Research Article

A Novel Green Synthesis of *Syzygium cumini* Leaves Extract Coated Silver Nanoparticles

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Abstract

Silver nanoparticles have been the subjects of researchers because of their unique properties. In last decades, numerous efforts were made to develop green methods of synthesis to avoid the hazardous byproducts. The investigation was aimed to synthesize silver nanoparticles using leaves extract of *Azadirachta indica* and *Syzygium cumini* leaves extract coated silver nanoparticles. Aqueous extract of plant was used and synthesis of nanoparticles confirmed by the colour change, followed by Transmission Electron Microscopy (TEM). The synthesized silver nanoparticles were spherical in shape. In TEM analysis, the average size of SNPs was found to be 24.38 nm and *Syzygium cumini* leaves extract coated silver nanoparticles were 28.11 nm in size. There was increase in size of plant extract coated silver nanoparticles as compared with silver nanoparticles which suggests that *Syzygium cumini* leaves extract has affinity for silver nanoparticles and capable of coating them.

Keywords: Silver nanoparticles (SNPs), *Syzygium cumini*, Transmission Electron Microscopy (TEM), *Azadirachta indica*

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Introduction

Nanotechnology is a 21st century science and because of its unique features it has found applications in a variety of biological domains. Metal nanoparticles less than 100 nm in diameter have made a substantial impact on diagnostic and medical devices for personalized healthcare practice. SNPs have great potential in a broad range of applications as antimicrobial agents, biomedical device coatings, drug-delivery carriers, imaging probes and diagnostic and optoelectronic platform [1]. Nanotechnology is rapidly growing and utilized in wide range of commercial products throughout the world i.e. electronics, bio-sensing, food industry, clothing, medical devices, paints, sunscreens, cosmetics, molecular diagnostics and therapeutics [2].

Different processes, such as chemical, physical, irradiation and biological methods, can be used to create nanomaterials. Chemical techniques involved in the synthesis of nanomaterials generate a substantial number of hazardous byproducts, the development of novel chemical or physical approaches has resulted in environmental damage. As a result, green nanotechnology is required, which involves a clean, safe, environmentally friendly and non-toxic method of nanoparticle manufacturing [3]. Among the various metallic nanoparticles, silver nanoparticles (SNPs) are one of the most important and fascinating nanomaterials that play a part in biomedical applications. SNPs play a significant role in nanoscience, nanotechnology, particularly in nanomedicine. Although a variety of noble metals have been employed in the past, SNPs have been studied for potential applications in cancer diagnosis and treatment [4].

Syzygium cumini Linn is a tropical evergreen plant. It is a member of the Myrtaceae family. Java plum, Portuguese plum, Malabar plum, purple plum, Black plum, Jamun and Indian Blackberry are all common names for this fruit. *Syzygium cumini* is also known as *Syzygium jambolanum, Eugenia cumini* and *Eugenia Jambolanala. Syzygium cumini* is a tiny black-purple drupe-bearing plant native to the Indian subcontinent with a wide range of biological functions. Which includes plants with anti-diabetic, anti-inflammatory and antioxidant properties. Activities include antipyretic, antioxidant, antimicrobial, anticancer, and antibacterial. *Syzygium cumini* has long been prized for its nutritional and therapeutic properties. It has a wide range of pharmacological activities, plant materials such as bark, leaves, seeds and fruit have all been employed medicinally in the treatment of a variety of illnesses. *Syzygium cumini* leaves have been utilised for antibacterial and antidysenteric purposes. *Syzygium cumini* seeds have antibacterial, anti-inflammatory, antioxidant and anticancer properties. *Syzygium cumini* bark has been used as diuretic. The ripe fruits of *Syzygium cumini* have been reported to have cardiovascular effects such as hypotensive, antioxidant and anti-inflammatory properties. *Syzygium cumini* has long been used to treat diabetes. Apart it also contains anti-allergic, anti-fungal, antibacterial, anti-cancer, anticlastogenic properties, radioprotective,

chemoprotective, antidiarrhoeal, antifertility, antihyperlipidemic, antihypertensive as well as hepatoprotective effects [5]. Flavonoids, alkaloids, tannins, glycosides, steroids, saponins, phenolics, terpenes, anthraquinones and essential oils are the phytochemicals present in *Syzygium cumini* plant [6].

