

BIOSYNTHESIS AND CHARACTERIZATION OF SILVER NANOPARTICLES FROM MANGROVE BARK – RHIZOPHORA MUCRONATA EXTRACTAJI JOVITHA AT^{1*}, DEIVASIGAMANI B²¹Centre for Advanced Studies in Marine Biology, Faculty of Marine Sciences, Annamalai University, Parangipettai, Tamil Nadu, India.²Centre for Advanced Studies in Marine Biology, Faculty of Marine Sciences, Annamalai University, Parangipettai, Tamil Nadu, India.

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ABSTRACT

Objective: The present study attempted to synthesize AgNPs from mangrove bark *Rhizophora mucronata* and analyze characteristics. The synthesized AgNPs analyzed with UV-vis spectroscopy, high-resolution transmission electron microscopy (HRTEM), and energy-dispersive X-ray (EDX) for confirming the nanoparticles.

Methods: The dried *R. mucronata* bark was powdered and kept in at 55°C for 15 min in a water bath and cooled at room temperature to get the extract. The *R. mucronata* bark extract was treated with silver nitrate and kept overnight in the dark environment which will turn the solution to dark brown color. The silver nanoparticles were characterized using UV-visible absorption at room temperature. Further characterization was also done with X-ray diffraction, high-resolution transmission electron microscope measurements, and DLS analysis.

Results: The synthesized AgNPs were analyzed with various analytical methods that revealed the abundant presence of silver nanoparticles. The UV-vis spectroscopy analysis exposed the surface plasmon resonance peak of 422 nm. High-resolution transmission electron microscopy (HRTEM) analysis indicated the size ranging from 10 nm to 200 nm in diameter and a spherical shaped poly dispersal of the particles. The energy-dispersive X-ray (EDX) and DLS also confirmed the presence of silver atoms.

Conclusion: Silver nanoparticles of *Rhizophora mucronata* bark revealed a well-defined structure and may be used in antimicrobial function in further researches.

Keywords: Biosynthesis, *Rhizophora mucronata*, Silver nanoparticles, HRTEM, UV-vis spectroscopy, EDX analysis. Dynamic light scattering.

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INTRODUCTION

Nanotechnology is one of the most promising areas in modern nanoscience and technology [1]. This technology includes the application of biological entities derived from prokaryotes to eukaryotes for the synthesis of nanoparticles, while plants assisted methods to involve simple, low-cost, eco-friendly, and fast procedures. Plants are containing different proteins and secondary metabolites such as alkaloids, quinines, flavonoids, terpenoids, and saponins [2,3] that are involved in the synthesis of AgNPs. There are several methods for the preparation of silver nanoparticles (AgNPs), including electrochemical method, laser ablation, microwave irradiation, thermal decomposition, and sonochemical synthesis. Moreover, these methods have some advantages over the conventional method, such as cost-effectiveness and in the process of production, it does not require high temperature, pressure, energy, and toxic chemicals. In general, metallic nanoparticles are mostly prepared by noble metals such as silver, platinum, and gold [4,5].

Thus, the synthesis of silver nanoparticles by chemical methods leads to the presence of some toxic chemical species observed on the surface. Therefore, the synthesis of nanoparticles using biological entities can potentially eliminate this problem by making the nanoparticle more biocompatible [6,7]. Among all the noble metal nanoparticles, silver nanoparticles are considered to be of great importance due to their high antiviral, antibacterial and antifungal properties combined with good electrical conductivity, chemical stability and catalytic activity [8,9]. Due to the above noble and proven qualities, silver nanoparticles are been used widely in medical, cosmetics, and pharmaceutical industries. [10].

The *Rhizophora mucronata* species of mangrove plants belong to the *Rhizophoraceae* family and a large distribution in the indo-pacific region. In the present study, AgNPs were synthesized from *Rhizophora mucronata*. They were characterized using UV-visible spectroscopy, high-resolution transmission electron microscopy (HRTEM), energy-dispersive X-ray (EDX) spectroscopy, and dynamic light scattering (DLS).

