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Green Synthesis of AgNPs Mediated by Using Aqueous Leaf Extract of *Coriandrum sativum*

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A B S T R A C T

Nanoparticles have attracted scientific responsiveness due to their fascinating properties, commercial and biotechnological applications advantageous over their bulk counterparts This is principally due to their small size and, subsequently, the remarkable surface area of NPs. Presently, advances in the synthesis, stabilization and production of AgNPs have adopted a new generation of commercial products and intensified scientific investigation within the nanotechnology field. In the present study, Ag NPs are synthesized in the presence of the aqueous leaf extract of *Coriandrum sativum. The biosynthesized nanoparticles were characterized by* UV– Vis spectra, FT-IR, XRD, and SEM analysis. Further, the antibacterial activity of the biosynthesized nanoparticles was tested against the selected bacterial isolates. Ag NPs exhibited maximum antagonistic activity towards *Pseudomonas aeruginosa.*

Introduction

Ever since the origin of life on the globe, biological entities and inorganic materials have been in persistent touch with each other. Due to this consistent interaction, life could endure on this world with a regimented deposit of minerals. Recently the global research focus is aimed towards the interaction between inorganic molecules and biological species (Tharanya *et al.*, 2015). Nanoparticles have attracted scientific responsiveness due to their fascinating properties, commercial and biotechnological applications advantageous over their bulk counterparts (Daniel and Astruc, 2004). Nanotechnology is termed as the study and utilization of structures between 1 to 100 nm in size. Nanotechnology combines chemical engineering, mechanical, microelectronics, electrical, material sciences, and biological screening. Currently, there are more than 300 declared products of nanotechnology in the market. Nanoparticles demonstrate novel or improved size-dependent properties contrasted with bigger particles of similar material (Patra *et al.,* 2018).

Nanoparticles are synthesized by a variety of physical, chemical, biological, and hybrid methods (Karthika *et al.,* 2015). Methods employed for the synthesis of nanoparticles are broadly classified under two processes such as “Top-down” process and the “Bottom-up” process. Top-down approach: Bulk material is broken down into particles at the nanoscale with various lithographic techniques e.g.: grinding, milling, etc. Bottom-up approach: Atoms self-assemble to new nuclei which grow into a particle of nanoscale (Kavitha *et al.,* 2013). Currently, the biological method is also followed for the green chemistry synthesis of metal nanoparticles due to it being free from hazardous chemicals. The metal nanoparticle synthesis method from plant extracts has more advantages over the microbial synthesis method because the microbial process is highly expensive due to the cost of microorganism isolation and their culture maintenance. Therefore, the development of safe, eco-friendly, reliable, and non-toxic methods for the synthesis of nanoparticles is of utmost importance to expand their biomedical applications (Li *et al.,* 2011).

Nanotechnology is currently employed as a tool to explore the darkest avenues of medical sciences to combat diseases caused by drug-resistant microbes (Singh *et al.,* 2014). Nanomedicines have become well esteemed in recent times owing to nanostructure’s utilization as delivery agents by encapsulating drugs and targeted delivery in specific tissues. Nanoparticle-based products have been developed both for imaging in cancer diagnosis and also for pharmacotherapeutic management (Haba *et al.,* 2007). Microbial drug resistance has emerged as a global health concern, as microbes acquire resistance by changing their metabolic activities and genetic structure. Nanotechnology is expected to open some new aspects to fight and prevent diseases using atomic-scale tailoring of materials (Afreen *et al.,* 2011). The present study deals with the eco-friendly biogenic approach for the synthesis of Ag NPs using the aqueous leaf extract of *Coriandrum sativum* and its characterization by UV– Vis spectra, FT-IR, XRD, and SEM analysis. Further, the antibacterial activity of the biosynthesized nanoparticles was tested against the selected bacterial isolates.

Materials and Methods

Collection of Plant Materials

Fresh healthy leaves of *Coriandrum sativum* without any infection were selected and collected from the areas in and around Vellore, Vellore District. The fresh and mature leaves of *Coriandrum sativum* were used for the study. Leaves were separated from the plant and washed thoroughly with running water to remove any dirt or debris on the surface and finally rinsed briefly in deionized water before use.

