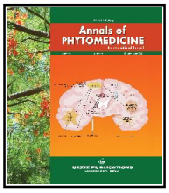
Annals of Phytomedicine 12(1): 453-459, 2023

DOI: http://dx.doi.org/10.54085/ap.2023.12.1.58

Annals of Phytomedicine: An International Journal
http://www.ukaazpublications.com/publications/index.php

Print ISSN : 2278-9839 Online ISSN : 2393-9885



Original Article : Open Access

Characterization and antibacterial response of biosynthesized silver nanoparticles using *Eclipta alba* (L.) Hassk. leaf extract

R. Anjali, Susan Kurian◆ and Anjana Rajan
Department of Zoology, Mar Ivanios College, Nalanchira, Thiruvananthapuram-695015, Kerala, India

Article Info

Article history
Received 5 April 2023
Revised 21 May 2023
Accepted 22 May 2023
Published Online 30 June-2023

Keywords
Silver nitrate
Nanoparticles,
Eclipta alba (L.) Hassk.
Antibacterial activity

Abstract

A variety of plant extracts have been used for the green synthesis of nanoparticles as they contain bioactive compounds with high metal-ion reducing potential. In this study, we investigated the green synthesis of silver nanoparticles (AgNPs) using leaf extracts of *Eclipta alba* (L.) Hassk. (Syn. *Eclipta postrata* (L.), as well as the antibacterial potential of the nanoparticles. Initial indications of AgNPs formation were a change in colour of the reaction mixture; namely, from green to brown, which was further confirmed by a characteristic SPR peak of 320 and 450 nm, respectively, measured by UV-vis spectroscopy. Scanning electron microscopy (SEM) showed the cuboidal morphology of AgNPs with the size range of 70 to 140 nm. The fourier transform infrared spectroscopy (FT-IR) spectra of AgNPs shows the presence of polyphenols and other plant derived compounds as well as proteins which reduces silver ion to AgNP's. Antimicrobial activity of green synthesized AgNPs against *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* were studied. Against *E. coli* and *P. aeruginosa,* AgNPs showed high antibacterial activity in comparison with *S. aureus*. In this study, it was demonstrated that eco-friendly green synthesized AgNPs may be more useful in biomedicine, water purification and nanobiotechnology in the near future.

1. Introduction

The nanotechnology field combines a wide range of scientific disciplines such as biology, chemistry and physics, where nanoparticles (NPs) possess more surface atoms than micropar-ticles, which make them more functional (Moteriya and Chanda, 2017). Materials, devices and systems can also be designed, characterized and applied through nanotechnology. This is done by controlling the size and shape of nanoparticles (Ndikau *et al.*, 2017). The advancement of green nanotechnology generates interest among researchers in the eco-friendly biosynthesis of nanoparticles. A wide range of nobel metals, including silver, gold, platinum, lead, copper, zinc and iron are used in the synthesis of nanosized particles. Due to their chemical-free nature and natural capping agents, plants make an ideal platform for nanoparticle synthesis (Haleemkhan *et al.,* 2015). In terms of silver nanotechnologies and their use as environmental disinfectants, the future is still unknown, since a lot of work and data is available and applications studied are going on**.** Silver nanoparticle research has attracted the attention of both the scientific community and the industry, regardless of the use of nanosilver in health-related fields. Studies on silver nanoparticles and their use in consumer products are emphasized in parallel to their syntheses and properties. For a low-cost, environmentally acceptable method, the synthesis of silver nanoparticles employing a variety of biological resources can be well suited. The rate at which metal ions are reduced rises when biological agents are used under standard temperature and pressure settings (Tran *et al.,* 2013).

Various chemical methods can be used to make nanomaterials. Nature has developed several methods for the synthesis of inorganic materials on the nano- and micro-length scales that focus on the creation of a relatively new and mostly untapped field of research based on the biosynthesis of nanomaterials (Mandal *et al*., 2023). The development of safe, non-toxic and ecologically acceptable "green chemistry" methods would be beneficial for the synthesis and assembly of nanoparticles. These methods are likely to involve organisms like bacteria, fungi, and even plants (Bhattacharya and Rajinder, 2005; Sastry *et al.,* 2004). The rapid expansion of research into nanomaterials and their applications has attracted particular interest in the realm of biomedical research. The use of hazardous chemicals and solvents compels scientists to advocate for more environmentally friendly, safe and biocompatible production methods, particularly for the future biological application of created nanomaterials. To address these issues, there is an increasing need for environmentally sound and hygienic ways to prepare nanoparticles using non-toxic chemicals and regenerative reducing agents (Moritz and Moritz, 2013).

NPs exhibit antimicrobial activity based on their composition, surface modification, intrinsic properties and microbial species. A cell membrane is bound to NPs by electrostatic interactions. NPs form pores on microbial surfaces and deposit on the surface (Hajipour *et al.,* 2012; Fröhlich *et al.,* 2012). Infiltrating into the cell wall causes structural changes to the plasma membrane, including increased permeability. By interacting with the phosphorus and sulfur elements of the DNA, AgNPs can disrupt DNA replication and terminate microorganisms (Prabhu *et al.,* 2012). Nanostructures are also capable of antimicrobial actions through the formation of biofilms. In recent

Corresponding author: Dr. Susan Kurian
Assistant Professor, Department of Zoology, Mar Ivanios College, Nalanchira, Thiruvananthapuram-695015, Kerala, India
E-mail: susan.kurian@mic.ac.in
Tel.: +91-8075558538

years, microbes have developed multiple properties that enable them to resist multiple drugs. Their ability to form biofilms is also an important characteristic.

