OPEN ACCESS

Manuscript ID:
ASH-2020-08012450

Volume: 8

Issue: 1

Month: July

Year: 2020

P-ISSN: 2321-788X

E-ISSN: 2582-0397

Received: 06.04.2020

Accepted: 20.06.2020

Published: 02.07.2020

Citation:
Goyal, Swati. "A Study on Synthesis of Silver Nanoparticles Using Ocimum Sanctum L (Tulsi) Leaf Extract & Their Antimicrobial Activity: A 'Green Chemistry Approach.'" *Shanlax International Journal of Arts, Science and Humanities*, vol. 8, no. 1, 2020, pp. 99–103.

DOI:
https://doi.org/10.34293/sijash.v8i1.2450

A Study on Synthesis of Silver Nanoparticles Using Ocimum Sanctum L (Tulsi) Leaf Extract & their Antimicrobial Activity: A Green Chemistry Approach

Swati Goyal
Department of Chemistry
Dr A.P.J Abdul Kalam University, Indore, Madhya Pradesh, India

Abstract
Nanoparticles (NPs) are being widely used in different fields; therefore, there is growing interest in the development of a biological and environmental safety method for their production. Now a day's chemical and physical methods are being used for the development of nanoparticle that is costly, time-consuming, and harmful for the environment. Plant-mediated synthesis of nanoparticles is a "Green chemistry" approach that connects different types of plants with nanotechnology. It has gained much more attention as a reliable, sustainable, and eco-friendly method for synthesizing a wide range of materials / nanomaterials. Plants are called nature's "chemical factories" therefore, plants and plant extracts are the best options to produce different types of nanoparticles. In this present study, silver nanoparticles were synthesized by using leaf extract of Ocimum sanctum and aqueous silver nitrate solution through a simple and eco-friendly method. Then the developed silver nanoparticles were characterized by using different types of techniques such as Scanning electron microscopy (SEM), Transmission electron microscopy (TEM), UV-Vis spectrophotometer, and Fourier transforms infra-red (FTIR) spectroscopy. Their antimicrobial activity was screened against microbial culture, and it was found that the synthesized silver nanoparticles have potential applications in antibacterial activity.

Keywords: Green synthesis, Ocimum Sanctum L, Silver Nanoparticles, Characterization and Antimicrobial activity.

Introduction

A convergence between biological-based technologies, green chemistry, and nanotechnology has become a major source of attraction from the past few years. Nanotechnology includes multidisciplinary research interests like Biochemistry, Material Science, Polymer Science, Botany, Biotechnology, etc and it has made nanotechnology crucial in many diverse fields like industry, agriculture, forestry medicine, and environmental management. Nanotechnology is the technique for synthesizing particles with the nano range from about 1 to 100 nm. They have a large surface area to volume ratio because of this. They possess optical properties. The nanoscale materials have accepted major significance for environmental remediation due to its abundant, ecofriendly, low cost, and non-toxic. These nano range particles are the primary building blocks of nanotechnology. The physicists, chemists, materials scientists, and engineers have a gain attraction towards the synthesis of metal and semiconductor nanoparticles due to their potential applications in different fields like catalysis, biosensing, recording media, and electronics. The most effective nanoparticles are being made by noble metals viz., Ag, Pt, Au, and Pd. Among those four noble metals, silver nanoparticles (AgNPs) have gained much importance because they play a significant role in the field of biological

systems, living organisms, and medicine. AgNPs are the most commercializing nanoparticles having some exclusive physicochemical properties like electric, optical, catalytic, and mainly medicinal properties.