MATERIALS AND METHODS**Sample collection and preparation**

To synthesize AgNPs, the bark of the mangrove plant *Rhizophora mucronata* was gathered from the Pichavaram mangrove forest located in the higher land of Vellar estuarine complex. The collected samples were first washed with seawater, then with tap water, and at last with distilled water to eliminate the foreign particles and contaminants. Then, they were shade-dried at room temperature for 2 weeks. A mixer grinder was used to powder the dried specimens [11]. Silver nitrate was purchased from HiMedia Laboratories, (Chennai, India).

Preparation of *R. mucronata* bark extract

The air-dried bark extract was ground to a coarse powder using a blender; 5.0 g of the dried bark powder sample was taken and mixed with 100 ml of glass distilled water. This mixture was kept at 55°C for 15 min in a water bath and cooled to room temperature and filtered through Whatman No. 1 filter paper. This aqueous bark extract was refrigerated and used for further experiments.

Synthesis of AgNPs

A total of 1 ml of the *R. mucronata* bark aqueous extract was added to 9 ml of 1 mM solution of silver nitrate in a 15 ml test tube. The

response of chemical was performed in the dark at room temperature overnight to minimize the photoreduction of silver nitrate. The aqueous bark extracts of *R. mucronata* and AgNO solution were used as control. After the desired reaction period, the solution containing the AgNPs was centrifuged at 10,000 rpm for 15 min. The pellet was collected and dispersed in glass-distilled water, removing any interactive biological molecules. This was repeated thrice to confirm better separation of the AgNPs and was used for characterization studies.

CHARACTERIZATION OF AGNPS

UV-visible spectroscopy

The formation of dark brown color during the synthesis was confirmed as the formation of AgNPs [12]. The bioreduction of the pure silver nanoparticles were recorded under UV-visible spectroscopy range between 400 nm and 700 nm. The UV-visible spectra of the mangrove bark extract and silver nitrate solution also recorded.

HRTEM

A drop of Silver nanoparticles was placed on carbon-coated copper grids and allowed to stand for 2 min, and the excess solution was eradicate using a blotting paper allowed to dry at room temperature. High-resolution transmission electron microscopy regards were made on an FEI-TECNAI G2 T-30 and S-Twin instrument for silver nanoparticles.

EDAX analysis

Energy-dispersive X-ray spectroscopy is an analytical technique used for the analysis of elements. EDX analysis is done for the further characterization of the synthesized silver nanoparticles. The presence of silver nanoparticles was confirmed by EDX analysis and mostly showed strong signal energy peaks for silver atoms in the range 2-4 KeV.

Dynamic light scattering

DLS is most commonly used to analyze nanoparticles. Nanoparticle size can be determined by measuring the random changes in the intensity of light scattered from a suspension or solution. However, this application is much less regular than particle sizing.

RESULTS AND DISCUSSION

UV-visible spectroscopy

R. mucronata bark (Fig. 1) aqueous extract added with silver nitrate at 1 mM showed a change in color from pale yellow to dark brown (Fig. 2). The UV-vis spectrum displayed surface plasmon resonance (SPR) peak bands centered at 420 nm. The reduction of AgNPs in the aqueous solution of the silver complex during the reaction with the bark extract of *R. mucronata* was confirmed by the UV-visible spectra. Absorption spectra of AgNPs at 422 nm after 24 h of incubation sharpening of peak indicated that the particles are found monodispersed (Fig. 3).

