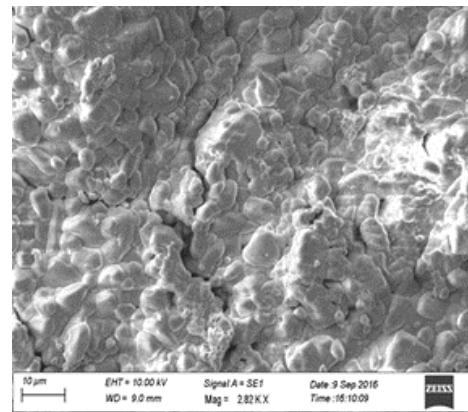


Fig. 7b: SEM image of silver nanoparticles from Tulasi at 10µm

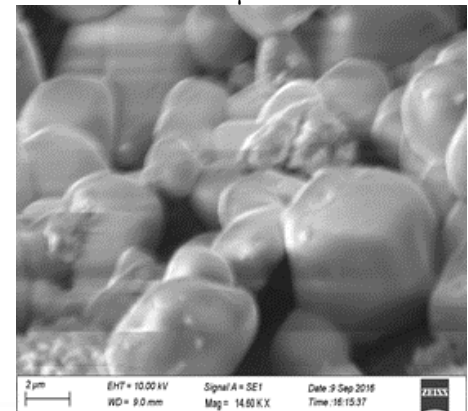


Fig. 7c: SEM image of silver nanoparticles from Tulasi at 2µm

IV. CONCLUSION

We have developed a fast, cheap, simple and green method for the synthesis of nanoparticle using *Azadirachta indica* and *Ocimum sanctum*. The bioreduction of Silver ions using leaf extracts of *Azadirachta indica* and *Ocimum sanctum* as reducing agent has been established. The synthesized nanoparticles were characterized by using UV-Vis Spectroscopy, FTIR and Scanning Electron Microscopy (SEM).

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