Various herbal plants have antimicrobial activity and are used for herbal medicinal products. *E. alba* is a traditional medicinal plant which has been used in various ailments (Mugale and Balachandran, 2020; Vijayakumaran *et al*., 2020; Saminathan, 2017; Yadav *et al*., 2017; Jha *et al.,* 2008). The present study demonstrates the synthesis of AgNPs using *E. alba* and to evaluate its antibacterial efficacy.

2. Materials and Methods

2.1 Collection of plant material

The plant material used for the study was the leaves of *E. alba* which were collected from Nalanchira, Thiruvananthapuram District, Kerala, India. The voucher specimen has been deposited in the Herbarium of Department of Botany, University of Kerala, with Voucher No. KUBH-11280 for future reference.

Figure 1: *Eclipta alba* (L.) Hassk.

2.2 Preparation of *E. alba* leaf extract

Fresh leaves of *E. alba* were collected and washed thoroughly with running tap water followed by Milli Q water, incised into small pieces and air dried. About 10 g of the leaves of *E. alba* were weighed and transferred into a 500 ml beaker containing 100 ml Milli Q water, mixed well and boiled for 10-15 min at 60^0C. A 250 ml Erlenmeyer flask was used to collect the filtrate after passing the extract through Whatman No.1 filter paper.

2.3 Synthesis of silver nanoparticles using leaf extract

To synthesize silver nanoparticles, 95 ml of 1 mM $AgNO_3$ solution was taken in a sterile conical flask and 5 ml of aqueous plant extract was added to it. For the reduction of Ag+ ions, the conical flask was covered with foil paper and kept in a dark room until the color of the solution changed from pale yellow to dark yellow. After 30 min of incubation, the creation of silver nanoparticles was confirmed by UV-visible spectroscopy, which causes the solution to change from pale yellow to dark brown. As a result, silver ion bioreduction was observed. Within 24 h, silver nanoparticles were synthesized.

2.4 Separation of silver nanoparticles

Using a REMI Centrifuge at 5000 rpm for 20 min, the produced silver nanoparticles were separated and purified. The obtained suspension was centrifuges at 10000 rpm for 45 min. The supernatant liquid was re-suspended thrice with sterile double distilled water. To remove any uncoordinated biomolecules, the procedure was repeated three times. The powder was created by drying the cleaned suspension. The dried AgNPs samples were scraped and then stored in screw-capped vials for further characterization analysis.

2.5 Characterization of silver nanoparticles

The UV-vis spectrum was measured to track the reduction of Ag+ using a UV-160A double-beam spectrophotometer (Shimadzu, Kyoto, Japan). The spectra were measured over the wavelength range of 200 to 800 nm at a scan speed of 480 nm/min and the greatest absorption wavelength was identified. Using Thermoscientific Nicolet iS50, fourier transform infrared (FT-IR) spectroscopy studies were performed. To check for functional groups in the phytoconstituents of the bioextracts, the dried samples were combined with KBr in a mortar, crushed into discs and then examined. In the transmittance mode, FT-IR spectra were scanned between 4000 cm^{-1} and 400 cm^{-1} at a resolution of 4 cm^{-1}. The sample's surface topography, composition and other characteristics such as electrical conductivity are all revealed *via* SEM analysis. Thermo Scientifics' SEM machine was used to conduct a scanning electron microscopic (SEM) analysis. A very little amount of the sample was used to create thin films on a copper grid that had been coated with carbon. Any excess solution was blotted away and the grid was then placed under a mercury lamp for five minutes to dry the films.

2.6 Evaluation of antibacterial activity of silver nanoparticles

Disc Diffusion Method as described in European Pharmacopeia with slight modification was used for antibacterial testing of the produced silver nanoparticles (Thankamani *et al.*, 2011; Wayne, 2009). Using Mueller-Hinton broth, gram-negative and gram-positive organisms, including *Pseudomonas aeruginosa* (ATCC 27853), *Staphylococcus aureus* (MTCC 96) and *Escherichia coli* (ATCC 25922) (clinical isolate) maintained at Biovent Innovations Pvt. Ltd, Department of Biotechnology, University of Kerala were used for the test. Fresh cultures were prepared by growing the bacterial strains to the exponential phase in Mueller- Hinton Agar (MHA) at 37ºC for 2 h to obtain a log phase cultures, opacity was checked with 0.5 McFarland turbidity standards. 100 µl of the pure cultures of test strains were swabbed uniformly using a sterile swab on the surface of MHA plate to obtain an even inoculum. After the plates dried for 5 min, different concentrations (45, 90, 180 and 360 µg/ml) of *E. alba* mediated synthesized AgNPs dissolved in sterile distilled water were loaded in the disc to evaluate antibacterial activity. Streptomycin (0.125 mg/ml) and sterile distilled water were used as positive and negative controls. The antibacterial activity was assessed by measuring the diameter of the zone of inhibition.

