

Research Article

Green Synthesis of Silver Nanoparticles using Spent Tea Leaves Extract with Atomic Force Microscopy

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Abstract

Green synthesis of nanoparticle is a unique method to synthesis nanoparticles by using biological sources. In this present study spent tea leaves extract was used synthesizing of AgNPs. Synthesizing of AgNPs was confirmed by changing their color to dark brown due to surface plasmon resonance (SPR) phenomenon. Process parameters such as AgNO₃ concentration (molarity), extract volume percent, sunlight exposure time and temperature were studied. UV-Vis spectroscopy showed the SPR peaks for AgNPs were between 370-435 nm using spent tea leaves extract. Atomic force microscopy (AFM) showed the average particle diameters of AgNPs were between 66 - 117 nm using spent tea leaves extract. Atomic Absorption Spectroscopy (AAS) showed the maximum extraction efficiency was 47% using spent tea extract.

Keywords: Green synthesis, Spent tea leaves extract, Silver Nanoparticles, AFM

1. Introduction

The synthesis of metal nanoparticles is a vast area of research due to its novel applications in many fields. Metal nanoparticles have unique physio-chemical properties and great applications in the fields of catalysts (Nair & Pradeep 2003), optics (Caro *et al.* 2010; Chien *et al.* 2008), magnetic (Laurent *et al.* 2010) and electrical (Lu & Chou 2008). Nanoparticles have very attractive optical properties. For example, a 20 nm gold nanoparticle has a wine red color (Chauhan *et al.* 2011), a yellowish gray for silver nanoparticles (Vankar & Shukla 2012) and dark color for platinum (Hei *et al.* 2012; Kanchana *et al.* 2010). Most of their physicochemical properties at the nanoscale, like a Localized Surface Plasmon Resonance (LSPR) has been employed for the development of new technology of biosensors (Doria *et al.* 2012).

Recently, biosynthetic methods are used to synthesize nanoparticles by employing naturally occurring reducing, capping, and stabilizing agents such as polysaccharides and biological microorganism (bacteria and fungus or plants extract), i.e. green chemistry (Abou El-Nour *et al.* 2010). Green chemistry is the development, implementation and design of chemical products and processes to eliminate or reduce the using and generation of hazardous substances on the environment and human health (Nadagouda & Varma 2008). Biological methods are an ecofriendly method used to synthesis of silver

nanoparticles without used of any toxic, expensive and harsh chemical substances (Korbekandi & Iravani 2010). Actually, the fit mechanism for metal nanoparticles synthesis by using biological substance or systems is not fully understood to this time (Rai *et al.* 2013).

Among the use of living organisms for nanoparticle synthesis, plants have found a larger area in application particularly in metallic nanoparticles synthesis. Using plants for synthesis of nanoparticles having many advantages over the chemical and microbe mediated synthesis method because they eliminate the culture maintaining process. They are safe to handle, easily available and hold a wide variability of metabolites, which may aid in reduction (Prathna *et al.* 2010). Nanoparticles produced by plants are more stable, are of various sizes and shapes. The rate of production is faster than in the case of microorganisms.

The synthesis of silver or gold nanoparticles by green methods consist of adding low concentration of gold or silver precursor to plant extract in solution to make up a final solution and centrifuged (Thakkar *et al.* 2010). The reduction of the Ag⁺ ions by plant extract occurs fairly rapidly. Plant parts like fruit, leaf, bark, seed, and stem extracts are being effectively used in green synthesis.

The objective of this work is to synthesis silver Nanoparticles (AgNPs) using Spent Black tea leaves extract as low cost material and ecofriendly environmental method. The effect of AgNPs synthesis

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parameters namely, Silver ion concentration, tea leaves extract concentration, and temperature have been studied. Also, the effect of electromagnetic radiation (sunlight) on AgNPs synthesis has been investigated. The AgNPs have been characterized by UV-visible spectroscopy, atomic absorption, AFM and FTIR will be used for characterization.

2. Materials and Methods

2.1 Materials

Silver nitrate (AgNO_3 · 99% purity, Sigma Chemical Co., Western Australia). Spent Black tea leaves were taken from home.

2.2 Preparation of Silver Nitrate Solution

Different molarities (1, 3, 5, 7 and 9 mM) of silver nitrate aqueous solution were prepared. The silver nitrate aqueous solution was used as precursor to synthesis AgNPs.

2.3 Preparation of Spent Black Tea Leaves Extract

The Spent black tea leaves used in this work was collected from waste of used tea at home. The spent tea leaves was washed with distilled water to remove any unwanted stakes, dried at room temperature and then stored to be used in tea extract preparation. A 10 g of dried spent tea leaves were boiled with 100 mL of distilled water at 80 °C for 5 minute (Salisu *et al.* 2014) in a 250 mL flask to obtained spent tea leaves extract. The resulted aqueous solution was filtered using Whatman filter papers (No. 2042-150) to obtained tea extract, which is yellow brown in color to be used in AgNPs synthesis.

2.4 Silver Nanoparticles Synthesis

A certain volume of tea leaves plant extract was added to AgNO_3 aqueous solution at specific molarities and mixed using hand mixer. The mix solution was allowed to react in sunlight for a specific time. Samples were then observed for the formation of AgNPs by observing the change in color of the mix solution.