Materials and Methods

Collection and authentication of plant material

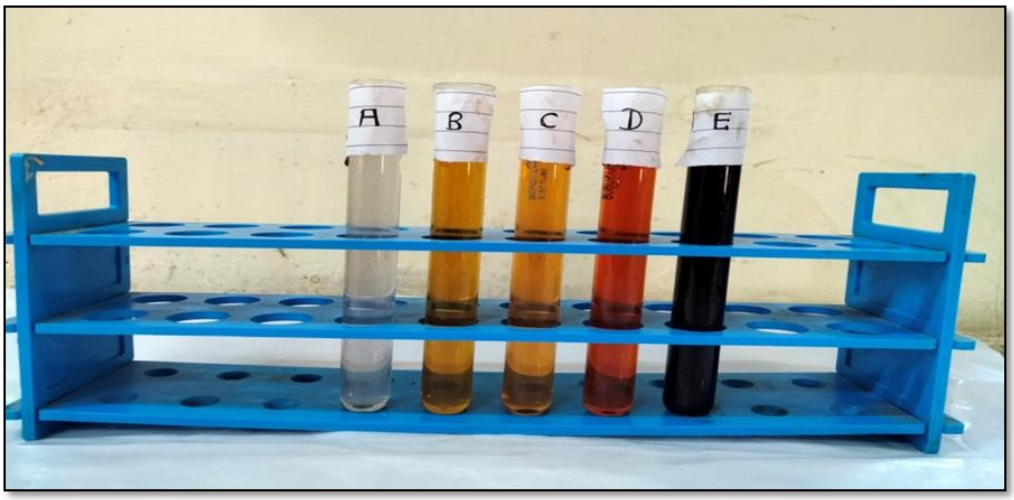
Department of Botany, Vasantrao Naik Marathwada Agricultural University, Parbhani, recognised the *Azadirachta indica* and *Syzygium cumini* plant material. The material was collected in bulk from the same farm and transported in batches to the laboratory, where it was processed for further research.

Preparation of Syzygium cumini leaves extract

Aqueous extract of *Syzygium cumini* Linn. leaves extract was prepared by cold extraction method. They were washed and allowed to dry completely under shade. Dried leaves were ground to powder with the help of an electrical grinder. Then, 20% of aqueous solution was made by dissolving 200 grams of powder in double distilled water and the final quantity was made to 1 liter. It was mixed thoroughly and allowed to soak for 48 hours at room temperature. It was shaken intermittently with an electrically operated flask shaker. Thus, resulting solutions was first filtered by using muslin cloth and then by ordinary filter paper onto glass plates. They were allowed to dry under shade and aqueous extract of *Syzygium cumini* was obtained. The weight of empty glass plates were noted and final weight after drying was obtained. The increase in weight was considered as the amount of aqueous extract obtained. Resulting extracts were scraped off the glass plate and stored in a container at 4^0C for preparing *Syzygium cumini* leaves extract coated silver nanoparticles.

Bio-Synthesis of Silver nanoparticles (SNPs)

The 5 gm washed and shade dried leaves of *Azadirachta indica* were cut into small pieces (do not grind), dispense in 100ml of sterile distilled water and kept in water bath for one hour at 80ºC. Then the leaves extract was collected in separate conical flask and filtered through Whatmans No. - 42 filter paper. 10^{-3} M Silver Nitrate ($AgNo_3$) solution was prepared and stored in brown bottles. 5ml of leaves extract was taken in BOD bottle separately and to this 95 ml of 10^{-3} M $AgNo_3$ solution was added. Then the BOD bottle were incubated for 28 hours at room temperature. The colour changes of the solution from pale green to dark brown was checked periodically. The colour change to brown indicates synthesis of SNPs.



Scheme 1. Silver Nanoparticles formation and their corresponding colour changes.

A. 1mM solution of Silver Nitrate ($AgNo_3$)
B. Natural colour of 5% aqueous leaves extract of *Azadirachta indica* (Neem) plant.
C. Ruby red colour formed after addition of 1 mM solution of Silver Nitrate ($AgNo_3$) and 5% aqueous leaves extract of *Azadirachta indica* (Neem) plant and 0.5 hour of incubation.
D. Ruby red colour darkens to reddish brown after addition of 1 mM solution of Silver Nitrate ($AgNo_3$) and 5% aqueous leaves extract of *Azadirachta indica* (Neem) plant and 2 hours of incubation.

E. Synthesis of *Syzygium cumini* leaves extract coated Silver Nanoparticles (SNPs).

Synthesis of Syzygium cumini leaves extract coated silver nanoparticles

The nanoparticle solution (0.17mg/kg body weight) was taken and coated by adding aqueous leaves extract of *Syzygium cumini* (800mg/kg body weight). The resultant solution was kept at room temperature for an hour while being swirled to covalently change the surface of SNPs with the phytochemicals of *Syzygium cumini* leaves extract.

Characterization of silver nanoparticles

Transmission Electron Microscopy imaging technique was used to characterise silver nanoparticles (SNPs). For TEM imaging, dip preparation for SNPs by floatation method was done [7]. A drop of sample was placed on a piece of parafilm and the carbon coated copper grid was placed. After 5-10 min, the excess was drained with the help of filter paper. The sample was washed with distilled water and then stained with 2% Uranyl acetate, air dried and observed under transmission electron microscope (Model – JEM-1400, HR) at various magnifications as per the standard protocol at NIAB (National Institute of Animal Biotechnology), Hyderabad.

