Vol. 20(5), pp. 210-215, May, 2021
DOI: 10.5897/AJB2020.17292
Article Number: 6EAF6E166849
ISSN: 1684-5315

http://www.academicjournals.org/AJB

African Journal of Plant Science

Full Length Research Paper

Synthesis, characterization and antibacterial activity of AgNPS mediated by *Ocimum sanctum*

A. V. Narasimha Swamy[1]* and R. Srinu Venkat Rao[2]

[1]Chemical Engineering Department, Faculty of Engineering, Jawaharlal Nehru Technological University (JNTU), Anantapur, India.
[2]Chemical Engineering Department, Faculty of Engineering, Bule Hora University, Ethiopia.

Received 5 December, 2020; Accepted 29 January, 2021

This research focused on isolation of Silver Nano Particles (AgNPs) using leaf filtrate of *Ocimum sanctum* and evaluation of their antibacterial action against *Bacillus aureus, Staphylococcus aureus and Escherichia coli*. It is known that plant extracts act as reducing agents, stabilizers and as capping agents. The *O. sanctum* plant was specifically selected due to its medicinal properties and abundant availability. The change in color of leaf extract was imminent when the silver nitrate was added to the basil leaf filtrates, indicating the formation of AgNPs. The UV-visible spectroscopy and SEM analysis confirmed the formation of AgNPs. Silver ions and nanoparticles are severely toxic to microorganisms; hence they are used predominantly as antimicrobial agents. The silver nanoparticles that were synthesized from *O. sanctum* leaf extract have exhibited antibacterial activity against *Klebsiella*, *B. aureus*, *S. aureus* and *E. coli* and the activity varied significantly with the different concentrations of silver nitrate used. The nano particles thus developed have been confirmed by UV along with SEM reports. AgNPs sizes varied from 53.3 to 121 nm. It has been observed that most of the plant extracts yielded silver nano particles of size less than 100 nm. However, the present experimental work yielded particle size of about 121 nm from *O. sanctum* leaf extract.

Key words: Silver nano particles (AgNPs), *Ocimum sanctum*, antibacterial activity.

INTRODUCTION

Nano technology is an interesting area of recent investigations with design, formation and structuring of particle in the range of 1-100 nm. Nano particle may be described as a minute matter that configures as a complete segment in relation to its momentum and character. AgNPs were used by professionals earlier in the fourth century in the well-known Roman Lycurgus cup made of dichroic glass, and also in the ninth century in Mesopotamia for developing sparking effect on pot surfaces. The luster comes along with thin layer that encompasses silver and copper NPs cover consistently in the glassy matrix of the ceramic glaze. These NPs were made by skilled people by pouring salts of Cu and Ag and oxides along with vinegar, ochre, and clay on the exterior of pottery that were sparkled previously.

The substance temperature was increased to 600°C in

*Corresponding author. E-mail: swamyn297@gmail.com.

