

Green Synthesis of Nanosilver Using an Ayurvedic Medicinal Plant *Bauhinia acuminata*

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ABSTRACT: Green chemistry is one of the burning research topics to researchers. The aim of the present work is to synthesis silver nanoparticles using bio reduction method mediated by a commonly seen plant in Kerala, *Bauhinia acuminata*. The nanosilver particles were synthesized by adding plant leaf extract to 1mM AgNO₃ solution. The Phytochemicals present in the leaf extract were qualitatively analyzed. The phytochemicals present in the leaf extract reduced the Ag⁺ ions in the silver nitrate solution to nanosilver. The formation of Ag nanoparticles was confirmed by UV- Vis spectroscopy, XRD, SEM, TEM, EDX, etc. The UV-Vis spectra have shown an absorbance maximum at 420nm, which is characteristic for Ag nanoparticles. The surface morphology of Ag nanoparticles was identified using SEM. The particle size of synthesized nanosilver was found to be 22nm by TEM analysis. EDX spectra shows excellent purity for synthesized silver nano particles. The different phases and crystalline size of silver nano particles were identified using XRD technique.

KEYWORDS: Bio reduction, Crystalline Size, Green Chemistry, Phytochemicals, Nanosilver,

I. INTRODUCTION

Nano particles are the particles having the size between 1- 100nm. They are having different properties than the bulk material of the same element due to variation in size, shape and distribution of the particles. Among all the noble metals nano silver is most important that it posses good conductivity, chemical stability, catalytic and antibacterial activity [1-5]. Due to these properties, silver nanoparticles can be used as catalysts in chemical reactions, optical elements, and in medical field.

There are various methods available for formation of Ag-NPs such as chemical reduction , heat evaporation , solvo thermal reduction, photochemical method , electrochemical technique , reverse micelles , microwave assisted methods, sol gel process etc [6-13]. Green chemistry is the sustainable chemistry that encourages the design of products and processes that minimize the use and generation of hazardous substances. Green methods are cost effective and environmentally friendly. The green synthesis of nano silver is available in the literature [14-19].

In this study green synthesis of silver nano particle was carried out using the aqueous leaf extract of *Bauhinia acuminata*, a commonly seen plant in Kerala. In Ayurveda, the plant leaf, root, bark, flower extracts were used to treat several diseases. The synthesized nano particles were characterized using UV-Vis spectroscopy, SEM, TEM, XRD, EDX etc.

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Fig 1: Image of *Bauhinia acuminata* plant

II. MATERIALS AND METHOD

Synthesis of nanosilver

10g of *Bauhinia acuminata* leaves were washed with distilled water and chopped into small pieces. It is transferred into 75ml distilled water in a beaker and boiled for 3minutes. It is filtered through a good quality filter paper.10mL of the leaf extract is mixed with 90mL, 0.001M silver nitrate solution in a conical flask and heated at 40°C until the colour changes to reddish brown.

Characterization of silver nano particles

UV-Vis Analysis: The optical property of Ag nano particles was determined by UV-Vis spectrophotometer (Schimadzu UV spectrophotometer). After the addition of AgNO₃ to the plant extract, the spectra were taken after 24h, between 350 nm to 500 nm.

SEM Analysis: The morphological features of synthesized silver nanoparticles from *Bauhinia* leaf plant extract were studied by Scanning Electron Microscope (JSM-6480 LV). The SEM slides were prepared by making a smear of the solutions on slides. A thin layer of platinum was coated to make the samples conductive. Then the samples were characterized in the SEM at an accelerating voltage of 20 KV.

XRD Analysis: Samples for X-ray powder diffractometer were first finely ground and then mounted on a glass slide. The sample was analyzed in a Bruker AX S D8 advance X-ray powder diffractometer.

EDX Analysis: It was done by JEOL-MODEL 6390 machine.

TEM Analysis:

Samples for TEM were prepared by drop-coating the Ag nanoparticle solutions on to carbon –coated copper grids. Micrographs were obtained using an EM 900 ZEISS transmission electron microscope.

PHYTOCHEMICAL SCREENING

Preliminary phytochemical screening was performed as per standardized procedure.