HRTEM and EDAX analysis

The structure and size of silver nanoparticles were characterized using HRTEM. The HRTEM images clearly visualize the AgNPs synthesized from the bark extract of *R. mucronata* (Figs. 4 and 5). The particles



Fig. 1: *Rhizophora mucronata* Bark

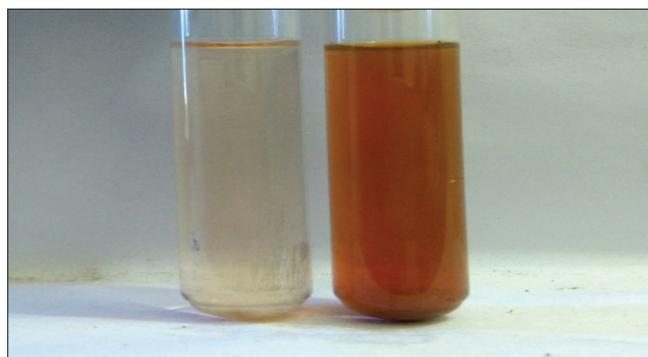


Fig. 2: Optical Photographs of synthesized AgNPs from *Rhizophora mucronata* Bark

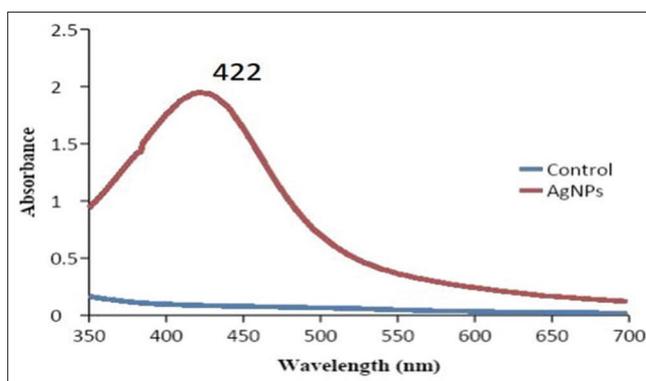


Fig. 3: UV-visible spectrum of the AgNPs synthesized from *Rhizophora mucronata* bark

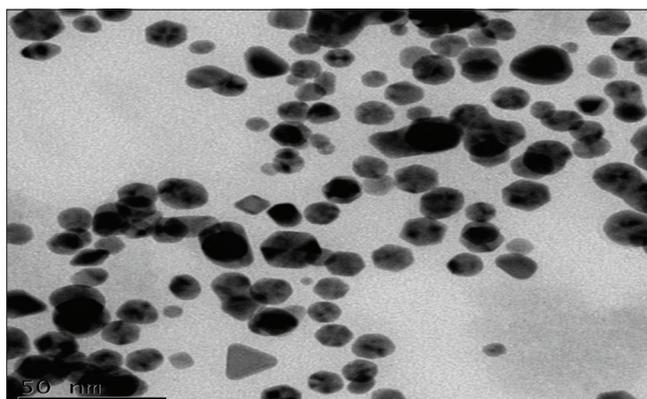


Fig. 4: High-resolution transmission electron spectroscopic image of AgNPs [scale bar 50 nm]



Fig. 5: High-resolution transmission electron spectroscopic image of AgNPs (scale bar 200 nm)