2.5 Effect of Silver Nitrate Concentration on AgNPs Synthesis

A 10 mL of Spent Black Tea leaves extract is mixed with 90 mL of aqueous solution of AgNO_3 of different concentration (1, 3, 5, 7 & 9) mM in a 250 mL flask. The solution was allowed to react at room temperature under sunlight for 10 minutes. The sunlight intensity is 95800 ± 500 LUX (as measured by Note3 Samsung mobile at IOS 800). The color of the solution mixture of silver nitrate and Spent Black Tea leaves extract was changed within 10 minutes from yellow brown to deep brown color under sunlight. This denotes the reduction of Ag^+ ions to Ag nanoparticles (Awwad *et al.* 2013).

The solution mixture was analyzed using UV, AFM and Atomic absorption. The best results were then selected at best concentration of AgNO_3 solution.

2.6 Effect of Plant Extract Ratio on AgNPs Synthesis

The effect of plant Extract ratio on AgNPs synthesis was examined at fixed exposure time to sunlight. Different volume ratio (5, 7, 10, 12 and 15 %) of Tea leaves extracts were mixed with the best molarity of AgNO_3 aqueous solution. At this stage 5, 7, 10, 12 and 15 mL of Spent Black Tea leaves extract were added to 95, 93, 90, 88 and 85 mL of AgNO_3 solution respectively and wait 10 min for the color change under sunlight at room temperature. The extract and AgNO_3 solution mixture was then analyzed using UV, AFM and Atomic absorption. The best results were then took at best plant extract.

2.7 Effect of sunlight exposure on AgNPs Synthesis

The effect of sunlight exposure on AgNPs synthesis for Spent Tea leaves extract was studied at different exposure time. The best volume of plant extract was added to the best molarity of AgNO_3 solution at room temperature and wait for the color changed under sunlight at different time (5, 7, 10, 12 and 15) min for each sample. The solution mixture was then analyzed using UV, AFM and Atomic absorption. The best results were then took at best exposure time.

2.8 Effect of Temperature on AgNPs Synthesis

The effect of temperature on AgNPs synthesis was investigated at different temperatures (25, 40, 50, 60, 70 and 80) °C using Lab-Tech water bath (model: LSB-015S, LabTech Co., Korea). The best volume of leaves extract was added to the best concentration of AgNO_3 to synthesis AgNPs at sunlight exposure time of 10 min and wait for color change. Each samples was then cool down to room temperature and analyzed using UV, AFM and Atomic absorption. The best results were then took at best temperature.

3. Analysis Methods

3.1 UV-Visible Absorption

The formation of silver nanoparticles by reducing silver metal ion solution with Spent Black Tea leaves extract were initially analyzed using UV-Visible Spectrophotometers (Jenway 6310, Bibby Scientific Limited, UK). UV-Vis spectrometer was observed at wavelength 320-800 nm with 1 nm as wavelength resolution.

3.2 Atomic Absorption Spectrometry

The initial and the final concentration of Ag^+ in the prepared nanosilver solution were obtained using the Atomic absorption (Nov AA350 Analytik Jena AG,

Germany). The concentration of Ag⁰ nanoparticles was calculated by difference between the initial and final concentration of Ag⁺.

3.3 Atomic Force Microscopy

Atomic Force Microscopy (CSPM-5500, Karaltay (Beijing) Instruments Co. Ltd., China) was used to analyze AgNPs. A thin film of prepared AgNPs was deposited on a silica glass plate by dropping few drops of the AgNPs on the plate and allowed to dry at room temperature in the dark. The deposited film glass plate was then scanned with the AFM.

3.4 Fourier-Transform Infrared Spectroscopy

The Spent Black Tea leaves extract were analyzed using Fourier-transform infrared (FTIR) Spectroscopy, (IRTracer-100 / Shimadzu Co. / USA). Several drops of freshly prepared Spent Black Tea leaves extract were gas bubbles are trapped.

4. Results and Discussion

4.1 FTIR of Spent Black Tea Leaves Extract

The spectra for spent tea leaves extract were obtained using an FTIR spectrophotometer (IRTracer-100, Shimadzu Co. Japan) as shown in Figure 1. Several peaks were observed indicating the spent tea is composed of various functional groups (Figure1). The broad band at about 3448.72 cm⁻¹ can be attributed to bond -OH groups. The band at about 2356.89 cm⁻¹ can be attributed to C=N stretching. While a 2067.69 peak indicate an alkynes group -C≡C- stretching. A N-H bond Amine I groups are also observed at 1635.64 cm⁻¹. The peak at 1543.05 cm⁻¹ is attributed to secondary amine groups. The peaks at 1458.18 and 1396.46 cm⁻¹ are both related to the symmetric bending of CH₃. While the peak at about 1107.14 cm⁻¹ can be attributed to C=O stretching vibrations groups. The peak at 470.63 cm⁻¹ correspond to stretching vibration of amine groups (Zuorro *et al.* 2013; Shojamoradi *et al.* 2013; Vitro *et al.* 2014).