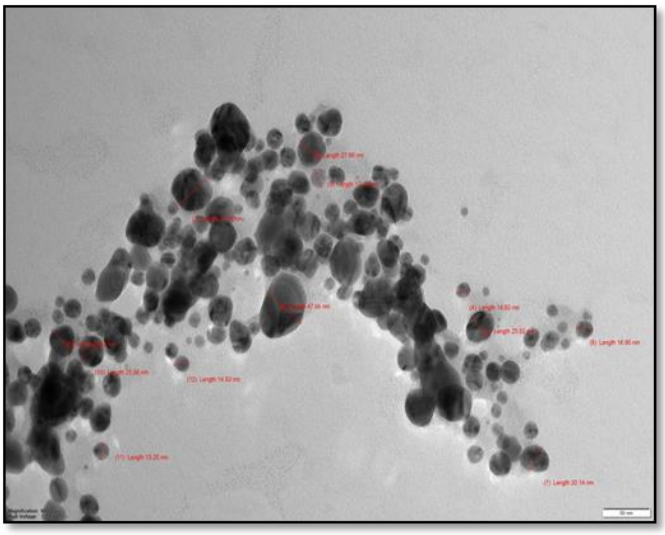
Results and Discussion

The purpose of present study was to conduct synthesis silver nanoparticles using leaves extract of *Azadirachta indica* and synthesis of *Syzygium cumini* leaves extract coated silver nanoparticles.

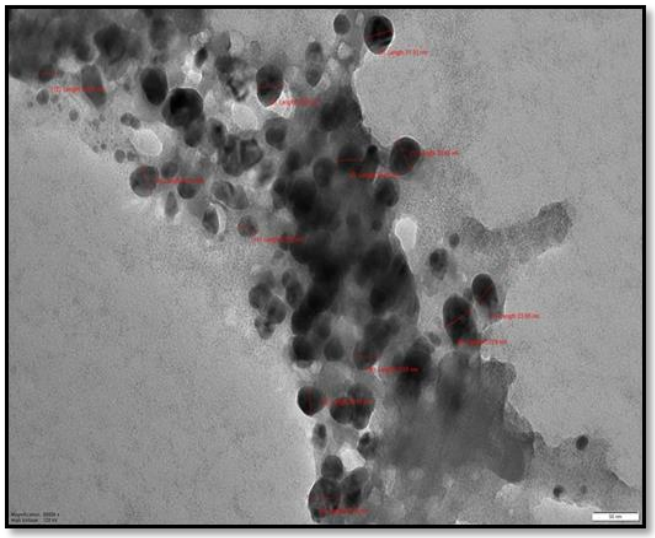
Syzygium cumini leaves extract

The current study involved the manufacture of an aqueous leaves extract of *Syzygium cumini*. Fresh *Syzygium cumini* leaves weighing 200 g were homogeneously mashed in a mortar and combined with 1000 ml distilled water and maintained for 24 hours. The slurries were then strained through two layers of muslin fabric and filtered with Whatman filter paper No. 42 then centrifuged for 10 minutes at 4500 rpm. The supernatant was regarded as containing 20% aqueous extract.

The moisture content of *Syzygium cumini* leaves was determined to be 64%. The leaves extract yield was 6.12 gm in distilled water from 50 gm powder material, with a percentage yield of 12.24%. The extract was reddish brown to greenish brown in colour, had a solid and sticky consistency, was astringent in flavour and had a dumpy aromatic odour. Our findings are in agreement with the reports of Wankhade., 2016 [8].



TEM image of Silver Nanoparticles (SNPs) (24.38 nm)



TEM image of *Syzygium cumini* leaves extract coated silver nanoparticles (28.11 nm)

Synthesis, coating and characterization of SNPs

Silver nanoparticles were synthesised using a biological process. In this experiment, dried leaves of *Azadirachta indica* were cut into small pieces distributed in 100 ml of sterile distilled water before being maintained in an 80℃ water bath for an hour. The leaf extract was then collected in a separate conical flask and filtered through Whatman

filter paper No.42. A 10^{-3} M silver nitrate solution was also made and stored separately in brown bottles. 5 mL leaves extract was added to 95 mL 10^{-3} M $AgNo_3$ solution and incubated for another 28 hours at room temperature. The change in colour from pale green to dark brown suggested that SNPs were being synthesised by the leaves which was very well visible to naked eyes. Similar observations were found by Dhaije., 2018 [9]. The TEM images showed the formation of silver nanoparticles of spherical shape. By measuring diameter of these nanoparticles average particle size found was 24.38 nm. Similar observations were obtained by Mishra., 2016 [10] and coating of silver nanoparticles was also obtained by TEM images. In these images *Syzygium cumini* leaves extract coated silver nanoparticles were found spherical in shape and 28.11 nm in size. There was increase in size of plant extract coated silver nanoparticles as compared with silver nanoparticles which suggests that *Syzygium cumini* leaves extract has affinity for silver nanoparticles and capable of coating them. Similar observation of increase in particle size after coating were also reported by Mishra., 2016 [10]. Transmission Electron Microscopy was used to characterise the produced and purified SNPs, according to the procedure of Gurunathan *et al*., 2009 [11].

Conclusions

From present investigation it can be concluded that *Azadirachta indica* (Neem) leaves can be successfully used in the biological reduction process to produce silver nanoparticles. *Syzygium cumini* leaves extract coated silver nanoparticles can be satisfactorily synthesised.

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Publication History
Received 24.05.2022
Revised 27.11.2022
Accepted 28.11.2022
Online 30.11.2022