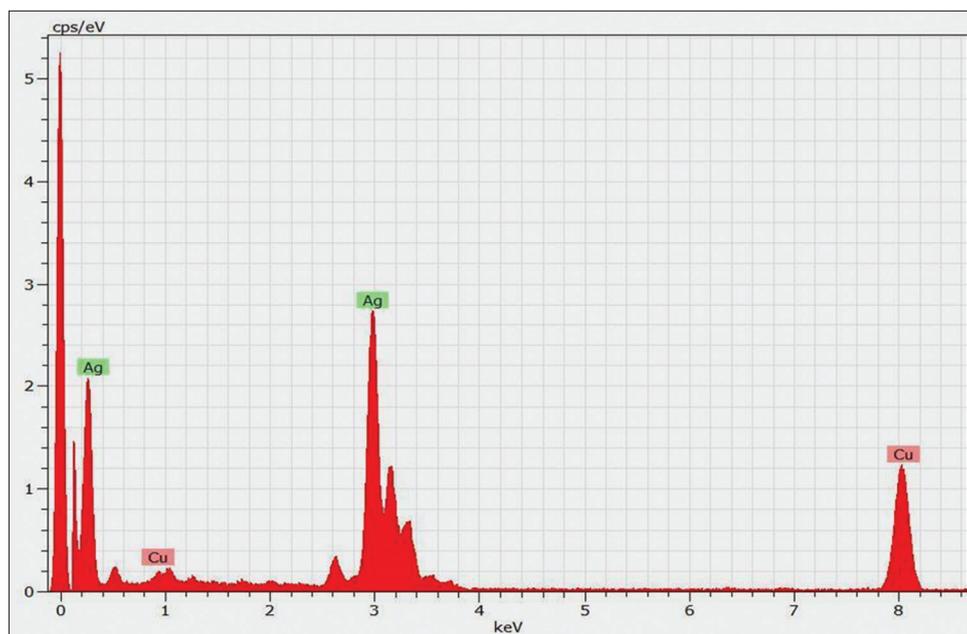


Fig. 6: Energy-dispersive X-Ray spectrum of AgNPs

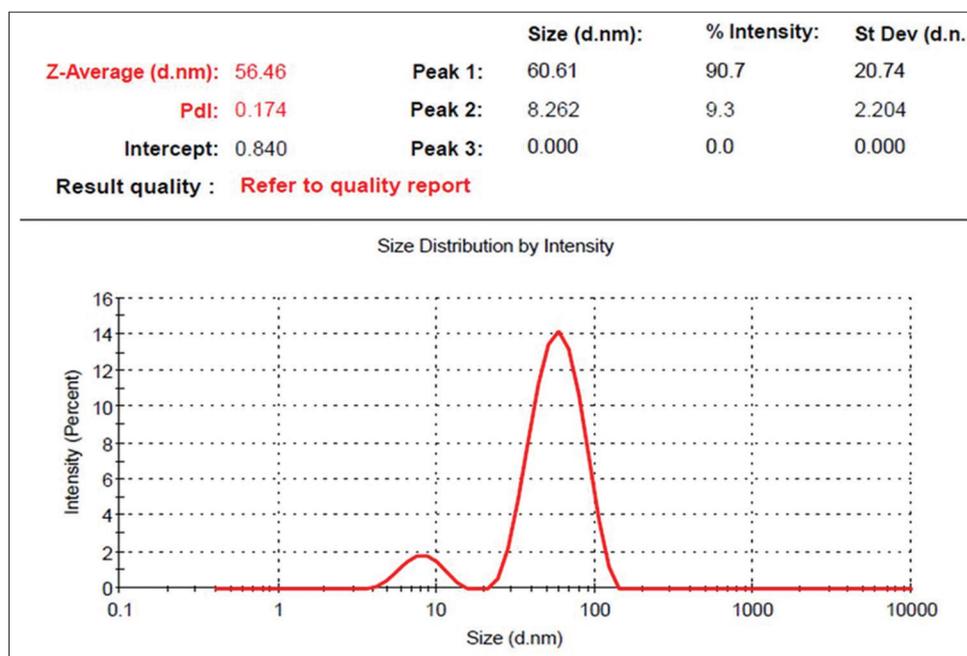


Fig. 7: DLS measurement analysis of AgNPs

are predominantly spherical in shape with a diameter ranging from 10 nm to 200 nm. The EDAX (Fig. 6) spectrum shows a single signal for silver, indicating that the synthesized AgNPs were free from impurity. The strong signal of the silver atoms of the *Rhizophora mucronata* bark extract indicated the crystalline property. Moreover, the AgNPs covered by phytoconstituents along with oxygen atom confirmed from the O peaks and the Ag signals on the image. The selected area electron diffraction pattern of the nanoparticles explained the face-centered cubic (fcc) crystalline structure of silver with different diffracting index.