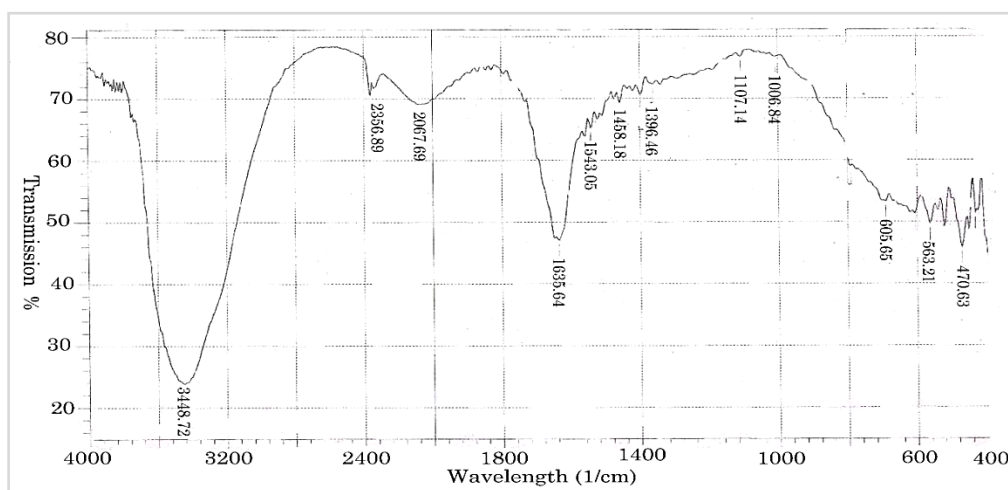


Figure 1 FTIR spectra of Spent Black Tea leaves extract

4.2 Green Synthesis of Silver Nanoparticles

4.2.1 Effect of AgNO₃ Concentration (Molarity)

To synthesis AgNPs, 10 mL of spent tea leaves extract was added to 90 mL of AgNO₃ aqueous solution at different concentration (1, 3, 5, 7 & 9) mM under sunlight for 10 minute at room temperature. The mix solution was characteristic using UV-visible and AFM.

Figure 2 shows the UV-visible spectra of AgNPs suspension at different concentration of AgNO₃ solution after dilution for five times. It is clear that increasing the molarity resulted in increasing of the absorbance spectra in the range for 1, 3 and 5 mM with shifting of SPR peak to longer wavelength direction. At 5 mM, the absorbance is maximum and the SPR band at 385 nm. For spent tea leaves extract and AgNO₃ solution, the SPR peaks was 370 – 385 nm for molarity between 1- 9 mM. With increase in morality to 9 mM,

the SPR peak is shifted toward the shorter wavelength with a decrease in the absorbance.

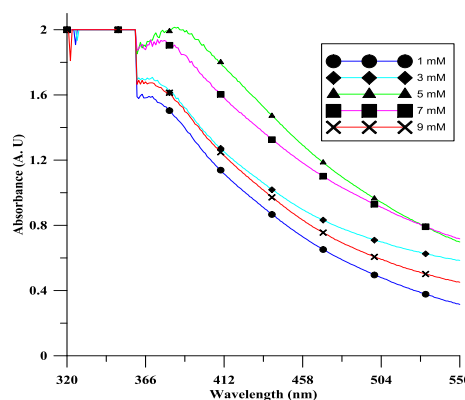
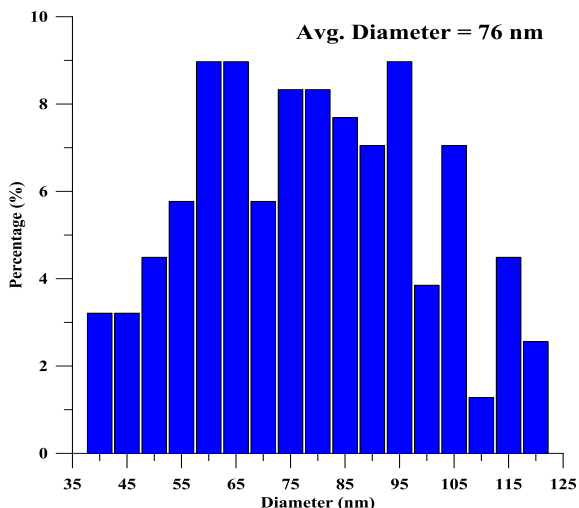
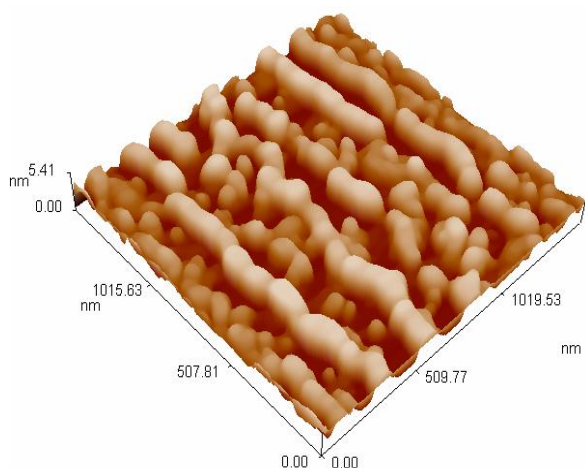