Preparation of plant extracts (Aqueous Extract)

20 g of fresh leaves of *Coriandrum sativum* were washed thoroughly with double distilled water, chopped into small pieces, grounded into a fine paste, and added to 100 ml of sterile double distilled water. Furthermore, the extract was then filtered using Whatman filter paper, and the filtrate was incubated at room temperature for 1 h to prepare the aqueous extract and was used for the synthesis of the Ag NPs. The extract was stored at 4°C for further experiments.

Biosynthesis of silver nanoparticles using the aqueous extract of *Coriandrum sativum*

1.5 ml of the aqueous leaf extract of *Coriandrum sativum* was added to 30 ml of 3 Mm $AgNO_3$ (99.99%) aqueous solution, stirred well, and incubated at room temperature. The experiment was done in triplicate for reproducibility. After 10 min, the color of the solution changed from yellow to a brown color indicating the formation of Ag nanoparticles. The solution containing silver nanoparticles was separated and concentrated by repeated washing and centrifugation at 10,000 rpm for 15 min. The final suspension was dried and the nanoparticle obtained was stored and used for further experimental studies.

Characterization of synthesized Ag NPs

The bioreduction of ions in the solution was monitored by periodic sampling of aliquots (1 mL) of the aqueous component after 20 times dilution and measured in the UV–Vis spectra. Samples were monitored as a function of time of reaction using Shimadzu 1601 spectrophotometer in the 100–700 nm range operated at a resolution of 1 nm. The purified and dried pellets of synthesized Ag NPs were subjected to X-ray diffraction (XRD) analysis. The IR spectra were recorded on a SHIMADZU FTIR-8400S spectrometer at room temperature. The particle size and morphology of the silver nanoparticles were examined using Scanning electron microscopic observations. SEM measurements were performed on a JEOL JSM 6390 instrument operated at an accelerating voltage at 15kV (Tharanya *et al.,* 2015).

Antibacterial Activity of *Coriandrum sativum* extracts and Ag NPs

Screening of antibacterial activity was performed by well diffusion technique. The Muller Hinton Agar (MHA) plates were seeded with 0.1 ml of the standardized inoculum of each test organism. The inoculum of *E. coli, Pseudomonas* sp., *Proteus* sp., and *Salmonella* sp. was spread evenly over the plate with a sterile swab.

A standard cork borer of 6 mm diameter was used to cut uniform wells on the surface of the MHA and 100 µl of *Coriandrum sativum* leaf extracts (Ethanolic, Hexane, and Aqueous) and Ag NPs (100 µg/ml) was introduced in 4 separate wells. The inoculated plates were incubated at 37 ^{o}C for 24 h and the zone of inhibition was measured.

Results and Discussion

Green nanotechnology in the development of material synthesis is of considerable importance to expand its biological applications. Currently, a wide array of inorganic nanoparticles has been synthesized by using biogenic enzymatic methods and their applications in many cutting-edge technological areas have been explored. Nanomedicine is a burgeoning field of research with tremendous prospects for the improvement of the diagnosis and treatment of human diseases.

Biogenic Approach for the Synthesis of Ag Nanoparticles

The Synthesis of Ag Nanoparticles by a biogenic approach using the aqueous leaf extract of *Coriandrum sativum* was shown in Fig.1. The color of the solution changed from yellow to dark brown indicating the formation of Ag nanoparticles.

The solution containing silver nanoparticles was separated, concentrated, and dried. The resultant nanoparticle powder obtained was stored and used for further experimental studies.

Characterization of Ag Nanoparticles

UV–VIS spectral analysis

UV–Vis spectroscopy is one of the most significant tools to characterize metal nanoparticles. In our results peak specific for the synthesis of silver nanoparticles was obtained at 464 nm by UV Visible spectroscope in the form of a sharp peak (Fig.2). The absorption behaviour arises due to surface Plasmon resonance (SPR), which originates from coherent oscillations of electrons in the conduction band of nanoparticles induced by the electromagnetic field. The SPR phenomenon arises when nanoparticles are irradiated with visible light, because of the collective oscillations of the conduction electrons (Anandh *et al.,* 2014).