Antibacterial activities have gained much attention because they potentially offer a solution to the problem of antibiotic resistance. AgNPs get attached to the cell wall and disturb cell wall permeability and cellular respiration; therefore, they can be used as an effective anti-microbial agent. Due to this property, AgNPs are now widely used in different fields of medicine like molecular imaging, diagnosis, and in the treatment of cardiovascular diseases and drug delivery. They can also be used in textile fabrics, as food additives, in the detection of DNA, and in packaging to eliminate microorganisms, etc. Various types of methods can be used to synthesize AgNPs depending on their specific requirements like their particular size and shape. The chemical and physical technologies that are being used for are quite expensive, and their by-products and wastes that are generated are toxic and harmful for the environment. As a result, there is a need to discover a technique that can be overcome this serious problem. Greener technology for the synthesis of silver nanoparticles is an environment-friendly, readily available, non-pathogenic, and less expensive technique, and it provides a single-step method for this process. In the present study, we have used Ocimum Tenuliflorum L. (Tulsi) for the production of AgNPs. Ocimum Tenuliflorum L. is a high-grade medicinal plant which has great medicinal properties. UV-Visible spectroscopy, Fourier transforms infrared (FTIR), Transmission Electron Microscope (TEM), and Scanning Electron Microscope (SEM) techniques have been used to identify the potential biomolecules. Furthers, its efficacy to inhibit different pathogenic bacterial growth was evaluated against human pathogens by the disc diffusion method.

Materials and Methods

Preparation of Silver Nitrate ($AgNO_3$) Solution

1 mM of the $AgNO_3$ solution was used for the preparation of AgNPs. For making AgNO3 solution 0.084, gm $AgNO_3$ is dissolved in double-distilled water in 500 mL volumetric flask. Then it was stored in Amber colored bottle to keep away from auto-oxidation of silver ions.

Preparation of extract of Tulsi (Ocimum sanctum) Leaves

Fresh leaves of Tulsi (Ocimum sanctum) were collected from the university campus and washed several times under running water and then soaked in deionized water for some time. After that, the water was removed, and about 50g of the leaves were ground into fine pieces. Then these ground leaves were kept in a 100 ml beaker having 20 ml methanol and 80 ml distilled water, and the beaker was boiled for at 90^{o}C 50 minutes. After that, the beaker was removed out and allowed to cool at room temperature. The solution was then filtered and preserved at 4^{o}C.

Green Synthesis: Silver Nanoparticles

AgNPs were synthesized using the made solutions silver nitrate, and Tulsi leaves extract. For this, 25 ml of Tulsi leaf extract was taken in a 250 ml conical flask containing 125 ml of freshly prepared 1mM $AgNO_3$ solution. Now the solution was heated up to 90^{o}C for 10 minutes, and changes were observed. It was found that the color gets changed to indicate the primary stage of Silver nanoparticles. The changed color solution was then centrifuged at 10000 rpm for 20 minutes, and the supernatant was collected in a clean beaker for further settlement of particles. This process ass repeated so many times to get a fine solution. After this microfiction technique was applied to get dried Silver nanoparticles and the flask was incubated at room temperature for 24 hrs in a dark chamber. The particles obtained by this technique were further characterized using different types of techniques.

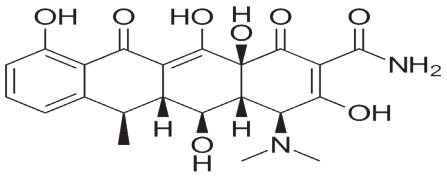
Characterization of AgNPs

Different types of spectroscopic and microscopic techniques were used to find out the morphology, characteristics, and functional group of the synthesized nanoparticles. Two microscopic techniques were performed for this purpose. Scanning Electron Microscopy (SEM) technique was used to find out the morphology of the nanoparticles, and Transmission electron microscopy (TEM)

was performed to determine the size and shape of nanoparticles. Another two spectroscopic techniques Fourier transformation infrared (FTIR) spectroscopy and UV-Vis spectroscopic were also performed. FTIR spectrometer was used to find out the bounding or interaction of element to element, and the desorption measurements were recorded on a Systronics spectrophotometer 166 (India) over the wavelength range 325–990 nm.