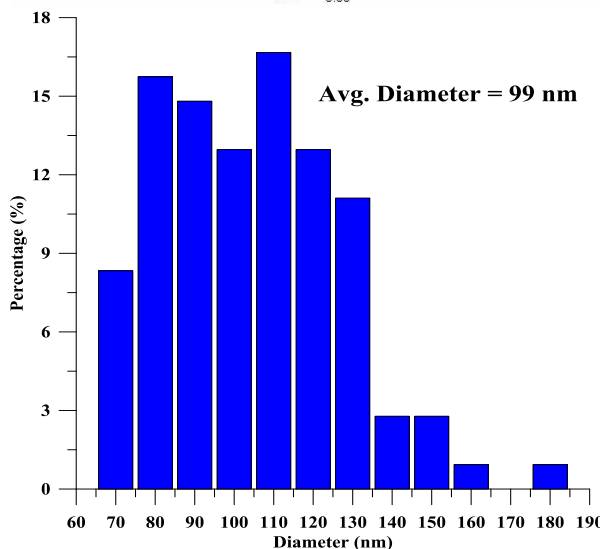
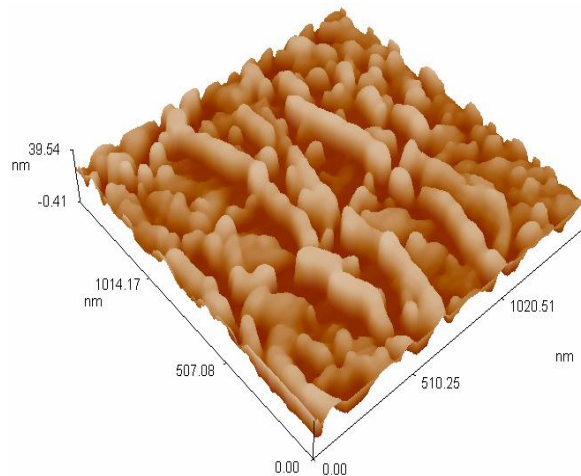
Figure 2 UV- Visible absorbance spectra of AgNPs suspension at different AgNO₃ concentration using Spent Black Tea leaves extract

This may be caused by increasing in destabilization of the particles as indicated by the decreased absorbance that may be due to increased particle aggregation and precipitation.

The AFM 3D images indicate the formation of homogeneous distribution of silver nanoparticles and no agglomeration was observed. Figure 3 shows the AFM images for (1, 5, 7 & 9) mM of AgNO₃ concentration where the particles shape and size was indicated. Some agglomeration was observed at 7 mM and 9 mM with large nanoparticles diameter. AFM shows the Granularity Accumulation Distribution (GAD), which gives the particles size distribution of AgNPs. The average particle diameter of the synthesized silver nanoparticles is 66, 76, 85 and 99 nm at 1, 5, 7 and 9 mM respectively. AFM image at 9mM, Figure 3 , indicates the particle diameter of the synthesized silver nanoparticles is between 70-180 nm. This large diameter could be due to sedimentation of the particles resulting in a decrease in absorbance (Delay *et al.* 2011).Therefore, the best AgNO₃ concentration (Molarity) for synthesis of AgNPs using spent tea leaves extract was taken at 5mM based on the absorbance, particles size and particles size distribution.



a) 5 mM



b) 9 mM

Figure 3 AFM images with nanoparticles size distribution of AgNPs synthesized using spent tea plant extracts: a) 5 mM AgNO₃ and b) 9 mM AgNO₃

4.2.2 Effect of Plant Extract Volume

The effect of different volume of plant extract for both Aloe Vera and spent tea leaves extract on AgNPs synthesis was investigated. A (5, 7, 10, 12 and 15 %) of plant extracts were added to the best molarity of AgNO₃ aqueous solution (5mM) for 10 min of sunlight exposure. Then the results were characterized using UV-Vis, AAS and AFM. Figure 4 shows the UV-Vis spectra of AgNPs suspension at different tea leaves extract (5-15 mL) where the samples were diluted for six time before UV-Vis characterization. The SPR peaks for 5 and 7 mL at 416 nm are very wide while SPR peaks for 10, 12 and 15mL extract are sharp and shifted to the longer wavelength direction at 428 nm. The maximum absorbance occurs at 10 mL extract with very narrow peak. Figure 5 shows the absorbance as a function of tea extract volume. The absorbance increases with increasing extract volume until the maximum was reached at 10% then start to decrease.

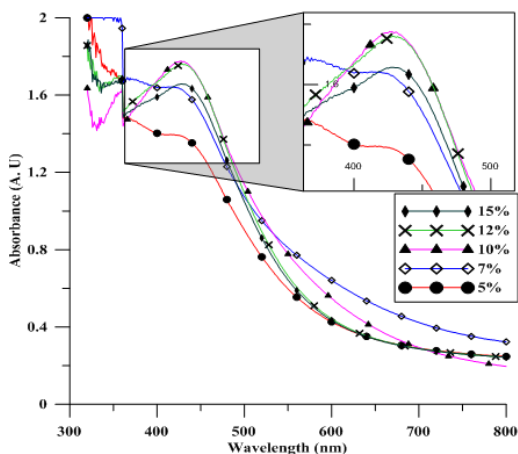


Figure 4 UV-Visible absorbance spectra of AgNPs at different spent tea extract volume