FTIR of Ag Nanoparticles

FTIR spectroscopy is used to probe the chemical composition of the surface and capping agents for the synthesis of NPs. FTIR analysis of synthesized Ag NPs using the aqueous extract of *Coriandrum sativum* is shown in Fig. 3. The synthesized Ag NPs showed the presence of bands due to O-H free bond (3259 cm^{-1}), aldehyde C-H stretching (2953 $cm^{-1}$and 2920 cm^{-1}). The peak at 1587 cm^{-1} corresponded to amide I, arising due to carbonyl stretch in proteins. The peak at 1041 cm^{-1} corresponded to C-N vibrations of the amine. Hence, it proves that Ag NPs have been synthesized with the aqueous extract of *Coriandrum sativum* involved in the biological reduction of the $AgNO_3$. Similar results were reported by Karthika *et al.*, (2015) and Tharanya *et al.,* (2015).

X-ray Diffraction Analysis

X-ray diffraction (XRD) is one of the most extensively used techniques for the characterization of NPs. Typically, XRD provides information regarding the crystalline structure, nature of the phase, lattice parameters, and crystalline grain size (Mourdikoudis *et al.,* 2018). The crystal structure of the Ag NPs was analyzed by an X-ray diffractometer. The formation of silver nanoparticles synthesized using the aqueous extract of *Coriandrum sativum* was supported by X-ray diffraction measurements.

X-ray diffractogram of the synthesized Ag NPs showed distinct diffraction peaks at 27.42°, 31.75°, 45.50°, 53.91°, 58.49°, 60.20°, 75.33° and 76.61° indexed to the planes 110, 111, 211, and 220 (Fig. 4).

SEM images of Ag NPs

The formation of Ag NPs, as well as their morphological dimensions in the SEM study, demonstrated that the average size was from 77.5-82.9 nm with interparticle distance, whereas the shapes were uniformed spherical and ellipsoidal (Fig. 5). The average size of biogenic nanoparticles synthesized using *Nyctanthes arbortristis* leaf extract was found to be between 100-150 nm, whereas the shapes were uniformly spherical (Sundarajan and Gowri, 2011). Similarly, the average size of biogenic nanoparticles synthesized using the bacterial culture supernatant of *Chromohalobacter* sp. was found to be between 96.8 -163.3 (Tharanya *et al.,* 2015).

Antibacterial activity of Silver Nanoparticles

Antibacterial activity of biogenic silver Nanoparticles, various extracts of *Coriandrum sativum* (Aqueous, Hexane, and Ethanol), and Ampicillin were comparatively investigated (Fig. 6-9). Among the above-mentioned, biogenic Ag NPs exerted maximum antagonistic activity against the tested bacterial strains, this was followed by the aqueous extract.