3. Results

3.1 Synthesis of silver nanoparticles

As a result of the synthesis of silver nanoparticles, it was noticed that the colour of the silver nitrate solution changed from colorless to brown with increasing intensity throughout the incubation period, suggesting the creation of silver nanoparticles. Because of the metal

nanoparticles' surface plasmon vibration, silver nanoparticles are widely recognized to exhibit a yellowish brown colour in water.

3.2 UV-vis spectrophotometer study on plant synthesized silver nanoparticles

This is a crucial method to determine nanoparticle creation and stability. It can be used to evaluate the size and shape of nanoparticles in aqueous suspension. The UV-vis spectrum of *E. alba* mediated synthesized silver nanoparticle was recorded in the range of 200-800 nm. The peak of absorbance for silver nanoparticles of *E. alba* was observed at 300 nm and 450 nm which confirmed the presence of silver nanoparticles, as indicated in Figure 2.

3.3 FTIR spectrum study on synthesized silver nanoparticles

FT-IR spectrum of synthesized nanoparticles (Figure 3) indicates clear peaks in 410.99, 425.13, 443.79, 2163.44 and 2916.37 cm^{-1}. The peaks 2916.37 and 2163.44 are assigned to the C-H alkaline group and alkynyl C ≡ C stretches of silver nanoparticles, respectively (Raju *et al.,* 2018, Banerjee *et al.,* 2014). The peak at 443.79 cm^{-1} is due to the O-Si-O network and ring opening vibration (Anandalakshmi *et al.,* 2015). Therefore, it might be said that AgNPs possessed a powerful capacity to attach to several functional groups of the different polyphenolics and other plant-derived compounds, as well as proteins, indicating the development of a layer that covers AgNPs inhibiting agglomeration by acting as a capping agent and giving the NPs stability (Ashour *et al.,* 2015).

3.4 SEM analysis for synthesized silver nanoparticles

By using SEM examination, green-produced AgNPs were further evaluated for their size, shape, morphology and surface chemistry. Surface morphological and nanostructural studies of SEM micrographs (Figure 4a) showed AgNPs to be cuboidal, cluster and spherical in form of varied sizes ranging from 70-140 nm (Annamalai and Nallamuthu, 2015; Lakkim *et al.,* 2020). The larger silver particles may be due to the aggregation of smaller ones, due to SEM measurements or possibly an artifact of the centrifugation and further drying required to prepare silver nanoparticle sample for SEM analysis (Moodley *et al.,* 2018). The average particle size within the selected area of SEM image was 70.80 nm, conforming to the nano range as indicated in Figure 4 b (Raja *et al.,* 2017).