Atomic Absorption Spectroscopy shows the concentration of Ag^0 and the extract reduction efficiency as a function of spent tea extract volume as shown in Figure 6. The AgNPs were characterized by using Atomic Force Microscopy (AFM). The topographical images of AgNPs synthesized by using different spent tea leaves extract ratio are shown in Figure 7. The AFM images clearly indicate that the average nanoparticle diameter of the AgNPs samples was found to be 86 nm, 78 nm and 92 nm corresponding to 5%, 10% and 15% spent tea extract volume respectively. Large nanoparticles size distribution from 65 nm to 135 nm was observed at 5% spent tea leaves extract ratio. This is in agreement with absorbance spectrum of AgNPs at 5mL, Figure 4. The peak is very broad and the absorbance is very small due to particles aggregation. From AFM image, about 3% of the particles are aggregated at 130 nm size, Figure 4.22. For 10% and 15% spent tea extract ratio, the nanoparticles size distribution is less compared with 5% spent tea extract ratio. Therefore, the increase of spent tea extract ratio will cause less nanoparticles distribution and more average nanoparticles diameter. In this work, it seems that nanoparticle aggregates occurs at low spent tea extract volume (5%) and at high spent tea extract volume (15%). However, at spent tea extract volume (10%), the $AgNO_3$ nanoparticles distribution is uniform. This is clearly supported by the SPR peak, Figure 4, where the maximum absorbance occurs at 10% extract. Therefore, and upon the UV-Vis beaks, amount of AgNPs, best efficiency and acceptable particle size, the best extract volume percent for synthesis of AgNPs using spent tea leaves extract was taken at 10% extract volume.

4.2.3 Sunlight Exposure Time

An 80 mL of $AgNO_3$ solution at 5 mM (best) concentration was mixed with 10 mL (best) spent tea leaves extract volume at sunlight exposure time of 5, 7, 10, 12 and 15 min in order to study the effect of

sunlight on synthesis of AgNPs. Figure 8 shows UV-Vis spectra of AgNPs suspension at different sunlight exposure time. Increasing the sunlight exposure time will increase the the absorbance for (5-10 min). The SPR peak is sharper and reaches maximum at 10 min. At 12 and 15 min exposure time the absorbance will decreased and the SPR peak will become very broad.

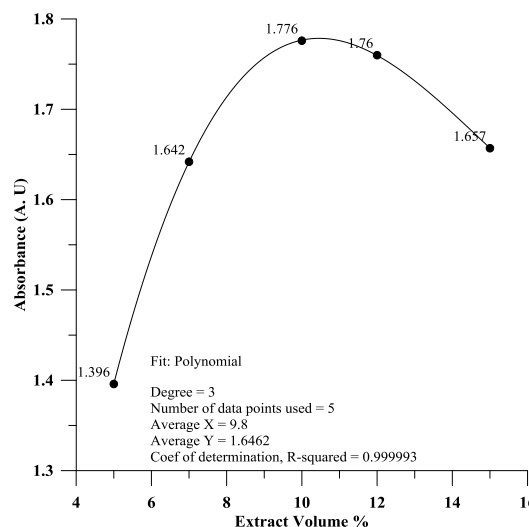


Figure 5 Absorbance as function of spent tea extract volume

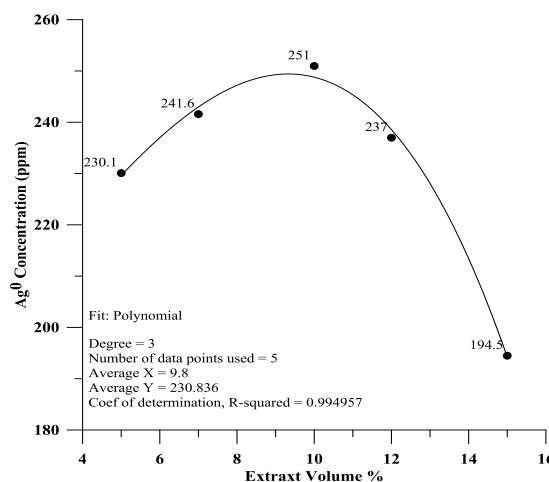
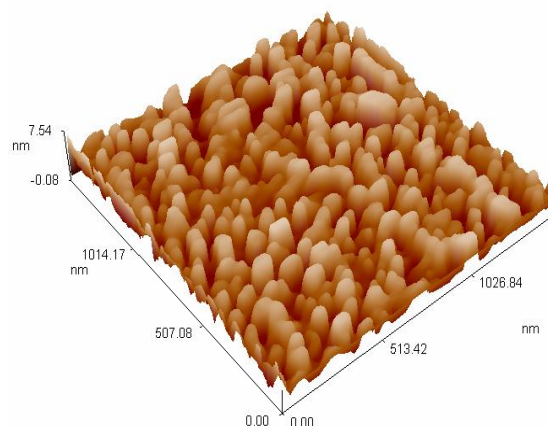