Fig.1 Synthesis of silver nanoparticles of *Coriandrum sativum*

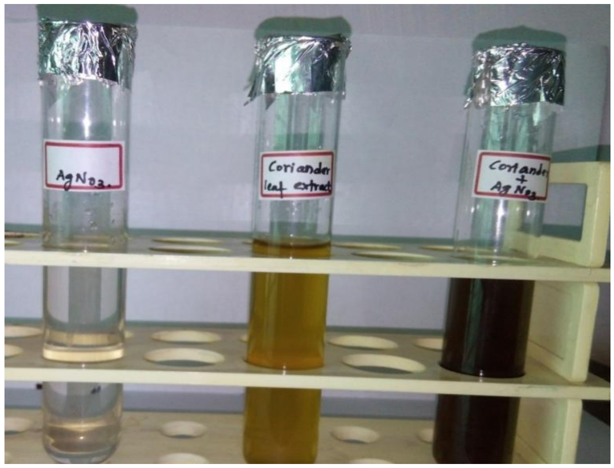


Fig.2 UV – Vis absorption spectrum of Ag NPs

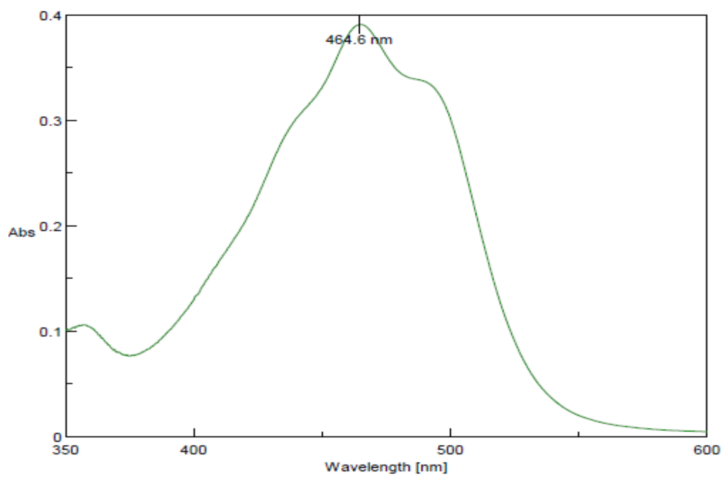


Fig.3 FTIR spectra of biosynthesized Ag NPs

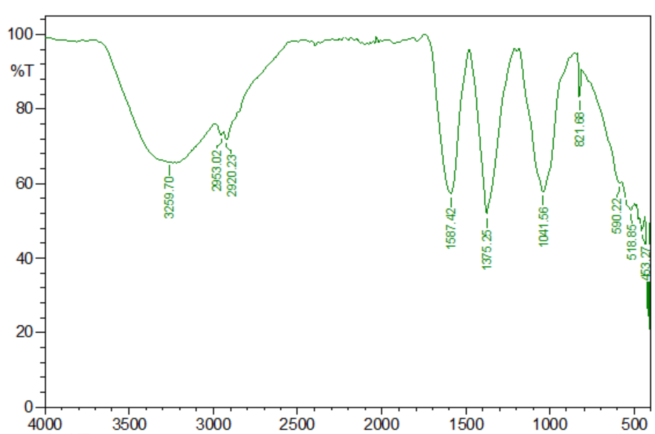


Fig.4 XRD pattern of Ag NPs

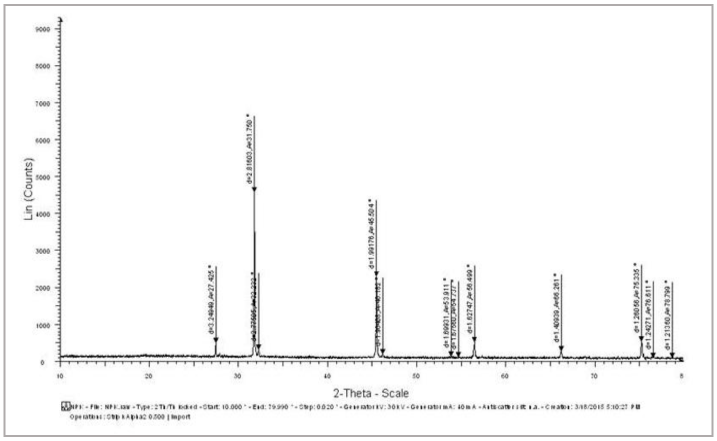


Fig.5 SEM analysis of Ag NPs

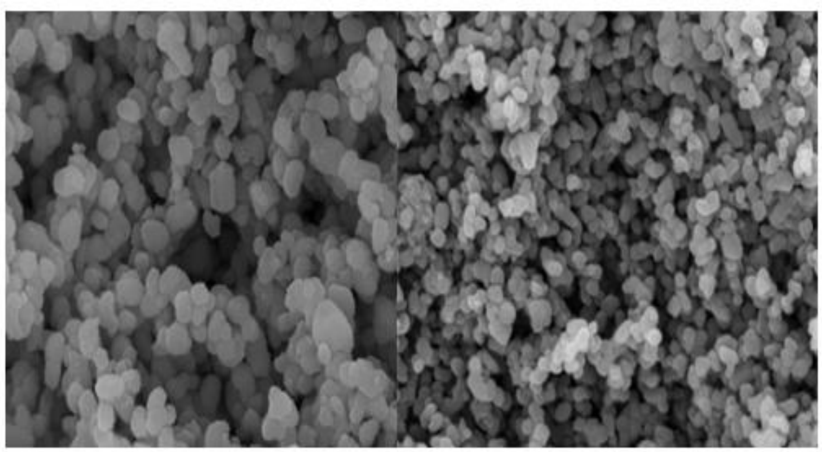


Fig.6 Antibacterial activity of Ag NP against *Pseudomonas* sp.