a reducing environment in a kiln. The glaze softens during hot condition, resulting in migration of Cu and Ag ions to outer layers of the glaze surface. The ions migrate back to metals in reduced environment, which congruently form as the nanoparticles resulting in color and light effects. Sheen technique manifest that the previous skilled people had sufficient information of substances. Taniguchi (1974), Kodali and Klabunde (2012), Maynard et al., 2010 of Tokyo University of Science introduced the term “nano-technology” during a conference in 1974. According to his definition, “Nanotechnology comprises, partition, coalition, and deformation process of substances by one atom or one molecule.” It would be appropriate to mention here that Drexler’s (1991) Ph.D. work at the MIT was the first doctoral degree in the area of molecular nanotechnology. The size of nanoparticles would be about 10^{-9}. AgNPs turn out to be significant in research domain because of its innumerable applications. Anti-bacterial activities of silver and silver compounds have been applied in dentil, catheters and burns to mention a few. AgNPs may be made by reducing $AgNO_3$ to Ag ions in the presence of plant isolate. AgNPs production by plant extract is a promising field since plant portion works as reducing and capping agent which is devoid of toxic compounds. The benefits of incorporating plant extract for AgNPs production includes power conservation, profitability and usefulness in safeguarding health. The applications of silver nano particles are many and a few are represented here. Gajendra et al., (2020) studied generation of AgNPS using silver nitrate and stem of *Andrographis paniculata*. The Ag-NPs showed antibacterial activity against *Streptococcus pneumonia*. Bilal et al. (2019) worked on AgNPs using callus extract of basil, and observed their anti- proliferative potential against liver carcinoma cells. Dakshayania et al. (2019) performed studies on synthesis and characterization of AgNPs with *Selaginella bryopteris* plant extract and Ag-NPs are expected to treat thrombotic disorders. Shahnaz and Mussarat (2016) conducted studies on AgNPS using *Salix alba* bark extract and antibacterial activity of these AgNPs was evaluated by disc diffusion, against bacteria isolated from dental plaque. Junwen et al. (2019) generated AgNPs along with an aqueous extract of *Coptis chinensis*, using ecofriendly green-synthesis and found that CC-AgNPs regulated apoptos is using the intrinsic pathway to inhibit A549-cell multiplication. Azam et al. (2017) studied generation of AgNPs from aqueous solution of $AgNO_3$ by using *Nigella arvensis* L. seed extract and generated NPs were also found to exhibit good cytotoxic effect on human breast cancer cells. Le Ouay and Stellacci (2015) worked on the antibacterial activity of AgNPS. Ibrahim (2015) studied about an ecofriendly, profitable, quick and simple method for synthesis of AgNPS using banana peel extract. Vijaya et al., (2013) studied the antigenotoxic effect of generated AgNPS of *Ocimum sanctum* leaf extract against cyclophosphamide. Diptendu and Goutam (2017) studied the generation of AgNPS by using mint leaves extracts and also evaluated their antimicrobial activity. The aim of the studies is to evaluate the antibacterial activity of the AgNPS against *Klebsiella pneumonia, Staphylococcus aureus, Enterobacter aerogenes, Escherichia coli and Pseudomonas aeruginosa*.

MATERIALS AND METHODS

Production of leaf extract

O. sanctum (Basil) leaves were taken and cleaned once with tap water, twice with DW, for removing dirt and dust. 10 g of cleaned leaves were made into small parts and boiled in 100 ml of DW with continuous mixing for 5 min. 10 ml of the filtrate was collected and stored for further use.

Synthesis of AgNPs

AgNPs were prepared by treating 90 ml of 2.0 mM $AgNO_3$ with 10 ml of filtrate. The reaction mixture was maintained at room temperature for 10 min, giving rise to the formation of dark brown color indicating the formation of silver nano particles. This blend was incubated at room temperature in dark conditions. About 1 ml of silver nanoparticle solution was taken (dilution with 1:2 V/V double distilled water) and observed in a UV-Vis Spectrophotometer (BIO SPECTROPHOTOMETER (BL 198) at different time intervals (20, 40, 60 and 80 min). The effect of $AgNO_3$ was monitored by varying its concentration as: 1, 2, 3, and 4 mM, respectively).

Characterization of silver nanoparticles

Silver nanoparticles exhibit unique optical properties. Preparation of AgNPs was ensured by studying the UV-Vis spectra. UV-Vis spectroscopy analysis was carried out on ELICO BIO SPECTROPHOTOMETER (BL 198) at 500 - 600 nm. The double distilled water was used as blank observation and the characteristics of AgNPS were determined by scanning electron microscopy.

Protocol of scanning electron microscope

The exterior pattern of the specimen was tested by employing scanning electron microscope (JEOL Ltd., Japan). A little quantity of powder was disseminated by hand onto a carbon tab attached to an aluminum stub and were smeared with a fine layer (300A) of gold by using POLARON-E 3000 sputter coater. The specimens were inspected by scanning microscope while direct data seized the picture.

Antimicrobial activity of formed AgNPS

The antibacterial work of AgNPs obtained at various $AgNO_3$ concentrations and plant extract was evaluated against *K. pneumonia, S. aureus, E. aerogenes, and P. aeruginosa.* Single cultures of bacteria were sub-cultured in Nutrient Agar. Wells of 5-mm diameter were made using gel puncture. Using a micro pipette, nanoparticle solution was poured into every well of all plates. The diameter of zone inhibition was measured in centimeter subsequent to incubation at 37°C for 24 h.