Alkaloids (Mayer's Test)

0.5 g of the extract was stirred with few ml of dilute hydrochloric acid and filtered. To a few ml of filtrate, one or two drops of Mayer's reagent were added to the sides of the test tube. A white creamy precipitate demonstrated the test as positive.

Steroids (Salkowski's Test)

0.5g of the extract was dissolved in 2ml of chloroform. Sulphuric acid was then carefully added to form a lower layer. A reddish-brown colour at the interface indicated the presence of steroids.

Flavonoids (Ferric chloride Test)

0.5 g of the extract was boiled with distilled water and then filtered. To 2 ml of the filtrate, few drops of 10% ferric chloride solution were then added. A green-blue or violet coloration indicated the presence of flavonoids.

Tannins (Neutral Ferric chloride Test)

0.5 g of the extract was stirred with 10ml of distilled water, filtered, and ferric chloride reagent was added to the filtrate. A blue-black, green or blue-green-precipitate was taken as evidence for the presence of tannins.

Phenols (lead acetate Test)

0.5 g of the extract was treated with lead acetate solution. Formation of precipitate indicated the presence of phenols.

Glycosides (Salkowski's Test)

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0.5 g of the extract was dissolved in 2ml of chloroform. Sulphuric acid was then carefully added to form a lower layer. A reddish-brown color at the interface demonstrated the presence of glycosides.

Saponin (Frothing Test)

To 1g of the extract about 3 ml of distilled water was added and shaken vigorously for about 5 min. frothing which persisted on warming was taken as an evidence for the presence of saponins.

Phlobotannins (Aqueous HCl Test)

Deposition of a red precipitate when an aqueous extract was boiled with 1% aqueous hydrochloric acid indicated the presence of phlobotannins.

Anthraquinones ((Borntrager's Test)

About 0.2 g of each portion to be tested was shaken with 10 ml of benzene and then filtered. 5ml of the 10% ammonia solution was then added to the filtrate. Appearance of a pink, red or violet color in the ammoniacal (lower) phase was taken as the evidence for the presence of free anthraquinones.

III. RESULTS AND DISCUSSION

Phytochemical screening

The result obtained for phytochemical screening of *Bauhinia acuminata* is given below.

Phytochemicals	Aqueous Leaf Extract
Tannin	+
Phlobotannins	-
Saponin	+
Flavonoids	-
Steroids	-
Terpenoids	+
Cardiac glycosides	+
Alkaloids	-
Phenols	+
Anthraquinol	-
Carbohydrate	-

Table 1: Qualitative analysis of phytochemicals in the leaf extract of *Bauhinia acuminata*

(+) – Indicates the presence of phytochemicals

(-) – Indicates the absence of Phytochemicals

Change in colour of the reaction mixture

It was observed that, after the addition of leaf extract to the AgNO₃ solution the colour changes from pale yellow to reddish brown, confirms the formation of Ag nanoparticles. The reddish brown color is due to the surface Plasmon resonance (SPR) of metal nanoparticles.

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Fig 2(a): AgNO₃ solution with leaf extract



Fig 2(b): Synthesized Ag nanoparticles

UV-Vis Spectrophotometer Analysis

Reduction of silver ions into silver nanoparticles during exposure to plant extracts was observed as a result of the color change. The reddish brown color is due to the Surface Plasmon Resonance phenomenon. The metal nanoparticles have free electrons, which give the SPR absorption band, due to the combined vibration of electrons of metal nanoparticles in resonance with light wave. The sharp bands of silver nanoparticles were observed around 421 nm, which is characteristic for Ag nanoparticles. The reduction of the metal ions occurs fairly rapidly; more than 90% of reduction of Ag⁺ ions is complete within 24 h.