Fig.7 Antibacterial activity of Ag NP against *Proteus* sp.

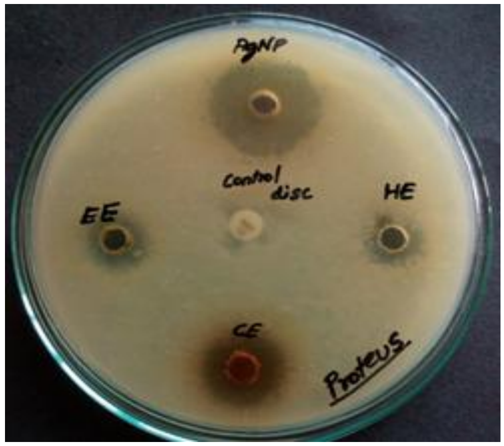


Fig.8 Antibacterial activity of Ag NP against *Salmonella* sp.

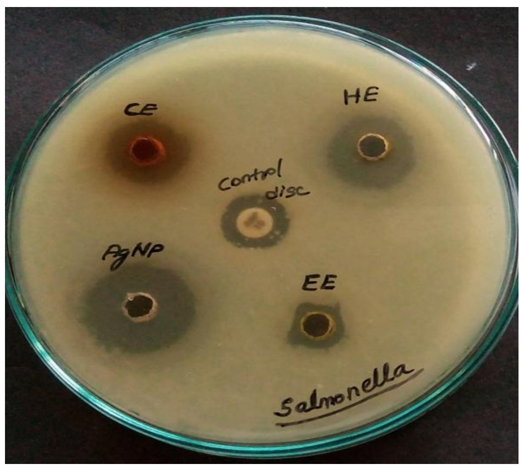
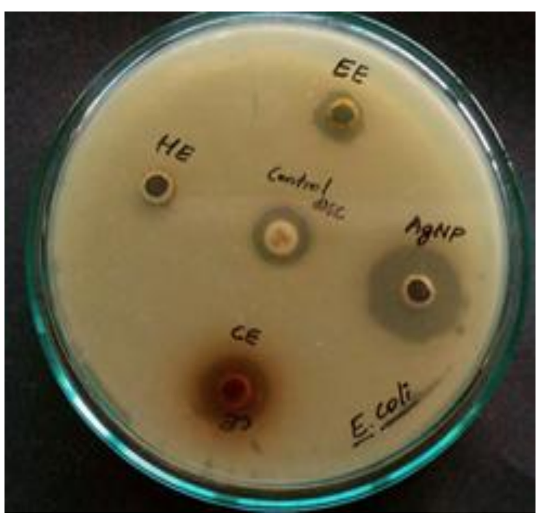


Fig.9 Antibacterial activity of Ag NP against *E.coli*



Biogenic silver nanoparticles exhibited significant activity against *Pseudomonas aeruginosa* (25 mm), *Shigella* sp. (22 mm), and *Proteus* sp. (22 mm). However, the antibacterial activity of hexane and Ethanolic extract of *Coriandrum sativum* was comparatively less. Antagonistic activity of the control (Ampicillin) was found to be very less than the plant extracts and silver nanoparticles (Table 1). According to Berger *et al.,* (1996), Silver ions exhibits strong inhibitory and bactericidal effects as well as possess a broad spectrum of antimicrobial activities It causes the efflux of K+ ions from bacterial cells; thus, the cytoplasmic membrane, which contains many important enzymes and DNA, becomes a significant target site of silver ions (Kim *et al.,* 2011). Owing to their smaller size, Ag-NPs can easily reach the nuclear content of bacteria and they present a large and impressive surface area. The antibacterial activity of silver nanoparticles synthesized by chemical means showed equal antibacterial activity against *S. typhimurium* (Gram -ve) and *S. aureus* (Gram +ve) (Shameli *et al.,* 2012). Ag NPs synthesized using prodigios in exerted maximum antibacterial activity against *E.coli* and *Pseudomonas* sp. (Karthika *et al.,* 2015).

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