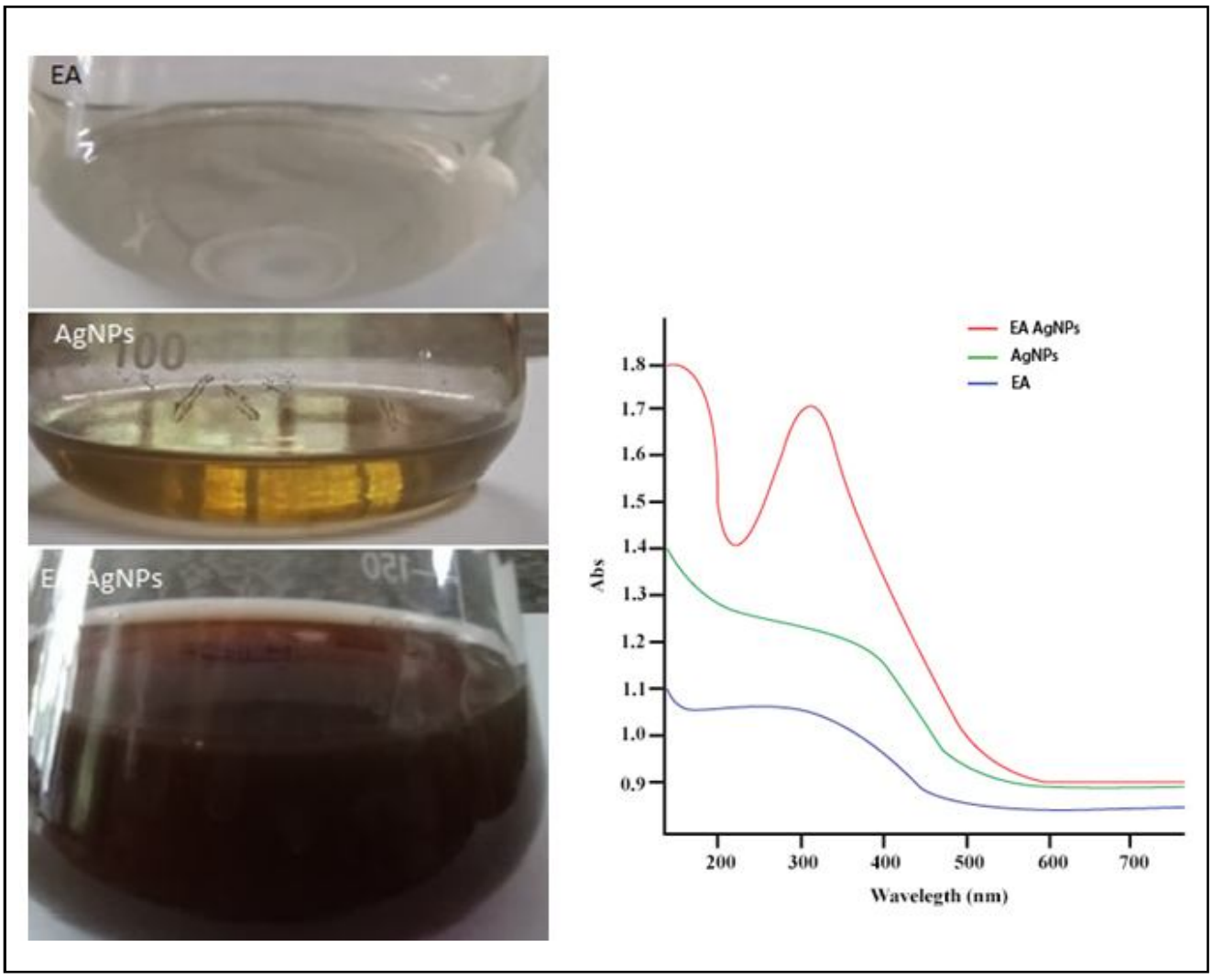


Figure 2: UV-vis spectroscopy analysis of biosynthesized silver nanoparticle of *E. alba* leaf extract.

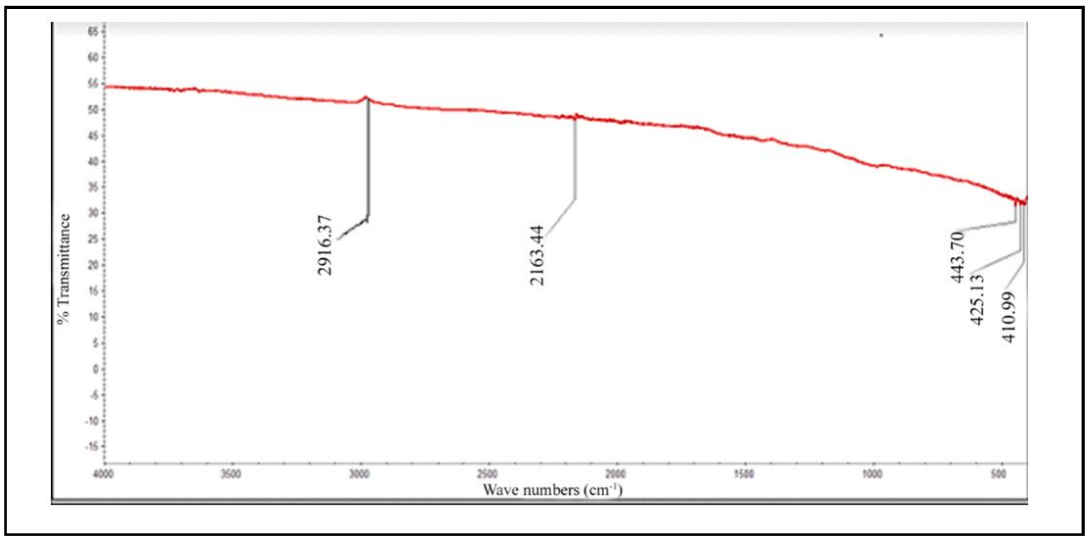


Figure 3: FT-IR spectra of bioreduced silver nanoparticles synthesized by *E. alba.*

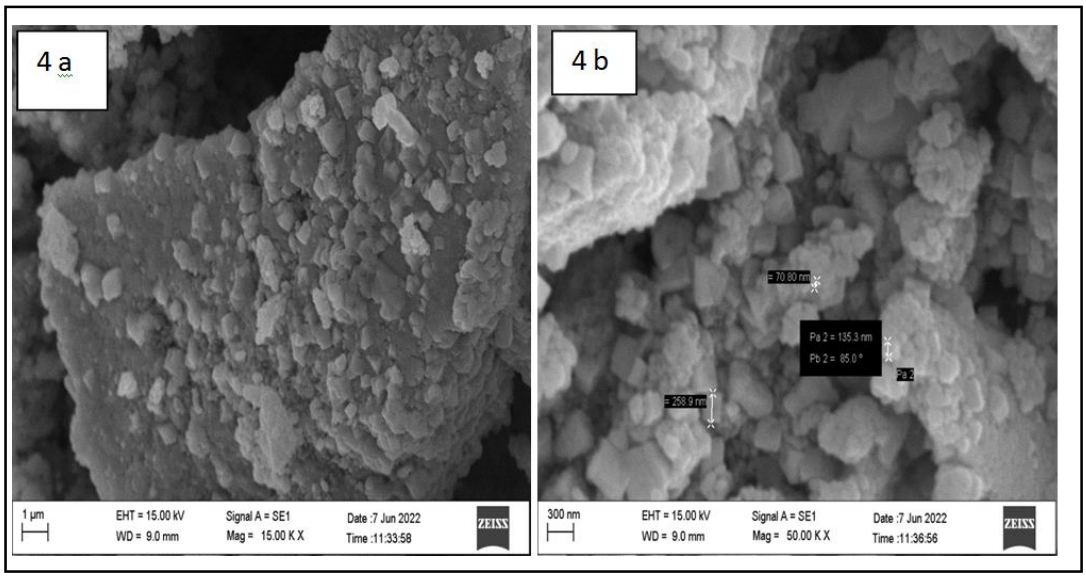


Figure 4: (a) SEM images of bioreduced silver nanoparticles synthesized by *E. alba;* (b) SEM image indicating size of bioreduced silver nanoparticles synthesized by *E. alba.*