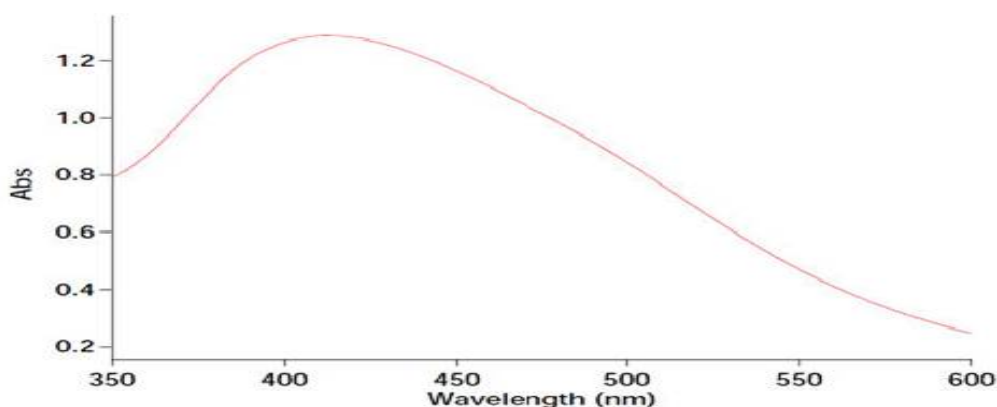


Fig 3: UV-Vis spectra of synthesized Ag nanoparticles after 24h.

XRD Analysis

The XRD analysis also confirms the formation of silver nano particles. The spectra shows diffractions at $2\theta = 24^\circ, 27^\circ, 32^\circ, 38^\circ, 44^\circ, 46^\circ, 64^\circ, 77^\circ$. Using Debye Scherrer equation, $D \approx 0.9\lambda/\beta\cos\theta$; Where D is the crystal size, λ is the wavelength of X-ray, θ is the Bragg's angle in radians and B is the full width at half maximum of the peak in radians, the average crystallite size was found to be 16.89nm. The lattice strain was found to be 0.0066.

Nano Silver

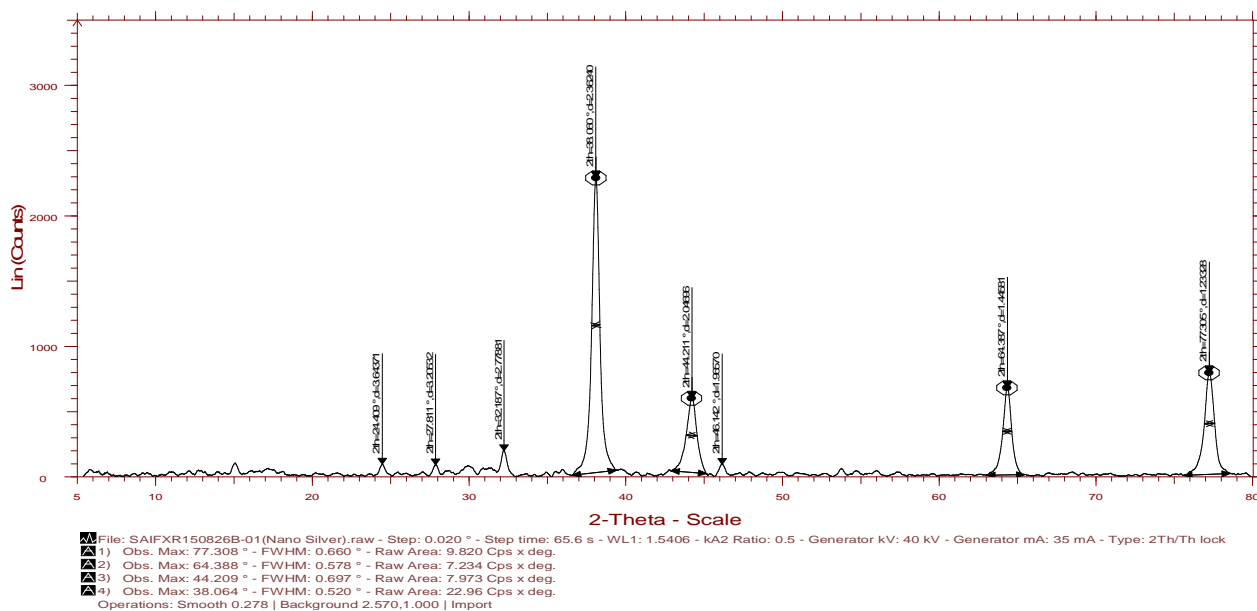


Fig 4: XRD of synthesized silver nano particles.