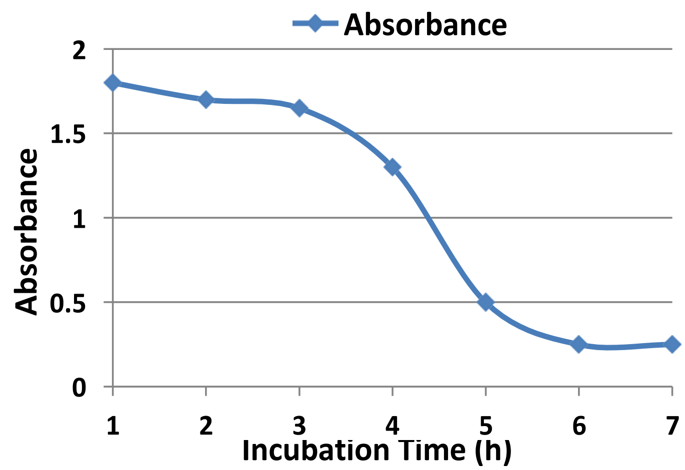

Figure 1. Incubation time.

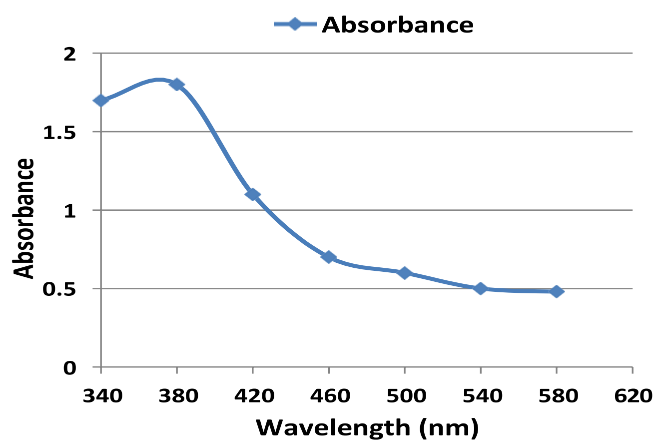

Figure 2. Spectra of leaf extract in 340 to 620 nm.

RESULTS

UV-Visible spectra analysis

The formed AgNPs were confirmed during visual observation by UV-Vis spectrometer. Absorption spectra of AgNPS formed in the solution at various nanometers. The *O. sanctum* isolates work as capping and stabilizing agents with *O. sanctum* having medicinal importance and is widely used in Ayurvedic medicine.

DISCUSSION

Reddish brown color indicates formation of AgNPs which exhibit reddish brown color in aqueous solution because of excitation of surface plasmon-vibrations. The dark brown color is formed in plant extract after addition of silver nitrate indicating the formation of silver nanoparticles as the reaction completes after 24 h (Shahnaz and Mussarat, 2016).

Absorption spectra of AgNPS formed in the solution at 380 nm showed the particle has increasingly sharp peak between 2nd and 3rd h and gradually decreasing with time (Figure 1). The synthesized silver nano particle using *O. sanctum* plant extracts were detected by UV-Vis spectrophotometer at various wavelengths (340, 380, 420, 460, 500, 540, 580 and 620 nm) with increasingly sharp absorbance maximum at 380 nm and gradually decreasing (Figure 2). Graph of time (x-axis) and optical density at 500 nm (y-axis) for various concentrations of $AgNO_3$ are thus plotted (Figure 3).

Antimicrobial analysis

The antibacterial activity of various plant extracts and the silver nanoparticle attained at various $AgNO_3$

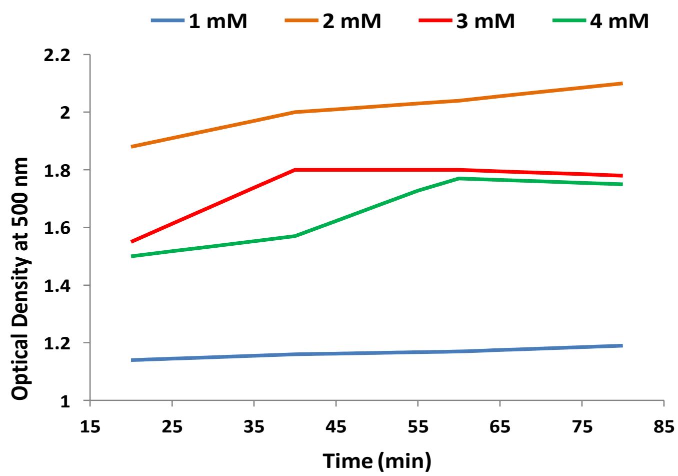

Figure 3. UV-VIS absorption spectroscopy of AgNPS.