Antibacterial Assay

The antimicrobial activity of developed nanoparticles was tested against two microorganisms – (Staphylococcus aureus and Escherichia coli) and the antibiotic Doxycycline (Scheme 1). Agar medium was used as a nutrient for the cultivation of the microorganisms, and the fresh grown culture of the microorganisms (50 µL) was spread on Petri plates by punching the holes in the media. A definite amount of AgNPs was injected into this hole using sterile equipment. Then all the plates were kept at room temperature for diffusion for 15 min. After that, the plates were kept in an incubator for 24 h at 300°C.



Scheme 1: Structure of Doxycycline

Results and Discussion

On the addition of leaf extract into solution of silver nitrate, the color of the solution changes from light yellow to brown. The brown color Silver nanoparticles occur due to the excitation of surface plasmon resonance. We observed the reduction rate and formation of nanoparticles according to change in temperature and concentration of silver nitrate as well as leaf extract in the solution.

Visual Properties Analysis

On adding the leaf extract of Ocimum Sanctum to an aqueous solution of AgNo3, surface plasmon resonance occurs due to which the color of the solution gets changed from yellow to brown. Due to surface plasmon resonance, silver ions get reduced, and it was observed by UV-Vis spectroscopy. UV-Vis spectroscopy can also be used to determine the size and shape of the nanoparticles. By the UV-Vis spectra, it was observed that the intensity of absorbance peak increased with time; therefore highest peak was observed at 427 nm (Fig.1).

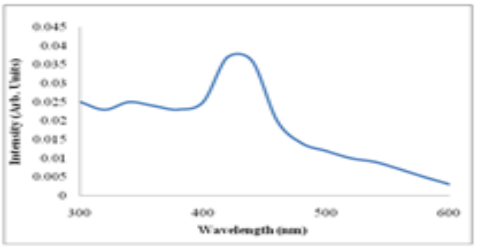


Figure 1: UV-Vis spectra of AgNPs

Particle Morphology Analysis

To determine the morphology of the silver nanoparticles, the sample was measured with a scanning electron microscope (SEM) and Transmission Electron Microscopy (TEM) Analysis. TEM study has been performed to find out the morphology and particle size silver nanoparticles. This technique helps us to understand the crystal structure, shape, and size of the silver particles. The TEM image of AgNPS has been showing in Fig.3. By the morphological study, it was found that nanoparticles are spherical, and the vacuums are well-dispersed with 20-50 nm size range.

SEM (Fig.2) study has also been done to get the knowledge about the dimensions of synthesized AgNPs. The sample was sonicated for 30 minutes at room temperature, and a small drop was dried on a glass slide. Then the slide was coated by gold particles and examined by scanning electron microscope. SEM analysis confirmed the spherical shape of the synthesized silver nanoparticles.

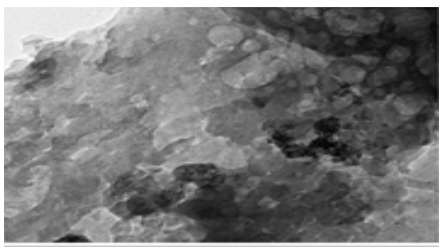


Figure 2: TEM image of AgNPs

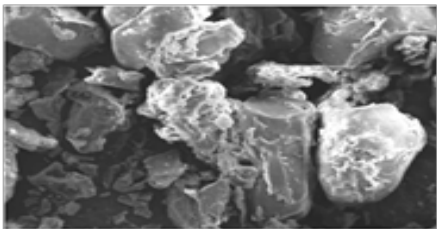


Figure 3: SEM image of AgNPs

Fourier Transform Infrared (FTIR) Analysis

Fourier Transform Infrared (FTIR) analysis has been done to find out the present functional groups in the synthesized nanoparticles. The spectra of AgNPs show sharp absorption peaks at two different wavelengths 1642 cm-1 and 3441 cm-1. The peak at 1642 cm-1 is arisen due to the stretching of carbonyl in proteins, which indicates the presence of amide I bond of proteins. Another peak that occurs at 3441 cm-1 is found due to OH stretching in alcohols and phenolic compounds. FTIR studies of synthesized nanoparticles confirmed the carbonyl group and proteins present in it have the stronger ability to get bind metal with metal ions.