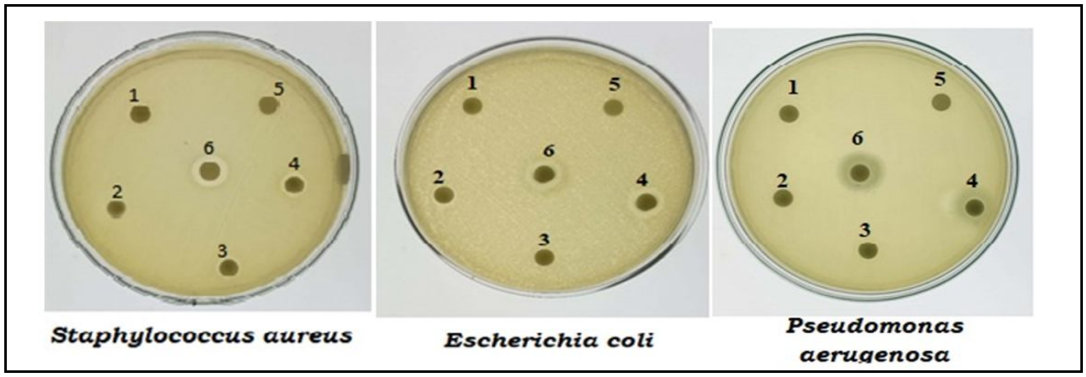


Figure 5: Antimicrobial activity of *E. alba* AgNPs against bacteria strain by disc diffusion assay. Different concentration of *E. alba* AgNPs sinificantly inhibited the growth of *Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa.* (1) 45 µg/ml (2) 90 µg/ml, (3) 180 µg/ml, (4)360 µg/ml, (5) Sterile distilled water (6) Streptomycin (0.125 mg/ml)

3.5 Antibacterial activity of synthesized silver nanoparticles

Table1 and Figure 6 show that biologically produced silver nanoparticles having strong antibacterial efficacy against clinically isolated multidrug-resistant human pathogens such as gram-positive bacteria, *Staphylococcus aureus* and gram-negative bacteria, *Pseudomonas aeruginosa* and *Escherichia coli*. Against *E. coli* and *P. aeruginosa, E. alba* mediated green synthesized AgNPs of 360 µg/ml concentration have shown significantly higher zones of inhibition with a diameter of 12 mm each (Figure 5). While minimum zone of inhibition was found in *Staphylococcus aureus* with a diameter of 10 mm corresponding to concentrations 360 µl/ml (Figure 5).

Table 1: Antibacterial activity of silver nanoparticles synthesized by *E. alba*

Samples/concentration (µg/ml)	Zone of inhibition (mm in diameter)		
	Staphylococcus aureus	*Escherichia coli*	*Pseudomonas aeruginosa*
45 µg/ml	NA	NA	NA
90 µg /ml	NA	NA	NA
180 µg /ml	9	NA	NA
360 µg/ml	10	12	12
Streptomycin	14	12	14
Sterile distilled water	NA	NA	NA

NA: No Activity

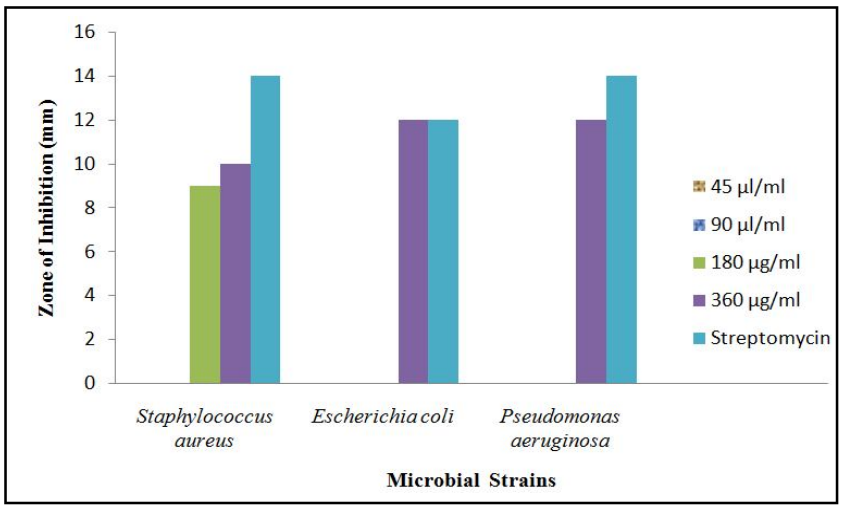


Figure 6: Antibacterial activity of silver nanoparticles synthesized by *E. alba*.