Figure 6 Ag^+ concentrations as a function of tea extract volume



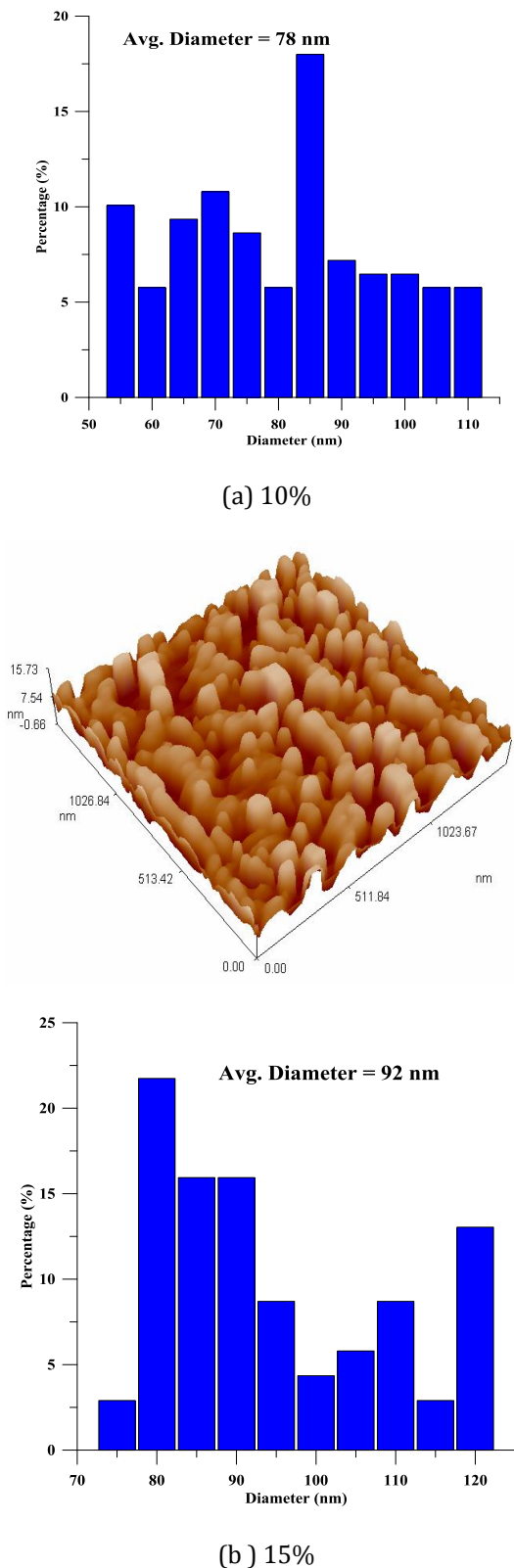


Figure 7 AFM images with nanoparticles size distribution of AgNPs synthesized using spent tea plant extracts: a) 10 % extract Volume ; b) 15% extract volume

Figure 9 shows the absorbance as function of sunlight exposure time using tea leaves extract. The absorbance increases with exposure time for (5, 7, 10 min) until reach a maximum at 10 min. At 12 and 15 min, the

absorbance starts to decrease. Atomic Absorption Spectroscopy results using tea leaves extract at different sunlight exposure time showed the Ag⁰ concentration reaches a maximum of 251 ppm at 10 min exposure time and then was sharply decreased at 12 min and 15 min exposure time. AFM images for AgNPs synthesis using spent tea leaves extract at different sunlight exposure time are shown in Figure 10. The image has an area of (1521 nm * 1540 nm) with (512*520) pixel which shows the topography of AgNPs. The 3D image show spherical nanoparticles shape with no agglomeration and uniform nanoparticles distribution for 5 min and 10 min of sunlight exposure time. Some agglomerations were observed at 15 min of sunlight exposure. The average nanoparticles diameter is 69 nm, 78 nm and 96 nm at 5, 10 and 15 min sunlight exposure time respectively. At 15 min large particles with diameter > 100 nm are formed. These particles will reduce the absorbance and this is supported by UV-Visible absorbance spectrum of AgNPs at 15 min sunlight exposure time, Figure 8. Therefore, and upon the UV-Vis beaks, amount of AgNPs, best efficiency and acceptable particle size, the best time of sunlight exposure for synthesis of AgNPs using spent tea leaves extract was taken at 10 min.

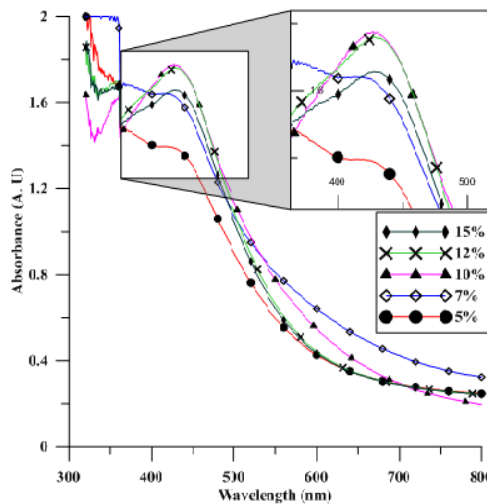


Figure 8 UV-Visible absorbance spectra of AgNPs at different sunlight exposure time using tea leaves extract

4.2.4 Effect of Temperature

A 80 mL of AgNO₃ solution at 5 mM (best) concentration was mixed with 10 mL (best) spent tea leaves extract volume for 10 min of sunlight exposure time to study the effect of temperature on synthesis of AgNPs. Figure 11 shows UV-Vis spectra for AgNPs suspension at different temperature 40, 50, 60, 70 and 80 °C after diluted for five time. Absorption spectra of silver nanoparticles formed in the reaction media have absorbance peaks between 430-435 nm and the absorbance decreases with increasing the temperature. The maximum SPR peak is 435 nm and occurs at 40 °C.