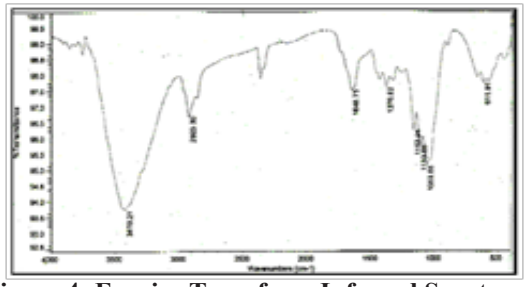


Figure 4: Fourier Transform Infrared Spectra of AgNPs

Antibacterial Activity Analysis

Antibacterial activity of synthesized AgNPs was observed against two bacteria viz.; Staphylococcus aureus and Escherichia coli bacteria. For this purpose, a disc diffusion method was used. This method was used to identify two important things, namely the sensitivity of bacterial strains and zone of inhibition towards the antibiotics. All the data of the present study are given in Table 1, which showed that the developed AgNPs exposed good inhibition against both of the bacterial strains (Staphylococcus aureus and Escherichia coli).

Table 1: Antibacterial activity of synthesized AgNPs using Ocimum Sanctum leaf extract

Name of Micro-organism	Zone of Inhibition in mm		
	Doxycycline (10 µg/disc)	Concentration of Silver Nanoparticles (µL)	Silver Nano-particles
Staphylococcus Aureus	20	15	1.8
	20	30	2.2
	20	45	2.5
Escherichia Coli	20	15	1.5
	20	30	1.9
	20	45	2.0

Conclusions

The plant-mediated nanoparticles can be produced at large scale, and they can have compatibility for medical and pharmaceutical applications. From the study, it can be concluded that the plant-mediated synthesis of nanoparticles is a simple, nontoxic, efficient, and ecofriendly method. In the present investigation leaves extract of Ocimum sanctum and a known concentration of silver nitrate solution was used to synthesize the AgNPs. Different types of Microscopic and Spectroscopic techniques were used to characterize the developed nanoparticles. The antimicrobial activity of the developed AgNPs was studied against Staphylococcus aureus and Escherichia coli bacteria, and it was found that the synthesized nanoparticles show good stability and good antimicrobial activity against both of the bacterial strain.

References

Borase, Hemant P, et al. "Plant Extract: A Promising Biomatrix for Ecofriendly, Controlled Synthesis of Silver Nanoparticles." *Applied biochemistry and biotechnology,* vol. 173, no. 1, 2014, pp. 1-29.

Cho, KH, et al. "The Study of Antimicrobial Activity and Preservative Effects of Nanosilver Ingredient." *Electrochimica Acta,* vol. 51, no. 5, 2005, pp. 956-960.

Elumalai, E.K. et al. "Green Synthesis of Silver Nanoparticle using Euphorbia Hirta L. and their Antifungal Activities." *Journal of Applied Science Research,* vol. 2, 2010.

Faramarzi, M.A. and Armin Sadighi. "Insights into Biogenic and Chemical Production of Inorganic Nanomaterials and Nanostructures." *Advances in Colloid and Interface Science,* vol. 189-190, 2013, pp. 1-20.

Gurunathan, S., et al. "Biosynthesis, Purification and Characterization of Silver Nanoparticles using Escherichia Coli." *Colloids and Surfaces B: Biointerfaces*, vol. 74, no. 1, 2009, pp. 328-335.

Hulkoti, N.I. and Taranath, T.C. "Biosynthesis of Nanoparticles Using Microbes - A Review." *Colloids and surfaces. B, Biointerfaces,* vol. 121, 2014, pp. 474-483.