Dynamic light scattering

Dynamic light scattering analysis was used to measure the hydrodynamic diameter using the diffusion coefficient of the monodisperse

colloidal particles present in the silver nanoparticles. From Fig. 7, the size and distribution of AgNPs were recorded at 60 nm. [1] reported that the particle size of AgNPs ranged from 5 nm to 15 nm, and more than 50% of the particles were observed at 20 nm to 110 nm.

CONCLUSION

The biocompatible AgNPs were successfully synthesized using *R. mucronata* bark extracts. The AgNPs were stable for up to 12 months. The green synthesis method used in this study for the preparation of AgNPs using *R. mucronata* bark is quite fast and of low-cost technique. The results revealed that the biologically synthesized AgNPs could be of immense use in the medical field for their antimicrobial function and would be an efficient immunostimulant for the marine animals.

CONFLICTS OF INTEREST STATEMENT

The authors declare that there are no conflicts of interest with any other study against this research.

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AUTHORS' CONTRIBUTIONS

Dr.B. Deivasigamani conceived the research and provided the methods, whereas A.T Aji Jovitha conducted experiment in the laboratory, analyzed and interpreted the obtained data. Both the authors contributed equally in drafting the manuscript.

REFERENCES

- Narayanan KB, Sakthivel N. Green synthesis of biogenic metal nanoparticles by terrestrial and aquatic phototrophic and heterotrophic eukaryotes and biocompatible agents. *Adv Colloid Interface Sci* 2011;169:2-59.
- Priya K, Krishnakumari S. Phytochemical analysis of *Achyranthes aspera* and its activity on sesame oil induced lipid peroxidation. *Anc Sci Life* 2007;27:6-10.
- Netala VR, Ghosh SB, Bobbu PL, Anitha D, Vijaya T. Triterpenoid saponins: A review on biosynthesis, applications and mechanism of their action. *Int J Pharm Pharm Sci* 2014;7:24-8.
- Kathiresan K, Manivannan S, Nabeel MA, Dhivya B. Studies on silver nanoparticles synthesized by a marine fungus *Penicillium fellutanum* isolated from coastal mangrove sediment. *Colloid Surf B* 2009;71:133-7.
- Leela A, Vivekanandan M. Tapping the unexploited plant resources for the synthesis of silver nanoparticles. *Afr J Biotechnol* 2008;7:3162-5.
- Srikanth K, Vikram G, Rama Swamy N. Eco-friendly and green synthesis of silver nanoparticles using leaf extract of *Strychnos potatorum* Linn. F. and their bactericidal activities. *3 Biotech* 2014;5:706-14.
- Sastry M, Ahmad A, Khan MI, Kumar R. Biosynthesis of metal nanoparticles using fungi and actinomycete. *Curr Sci India* 2003;85:162-70.
- Thakkar KN, Mhatre SS, Parikh RY. Biological synthesis of metallic nanoparticles. *Nanomedicine* 2010;6:257-62.
- Rogers JV, Parkinson CV, Choi YW, Speshock JL, Hussain SM. A preliminary assessment of silver nanoparticle inhibition of monkey pox virus plaque formation. *Nanoscale Res Lett* 2008;3:129-33.
- Panacek A, Kolar M, Vecerova R, Prucek R, Soukupová J, Krystof V, et al. Antifungal activity of silver nano particles against *Candida* spp. *Biomaterials* 2009;30:6333-40.
- Keat CL, Aziz A, Eid AM, Elmarzugi NA. Biosynthesis of nanoparticles and silver nanoparticles. *Bioresour Bioproc* 2015;2:47.
- Zhang XF, Liu ZG, Shen W, Gurunathan S. Silver nanoparticles synthesis, characterization, properties, applications, and therapeutic approaches. *Int J Mol Sci* 2016;17:1534.
- Patil RS, Kokate MR, Kolekar SS. Bioinspired synthesis of highly stabilized silver nanoparticles using *Ocimum tenuiflorum* leaf extract and their antibacterial activity. *Spectrochim Acta A Mol Biomol Spectrosc* 2012;91:234-8.