4. Discussion

It is important to develop easy, reliable and eco-friendly methods to increase interest in the synthesis and application of nanoparticles that are beneficial to mankind. It has been observed that silver ions are reduced into silver nanoparticles as a result of exposure to plant extracts. In an aqueous solution, silver nanoparticles showed dark reddish-brown colour because of surface plasmon resonance. In the current study, silver nanoparticles were synthesized using leaves extract of the *E. alba* plant. Interestingly, the silver nanoparticles synthesized within one hour of incubation made it one of the fastest methods for producing silver nanoparticles, and there was no significant change in their properties afterwards (Sulaiman *et al.,* 2013). The synthesis of *E. alba* was initially confirmed by characteristic colour change.

Spectrophotometry was used to evaluate the silver nanoparticles produced from the aqueous leaf extract of *E. alba* at a wavelength range of 200-800 nm. As a result of surface plasmon resonance of electrons induced by *E.alba* leaf extracts, the peak observed between 320-400 nm indicates the presence of silver nanoparticles. The broadening of peaks in the UV-vis spectrum indicates polydisperse particles (Ojo *et al.,* 2017). Various factors influence the frequency and width of surface plasmon absorption, including the metal nanoparticles' shape and size, as well as the dielectric constants of the metal itself and the surrounding medium. Nanoparticles in aqueous suspension can be examined by UV-vis spectroscopy for their size- and shape-controlled characteristics (Sulaiman *et al.,* 2013).

FTIR spectroscopy allows the detection of organic and inorganic species in small quantities with high sensitivity. The synthesised nanoparticle's FTIR spectra show distinct peaks at 410.99, 425.13, 443.79, 2163.44 and 2916.37 cm^{-1}. The alkynyl C-C stretches and C-H alkaline groups of silver nanoparticles are attributed to the peaks 2916.37 and 2163.44, respectively (Raju *et al.,* 2018; Banerjee *et al.,* 2014). Ring-opening vibration and the O-Si-O network are responsible for the peak at 443.79 cm^{-1} (Anandalakshmi *et al.,* 2016). Therefore, it could be said that AgNPs had a strong ability to bind to several functional groups of different polyphenolics and other plant-derived compounds, as well as proteins. This tends to suggest the

growth of a layer that covers AgNPs, inhibiting agglomeration by serving as a capping agent and giving the NPs stability (Huq and Akter, 2021; Ashour *et al.,* 2015).

The size, shape, morphology and surface chemistry of green-produced AgNPs were assessed using SEM analysis. Ambiguous evidence that AgNPs were cuboidal in form came from microscopic inspection (SEM). Furthermore, the particles looked to be heavily aggregated, perhaps as a result of the physical dehydration utilized during the SEM sample preparation process (Moodley *et al.,* 2018). Additionally, it can be deduced from the SEM image that silver nanoparticles typically range in size between 70 to 140 nm, which is inside the nano range.

It is reported that many of the nanoparticles are used as antimicrobial agents against antibiotic resistant bacteria and cytotoxicity activity. Among the different NPs, AgNPs have shown a significant applications against pathogenic microbes (Ribeiro *et al.*, 2023; Hug *et al.*,2022; Ahmad *et al*., 2020). It seemed to have the strongest antimicrobial effects against both gram-negative and gram-positive bacteria. It inhibits bacterial protein synthesis and DNA replication by interacting with thiol groups in proteins. Nanotechnology exploits the antibacterial properties of silver in the form of nanoparticles. Against gram-positive (*S. aureus*) and gram-negative bacteria (*E. coli* and *P. aeruginosa*), silver nanoparticles exhibit a substantial inhibitory action. It was specifically observed that the highest zone of inhibition was found against the gram-negative bacterium, *E coli* and *S. aureus* (Table 1 and Figure 6). However, gram-positive bacteria showed a reduced zone of inhibition compared to gram-negative bacteria, which may be because the cell wall's thick, hard coating of peptidoglycan complex prevented the entry of silver nanoparticles (Wasileska *et al*., 2023). AgNPs may easily access the nuclear material of bacteria due to their size, and they have a sizable and outstanding surface area that enables broad contact with bacteria. This might explain why they have the strongest antibacterial activity (Alduraihem *et al*., 2023; Ojo *et al.,* 2017). AgNPs can bind to the cell membrane suface and it can cause the alteration in the permeability and respiratory properties of the member (Alzubaidi *et al*., 2023). The interaction of particles relay on how accessible its surface area is. The smaller particles have greater antibacterial activity than larger particles due to their larger surface area (Lee and Jun, 2019).

From the current work, eco-friendly silver nanoparticles were effectively created. The present study conclusively proves that green synthesized silver nanoparticle of *E. alba* have high potential as an antibacterial agent.

5. Conclusion

The biosynthesis of approximately cuboidal, stable silver nanoparticles utilising *E. alba* leaf extract as a reducing and capping agent has been established in the current study. The biosynthesized nanoparticles showed potent antibacterial activity against both gram-positive (*S. aureus*) and gram-negative (*P. aeruginosa* and *E. coli*) pathogens. Against *E. coli* and *P. aeruginosa*, the *E. alba* mediated green synthesized AgNPs shown significantly higher zones of inhibition. While minimum zone of inhibition was found in *S. aureus*. The colour change was used to validate the creation of silver nanoparticles and UV-vis, SEM and FT-IR techniques were used to characterize them. Using UV-vis spectroscopy, the naturally innocuous AgNPs were further validated by analyzing the characteristic peak obtained at 320 nm and 400 nm. Some organic bio-compounds that function as a reducing and stabilising agent can be identified in leaf extract and are supported by FTIR peaks. Cuboidal AgNPs with diameters between 70 and 140 nm were validated by the SEM investigation. Finally, compared to chemical and physical synthesis methods, the green production of silver nanoparticles utilizing plant material was determined to be the most traditional and environmentally beneficial method.