The spectrum surface plasmon resonance of nanoparticles is influenced by the size, shape, and free electron density, surrounding medium and inter particles interactions (Ghosh & Pal 2007). The variation in the peak intensity and bandwidth resulting from the varying size and size distribution of the particles. Although the band broadening are the same for all the temperatures 40, 50,60,70,80 °C, but the variation in the peak intensity are different. At 70 and 80 °C the absorbance are the lowest that can be attributed to aggregation (Desai *et al.* 2012).

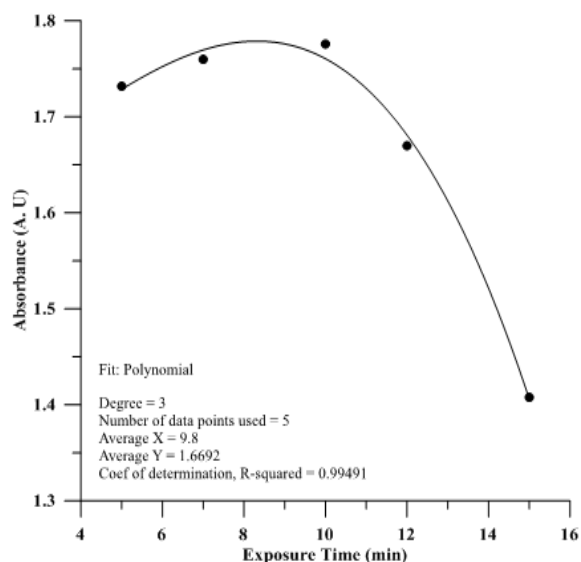


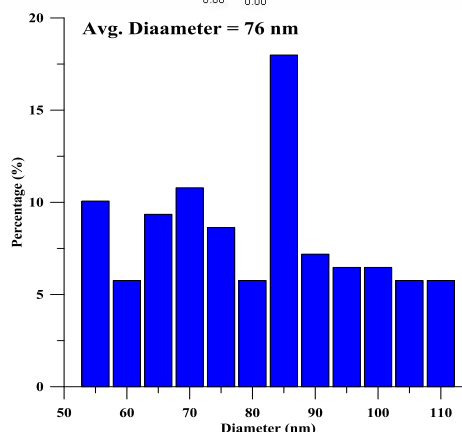
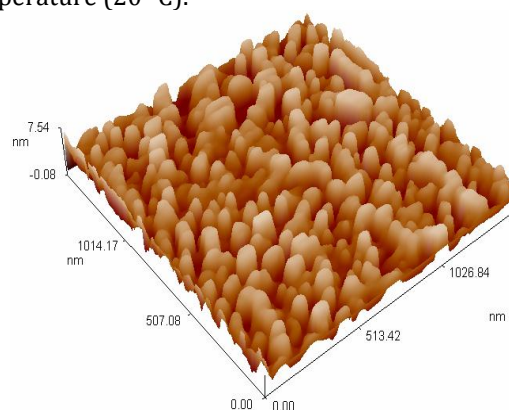
Figure 9 UV-Visible spectrums as function of exposure time

AFM images for AgNPs synthesis using spent tea leaves extract at different temperature are shown in Figure 12. The image has an area (1521 nm * 1549 nm) with (500*496) pixel shows the topography of AgNPs. At 40, 50, 60 and 70 °C the images show a spherical nanoparticles shape with no agglomeration. At 80 °C the image shows some agglomeration for some nanoparticles. The average nanoparticles diameter was 78.89, 82.41, 85.76, 90.53 and 101.64 at 40, 50, 60, 70 and 80 °C respectively. The particles diameter distribution for 40, 50, 60 and 70 °C is between 50- 135 nm, while at 80 °C the range from 100- 175 nm with present of 1.85% at 210 nm. According to above result, and upon the UV-Vis peaks, amount of AgNPs, best efficiency and acceptable particle size, the best condition for synthesis of AgNPs using spent tea leaves extract was taken at 5 mM AgNO₃ concentration (molarity), 10% extract volume percent, 10 min under sunlight exposure and room temperature (20 °C).

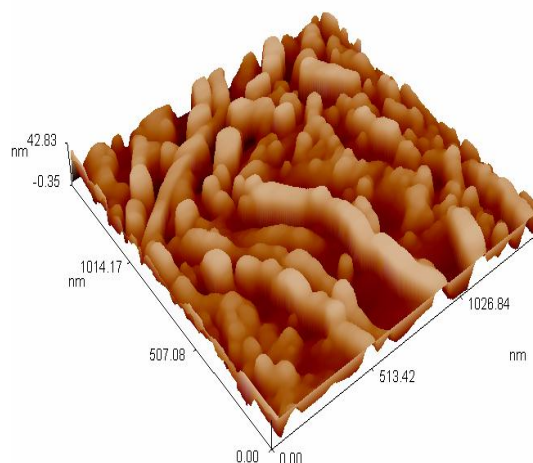
Conclusions

The green synthesis of AgNPs using spent tea leaves extract has many advantages namely waste material, simple, fast, economic, and ecofriendly method. Throughout this work, all the synthesis experimental for AgNPs were done under sunlight. AgNPs synthesis under sunlight is very efficient approach compare with that in the dark. It is well known that for AgNPs the

absorption band appears at about 380- 420 nm because of the surface plasmon resonance in Ag nanoparticles. The FTIR spectra for Spent Black Tea extract show several peaks and signify various functional groups. The UV-Vis was used to characterize the formation of silver nanoparticles from AgNO₃ solution mixture with plant leaves extract. The 3D images results by AFM indicate the formation of homogeneous distribution of silver nanoparticles and no agglomeration was observed. Atomic Absorption Spectroscopy was used to determine the concentration of AgNPs changes with synthesis process. The results revealed the best condition for synthesis of AgNPs using spent tea leaves extract was taken at 5 mM AgNO₃ concentration (molarity), 10% extract volume percent, 10 min under sunlight exposure and room temperature (20 °C).