SEM Analysis

SEM provided further insight into the morphology and size details of the silver nanoparticles. From the SEM analysis it was found that majority of the synthesized Ag nano particles were spherical in shape. The silver nanoparticles were very much dispersed and homogeneous.

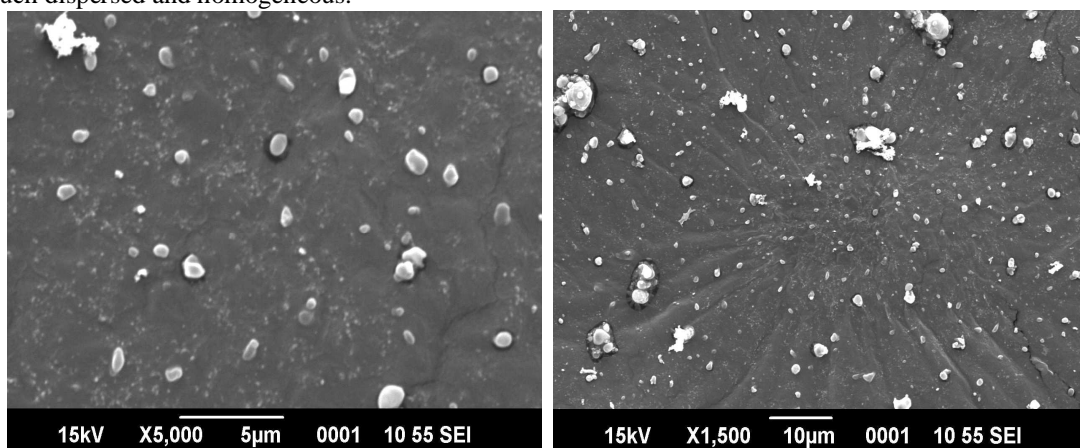


Fig 4: SEM images of the synthesized silver nanoparticles

EDX Spectra

The EDX spectra also confirms the formation of Ag nanoparticles. The spectra shows peaks only for Ag, that shows the high purity of synthesized Ag nanoparticles.

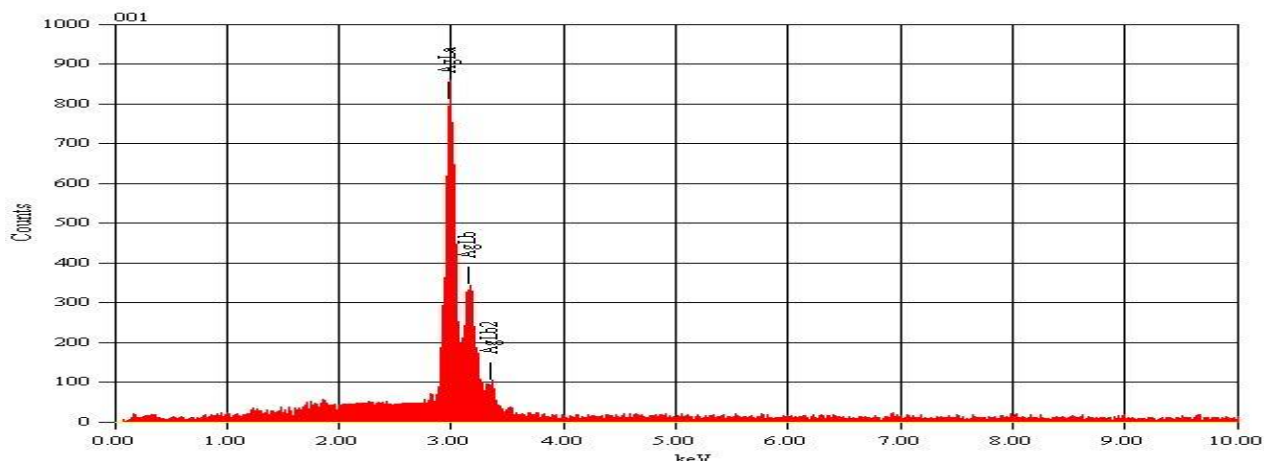


Fig 5: EDX spectra of synthesized silver nanoparticles

Element	(keV)	Mass%	Atom%	K
Ag L	2.984	100	100	1
Total		100	100	

TEM Analysis

The particle size of synthesized silver nano particles was determined using TEM. The analysis shows that the particle size ranges from 5-50nm and the average particle size is found to be 20nm. The particles are highly dispersed, spherical in shape and homogenous. The diffraction pattern also shows perfect arrangement of silver nanoparticles in the crystal lattice.