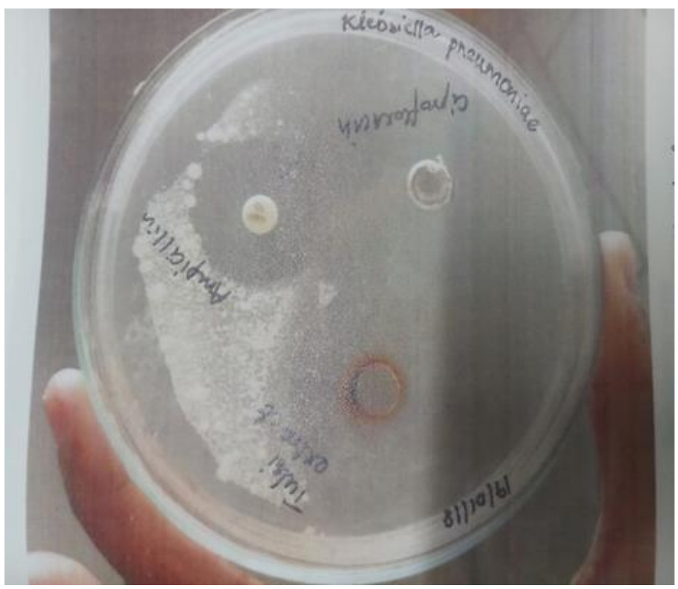

Figure 4. Zone of inhibition values for *Klebsiella pneumonia.*

concentration was evaluated against *K. pneumoniae*. After incubation at 37°C for 24 h, the zone of Inhibition diameter was measured.

The zone inhibitions are shown in Figures 4 to 8 and the corresponding values for different organisms with pertaining antibiotics are shown in Table 1.

SEM analysis

The scanning electron microscopic (SEM) image shows that high density AgNPS produced by *O. sanctum* plant extracts ultimately confirmed the presence of AgNPS (Figure 9). The picture shows consistently spherical silver nano particles having diameter of 53.3 to 121 nm. The nanoparticles stayed apart in the aggregates, showing the stabilization of the NP by a capping agent.

Conclusion

AgNPS prepared from leaf extract showed antibacterial activity against various pathogenic microorganisms and this activity varied with different concentrations of $AgNO_3$ used. The average particle size using *O. sanctum* is in the range of 53.3 to 121 cm. The inhibition zones found in these experiments by the authors are reported here. There is a good scope for these AgNPS in medicinal

Table 1. Zone of inhibition values for different organisms with antibiotics.

S/N	Organism	Disease caused	Antibiotic	Inhibition value CM
1	*Klebsiella pneumonia*	Pneumonia	Ciprofloxacin Ampicillin	50 2.6
2	*Staphylococcus aureus*	Endocarditis	Oxacillin Fluconazole	0.70 0.10
3	*Enterobacter aerogenes*	Infectious and opportunistic bacteria	Erythromycin Clindamycin	0.10 1.90
4.	*Escherichia coli*	Gastero-enterititis	Amoxicillin Ciprofloxacin	1.75 3.50
5	*Pseudomonas aeruginosa*	Skin infections etc.	Gentamycin Ciprofloxacin	3.55 2.40

Figure 5. Zone of inhibition values for *Staphylococcus aureus.*

Figure 7. Zone of inhibition for *Escherichia coli.*

Figure 6. Zone of inhibition values for *Enterobacter aerogenes.*

Figure 8. Zone inhibition of values for *Pseudomonas aerureginosa.*

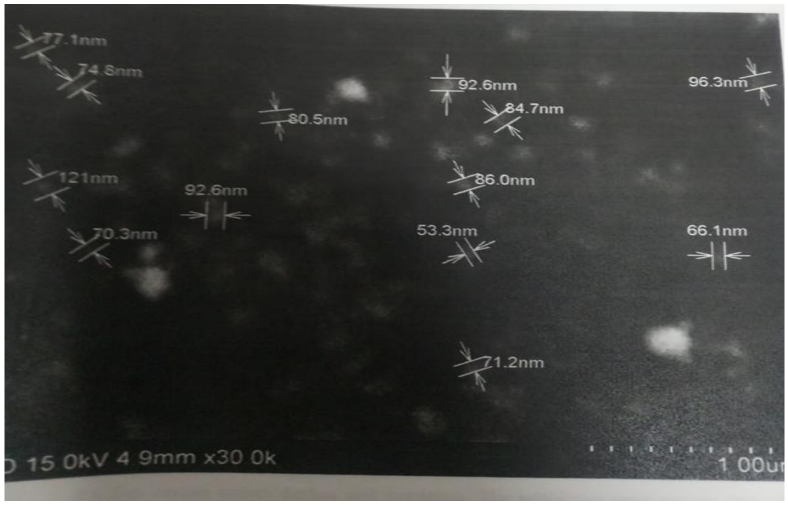

Figure 9. Scanning electron microscope analysis of silver nano particles.