Acknowledgements

Authors are very much thankful to the Principal, Mar Ivanios College, Thiruvananthapuram and the Central Laboratory for Instrumentation and Facilitation, University of Kerala, Thiruvananthapuram for providing all the facilities in the conduct of the wok.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

References

Ahmad, S. A.; Das, S. S.; Khatoon, A.; Ansari, M. T.; Afzal, M.; Hasnain, M. S. and Nayak, A. K. (2020). Bactericidal activity of silver nanoparticles: A mechanistic review. Materials Science for Energy Technologies, **3**: 756-769.

Alzduraihem, N. S.; Bhat, R. S.; Al-Zahrani, S. A.; Elnagar, D. M.; Alobaid, H. M. and Daghestani, M. H. (2023). Anticancer and antimicrobial activity of silver nanoparticles synthesized from pods of *Acacia nilotica*. Processes, **11**(2):301.

Alzubaidi, A. K.; Al-Kaabi, W. J.; Ali, A. A.; Albukhaty, S.; Al-Karagoly, H.; Sulaiman, G. M. and Khane, Y. (2023). Green synthesis and characterization of silver nanoparticles using flaxseed extract and evaluation of their antibacterial and antioxidant activities. Applied Sciences, **13**(4):2182.

Anandalakshmi, K.; Venugobal, J. and Ramasamy, V. (2016). Characterization of silver nanoparticles by green synthesis method using *Pedalium murex* leaf extract and their antibacterial activity. Applied Nanoscience, **6**(3):399-408.

Ashour, A. A.; Raafat, D.; El-Gowelli, H. M. and El-Kamel, A. H. (2015). Green synthesis of silver nanoparticles using cranberry powder aqueous extract: Characterization and antimicrobial properties. International Journal of Nanomedicine, **10**:7207.

Banerjee, P.; Satapathy, M.; Mukhopahayay, A. and Das, P. (2014). Leaf extract mediated green synthesis of silver nanoparticles from widely available Indian plants: Synthesis, characterization, antimicrobial property and toxicity analysis. Bioresources and Bioprocessing, **1**(1):1-10.

Bhattacharyya, A.; Bhaumik, A.; Rani, P. U.; Mandal, S. and Epidi, T. T. (2010). Nano-particles-A recent approach to insect pest control. African Journal of Biotechnology, **9**(24):3489-3493.

Fröhlich, E. (2012). The role of surface charge in cellular uptake and cytotoxicity of medical nanoparticles. International Journal of Nanomedicine, **7**:5577.

Hajipour, M. J.; Fromm, K. M.; Ashkarran, A. A.; de Aberasturi, D. J.; de Larramendi, I. R.; Rojo, T. and Mahmoudi, M. (2012). Antibacterial properties of nanoparticles. Trends in Biotechnology, **30**(10):499-511.

Haleemkhan, A. A.; Naseem, B. and Vardhini, B. V. (2015). Synthesis of nanoparticles from plant extracts. Int. J. Mod. Chem. Appl. Sci., **2**(3):195-203.

Huq, M. A. and Akter, S. (2021). Bacterial mediated rapid and facile synthesis of silver nanoparticles and their antimicrobial efficacy against pathogenic microorganisms. Materials, **14**(10):2615.

Huq, M. A.; Ashrafudoulla, M.; Rahman, M. M.; Balusamy, S. R. and Akter, S. (2022). Green synthesis and potential antibacterial applications of bioactive silver nanoparticles: A review. Polymers, **14**(4):742.

Jha, A. K.; Prasad, K.; Kumar, V. and Prasad, K. (2009). Biosynthesis of silver nanoparticles using *Eclipta* leaf. Biotechnology Progress, **25**(5): 1476-1479.

Lakkim, V.; Reddy, M. C.; Pallavali, R. R.; Reddy, K. R.; Reddy, C. V.; Bilgrami, A. L. and Lomada, D. (2020). Green synthesis of silver nanoparticles and evaluation of their antibacterial activity against multidrug-resistant bacteria and wound healing efficacy using a murine model. Antibiotics, **9**(12):902.

Lee, S. H. and Jun, B. H. (2019). Silver nanoparticles: Synthesis and application for nanomedicine. International Journal of Molecular Sciences, **20**(4):865.

Mandal, S.; Tiwari, P. K.; Kumari, A.; Bayal, N.; Anchal, A. and Upadhyay, K. (2023). Role of nanomaterials in microbial studies. Sustainable nanomaterials for biomedical engineering: Impacts, Challenges, and Future Prospects, pp:345.

Moodley, J. S.; Krishna, S. B. N.; Pillay, K. and Govender, P. (2018). Green synthesis of silver nanoparticles from *Moringa oleifera* leaf extracts and its antimicrobial potential. Advances in Natural Sciences: Nanoscience and Nanotechnology, **9**(1):015-011.