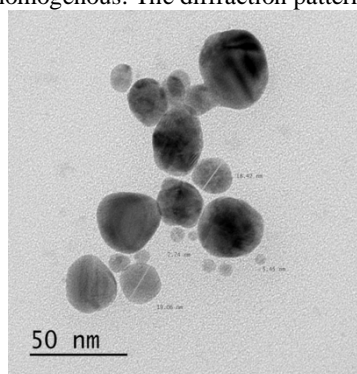


Fig 5: a

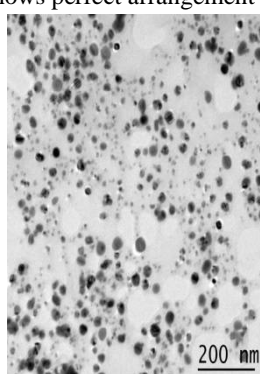


Fig 5: b

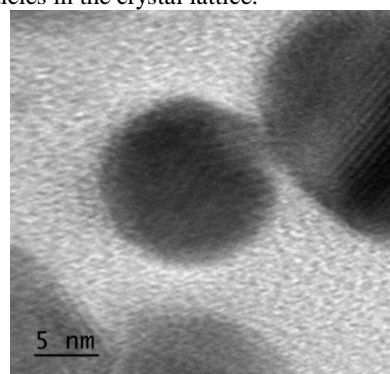


Fig 5: c

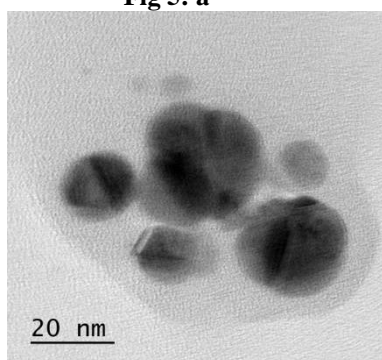


Fig 5: d

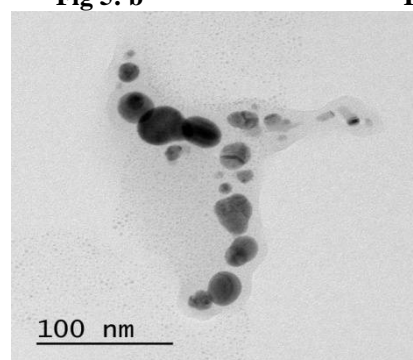


Fig 5: e

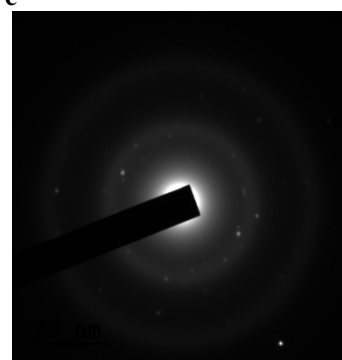


Fig 5: f

Fig 5 : TEM image of synthesized silver nanoparticles under different magnifications.

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1V. SUMMARY AND CONCLUSION

Nano silver particles can be synthesized by the green bioreduction method using the plant leaf extract *Bauhinia acuminata*. The reddish brown color of the product confirms the formation of silver nanoparticles and it is due to the surface plasmon vibrations of silver particles. The plant leaf extract contains different phytochemicals such as tannin, saponin, phenol etc. may cause the reduction of Ag ions into nanoAg. The UV-Vis spectrum shows an absorbance peak at 420nm which is characteristic for nano silver. The SEM analysis shows that Ag nanoparticles have spherical morphology. From The TEM analysis the particle size of silver nanoparticles ranges from 5nm-50nm and the particles are found to be highly dispersed.EDX spectra confirms the high purity of silver nano particles .From XRD analysis the crystallite size of silver nanoparticles is found to be 16.89nm.

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