Jain, D., et al. "Synthesis of Plant-Mediated Silver Nanoparticles Using Papaya Fruit Extract and Evaluation of Their Anti-Microbial Activities." *Digest Journal of Nanomaterials and Biostructures*, vol. 4, no. 3, 2009.

Kolmas, J., et al. "Substituted Hydroxyapatites with Antibacterial Properties." *Biomed Research International,* vol. 2014.

Levy, Ilan, and Oded Shoseyov. "Cross Bridging Proteins in Nature and their Utilization in Bio- and Nanotechnology." *Current Protein & Peptide Science,* vol. 5, no. 1, 2004.

Mallick, K., et al. "Self Assembly of Silver Nanoparticles in a Polymer Solvent: Formation of a Nano Chain through Nanoscale Soldering," *Materials Chemistry and Physics*, vol. 90, no. 2-3, 2005, pp. 221-224.

Panigrahi, S, et al. "General Method of Synthesis for Metal Nanoparticles." *Journal of Nanoparticle Research*, vol. 6, 2004.

Parashar, Vyom, et al. "Parthenium Leaf Extract Mediated Synthesis of Silver Nanoparticles: A Novel Approach Towards Weed Utilization." *Digest Journal of Nanomaterials and Biostructures*, vol. 4, no. 1, 2009, pp. 45-50.

Poulose, Subin, et al. "Biosynthesis of Silver Nanoparticles." *Journal of Nanoscience and Nanotechnology,* vol. 14, no. 2, 2014.

Sathyavathi, R. et al. "Biosynthesis of Silver Nanoparticles using Coriandrum Sativum Leaf Extract and their Application in Nonlinear Optics." *Advanced Science Letters*, vol. 3, no. 2, 2010, pp. 138-143.

Sato, K. and Kitamori, T. "Integration of an Immunoassay System into a Microchip for High-Throughput Assay." *Journal of Nanoscience and Nanotechnology,* vol. 4, no. 6, 2004, pp. 575-579.

Schrand, A.M., et al. "Toxicity Testing of Nanomaterials." *Advances in Experimental Medicine and Biology,* Springer, 2012.

Singh, M, et al. "Nanotechnology in Medicine and Antibacterial Effect of Silver Nanoparticles." *Digest Journal of Nanomaterials and Biostructures*, vol. 3, no. 3, 2008.

Thompson, D.G. et al. "Ultrasensitive DNA Detection using Oligonucleotide - Silver Nanoparticle Conjugates." *Analytical chemistry*, vol. 80, no. 8, 2008, pp. 2805-10.

Wang, H. et al. "Responsive Polymer-Flurorescent Carbon Nanoparticle Hybrid Nanogels for Optical Temperature Sensing, Near-Infrared Light-Responsive Drug Release and Tumor Cell Imaging." *Nanoscale,* vol. 6, no. 13, 2014, pp. 7443-7452.

Xia, Yu, et al. "A Nontoxic and Low-Cost Hydrothermal Route for Synthesis of Hierarchical Cu_2ZnSnS_4 Particles." *Nanoscale Research Letters,* vol. 9, 2014 pp. 208-214.

Yang, Hung-Wei, et al. "Potential of Magnetic Nanoparticles for Targeted Drug Delivery." *Nanotechnoly Science and Applications,* vol. 5, 2012, pp. 73-86.

Yu, Da-Guang. "Formation of Colloidal Silver Nanoparticles Stabilized by Na^+ Poly (-glutamic acid)-Silver Nitrate Complex Via Chemical Reduction Process." *Colloids and surfaces. B, Biointerfaces,* vol. 59, no. 2, 2007, pp. 171-178.

Author Details

Swati Goyal, *Department of Chemistry, Dr A.P.J Abdul Kalam University, Indore Madhya Pradesh, India.*
***Email ID**: swatipc2011@gmail.com.*