Moritz, M. and Geszke-Moritz, M. (2013). The newest achievements in synthesis, immobilization and practical applications of antibacterial nanoparticles. Chemical Engineering Journal, **228**:596-613.

Moteriya, P. and Chanda, S. (2017). Synthesis and characterization of silver nanoparticles using *Caesalpinia pulcherrima* flower extract and assessment of their *in vitro* antimicrobial, antioxidant, cytotoxic, and genotoxic activities. Artificial cells, Nanomedicine, and Biotechnology, **45**(8):1556-1567.

Mugale, M. N. and Balachandran, C. (2020). Haematological protective effect of raw extract and nano-*Eclipta alba*–treated rats in experimentally induced hepatocarcinogenesis. Comparative Clinical Pathology, **29**: 547-552.

Ndikau, M.; Noah, N. M.; Andala, D.M. and Masika, E. (2017). Green synthesis and characterization of silver nanoparticles using *Citrullus lanatus* fruit rind extract. International Journal of Analytical Chemistry, pp:10.

Ojo, O. A.; Oyinloye, B. E.; Ojo, A. B.; Afolabi, O. B.; Peters, O. A.; Olaiya, O. and Osunlana, O. (2017). Green synthesis of silver nanoparticles (AgNPs) using *Talinum triangulare* (Jacq.) Willd. leaf extract and monitoring their antimicrobial activity. Journal of Bionanoscience, **11**(4):292-296.

Prabhu, S. and Poulose, E. K. (2012). Silver nanoparticles: Mechanism of antimicrobial action, synthesis, medical applications, and toxicity effects. International Nano Letters, **2**(1):1-10.

Raja, S.; Ramesh, V. and Thivaharan, V. (2017). Green biosynthesis of silver nanoparticles using *Calliandra haematocephala* leaf extract, their antibacterial activity and hydrogen peroxide sensing capability. Arabian Journal of Chemistry, **10**(2):253-261.

Raju, L. and Manosathiyadevan, M. (2018). Green synthesis of silver nanoparticles from *Eclipta alba* (L.) collected from kolli hills and their larvicidal activity Against *Aedes egypti* (L.). Reviews, pp:10.

Ribeiro, A. I.; Vieira, B.; Dantas, D.; Silva, B.; Pinto, E.; Cerqueira, F. and Zille, A. (2023). Synergistic antimicrobial activity of silver nanoparticles with an emergent class of Azoimidazoles. Pharmaceutics, **15**(3):926.

Saminathan, K. (2015). Herbal synthesis of silver nanoparticles using *Eclipta alba* and its antimicrobial activity. Int. J. Curr. Microbiol. App. Sci., **4**(3):1092-1097.

Sulaiman, G. M.; Mohammed, W. H.; Marzoog, T. R.; Al-Amiery, A. A. A.; Kadhum, A. A. H. and Mohamad, A. B. (2013). Green synthesis, antimicrobial and cytotoxic effects of silver nanoparticles using *Eucalyptus chapmaniana* leaves extract. Asian Pacific Journal of Tropical Biomedicine, **3**(1):58-63.

Thankamani, V.; James, J.; Arunkumar, T.V. and Dev, L.M.S. (2011). Phytochemical screening and antimicrobial activity of *Alstonia scholaris* flower (L) R.Br. Int. J. Pharm. Res. Dev., **3**(4):172-178.

Tran, Q. H. and Le, A. T. (2013). Silver nanoparticles: Synthesis, properties, toxicology, applications and perspectives. Advances in natural sciences: Nanoscience and Nanotechnology, **4**(3):033001.

Vijayakumar, S.; Vinayagam, R.; Anand, M. A. V.; Venkatachalam, K.; Saravanakumar, K.M.; Wang, M. H. and David, E. (2020). Green synthesis of gold nanoparticle using *Eclipta alba* and its antidiabetic activities through regulation of Bcl-2 expression in pancreatic cell line. Journal of Drug Delivery Science and Technology, **58**:101786.

Wasilewska, A.; Klekotka, U.; Zambrzycka, M.; Zambrowski, G.; Œwiêcicka, I. and Kalska-Szostko, B. (2023). Physicochemical properties and antimicrobial activity of silver nanoparticles fabricated by green synthesis. Food Chemistry, **400**:133960.

Wayne, P.A. (2009). Performance standards for antimicrobial susceptibility testing, nineteenth informational supplement CLSI document, M100-S19. Clinical and Laboratory Standards Institute,USA. pp:136-139

Yadav, N.KJ., R.K.; Arya, K. Dev, C.; Sharma, Z.; Hossain, S.; Meena, K.R.; Arya, J.R.; Gayen, D.; Datta, and R.K. Singh. (2017). Alcoholic extract of *Eclipta alba* shows *in vitro* antioxidant and anticancer activity without exhibiting toxicological effects, Oxid. Med. Cell Longev. **90**:94641.

Citation **R. Anjali, Susan Kurian and Anjana Rajan (2023). Characterization and antibacterial response of biosynthesized silver nanoparticles using *Eclipta alba* (L.) Hassk. leaf extract. Ann. Phytomed., 12(1):453-459. http://dx.doi.org/10.54085/ap.2023.12.1.58.**