applications also. The antibacterial activities of AgNPS on some pathogenic micro-organisms studied, thus reveals that the green synthesis method for the isolation of nano-particles provide rapid, stable, economic, simple and environmentally friendly mechanism.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

The authors gratefully appreciate the JNTUA, Anantapuramu, India for providing basic facilities for carrying out this research.

REFERENCES

Azam C, Naser K, Ali F (2017). Biosynthesis, Characterization, Antimicrobial and Cytotoxic Effects of Silver Nanoparticles Using *Nigella arvensis* L. seed extract. Iranian Journal of Pharmaceutical Research 16(3):1167-1175.

Bilal HA, Munazza N, Wali M, Syed SH, Rashda A, Lubna R, Christophe H (2019). A Comparative Evaluation of the Antiproliferative Activity against HepG2 Liver Carcinoma Cells of Plant-Derived Silver Nanoparticles from Basil Extracts with Contrasting Anthocyanin Contents. Biomolecules 9(8):320.

Dakshayani SS, Marulasiddeshwara MB, Sharath Kumar MN, Ramesh G, Raghavendra KP, Devaraja S, Rashmi H (2019). Antimicrobial, anticoagulant and antiplatelet activities of green synthesized silver nanoparticles using Selaginella (Sanjeevini) plant extract. International Journal of Biological Macromolecules 131(6):787-797.

Diptendu S, Goutam P (2017). Green synthesis of silver nano particles using *Mentha asiatica* (mint) Extract and Evaluation of their antimicrobial potential. International Journal of Current Research in Biosciences and Plant Biology 4(1):77-82.

Drexler (1991). Molecular Machinery and Manufacturing with Applications to Computation (PDF) (Ph.D. thesis). Massachusetts Institute of Technology.

Gajendra NM, Prasenjit M,Indranil C, Ganesh CS, Nirmal M, Kaushik G, Nandan B, Sudipta D, Soumitra M (2020). Green synthesis, characterization, antimicrobial and cytotoxic effect of silver nanoparticles using arabinoxylan isolated from Kalmegh. International Journal of Biological Macromolecules 162(11):1025-1034.

Ibrahim HMM (2015) Green Synthesis and Characterization of Silver Nanoparticles Using Banana Peel Extract and Their Antimicrobial Activity against Representative Microorganisms. Journal of Radiation Research and Applied Sciences 8:265-275.

Junwen P, Binfan F, Lifeng J, Taizhen S (2019). Biosynthesis, characterization, and anticancer effect of plant-mediated silver nanoparticles using Coptis chinensis. International Journal of Nanomedicine 14:1969-1978.

Le Ouay B, Stellacci F (2015). Antibacterial activity of silver nanoparticles: A surface science insight. Nano Today 10(3):339-354.

Maynard DA, Bowman DM, Hodge GA (2010). Tracing and disputing the story of nanotechnology. In Hodge, Graeme A, Bowman Diana M. Maynard Andrew D. International hand book on regulating nanotechnologies. Cheltenham, UK: Edward Elgar. p. 54. ISBN 9781849808125.

Shahnaz M,Mussarat K (2016). Green Synthesis of Silver Nanoparticles using Bark Extract of Salix Alba and Its Antimicrobial Effect Against Bacteria Isolated from Dental Plaque. Oriental Journal Chemistry 32(3):1611-1618.

Taniguchi N (1974). On the Basic Concept of 'Nano-Technology', Proceedings of the International Conference on Production Engineering, Part II, Japan Society of Precision Engineering.

Vijaya PP, Rekha B, Mathew AT, Syed MA, Yogananth N, Anuradha V, Kalitha PP (2014). Antigenotoxic effect of green-synthesised silver nanoparticles from sanctum leaf extract against cyclophosphamide induced genotoxicity in human lymphocytes-in vitro. Applied Nanoscience 4:415-420.