(a)



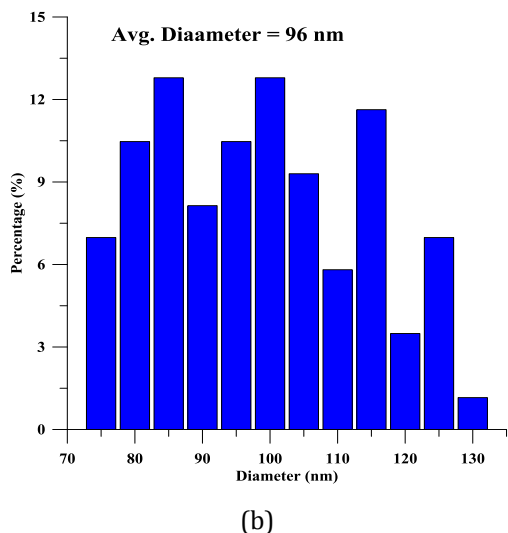


Figure 10 AFM images with nanoparticles size distribution of AgNPs: a) 10min sunlight exposure ; b) 15 min sunlight exposure

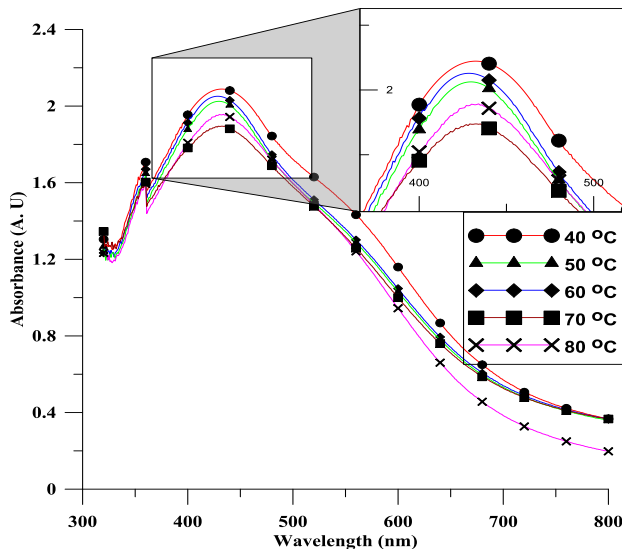


Figure 11 UV-Visible absorbance spectra of AgNPs at different temperature using tea leaves extract

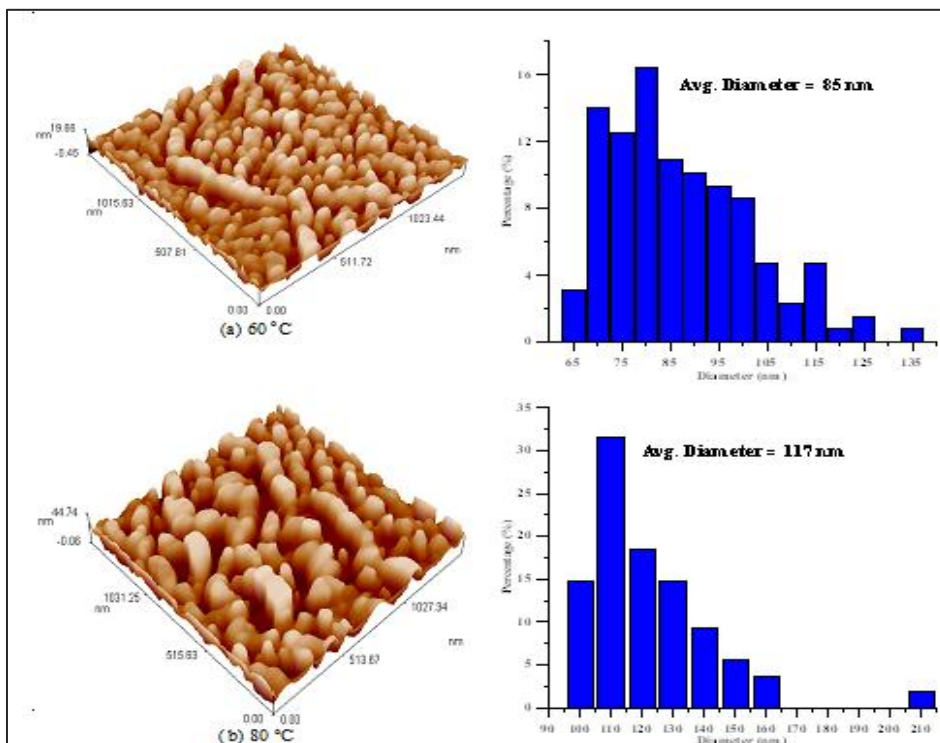


Figure 12 AFM image of AgNPs at different temperature using spent tea leaves extract: a) 60 °C, b) 80 °C

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