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BIOGENIC SYNTHESIS OF SILVER NANOPARTICLES FROM THE LEAF EXTRACT OF *SYZYGIUM CUMINI* (L.)AND ITS ANTIBACTERIAL ACTIVITY

RAM PRASAD[1]*,V. SATYANARAYANA SWAMY[1], KUMAR SURANJIT PRASAD[2] AND AJIT VARMA[1]

[1]*Amity Institute of Microbial Technology, Amity University Uttar Pradesh, Sector-125, Noida-201303, UP, India*
[2]*Department of EnvirEnvier. Microbiology & Biotechnology, Ashok and Rita Patel Institute of Integrated Study and Research in Biotechnology and Allied Sciences (ARIBAS), Anand, Gujarat, India*

ABSTRACT

The antimicrobial activity of silver nanoparticles with the clinically important microbes is a major area of research in the field of nanobiotechnology. *Syzgium cumini* (L.) is used as medicinal plant for anti-diabetic property. The leaves and bark are used for controlling blood pressure and gingivitis. In this present study, we have synthesized silver nanoparticles using *Syzygium cumini* leaf extract. The leaf extract of *S. cumini* is used as capping and reducing agent in the synthesis of silver nanoparticles. The formation of nanoparticles was confirmed by UV-Vis spectrophotometer, Atomic Force Microscopy (AFM) and Scanning Electron Microscopy (SEM). Further antimicrobial activity of silver nanoparticles was studied against *Escherichia coli* MTCC 1302, *Staphylococcus aureus* MTCC 740, *Pseudomonas aeruginosa* MTCC 2295, *and Bacillus licheniformis* MTCC 9555. Surface Plasmon resonance of silver nanoparticles was recorded at 400 nm by UV-Vis spectrophotometer. The synthesized nanoparticles are spherical and having size ranging from 100- 160 nm.

KEY WORDS :***Syzygium cumini,*** **Silver nanoparticles, Antimicrobial activity, Atomic force microscopy, Scanning electron microscopy, leaf extract.**

RAM PRASAD
Amity Institute of Microbial Technology, Amity University Uttar Pradesh, Sector-125, Noida-201303, UP, India

***Corresponding author**

INTRODUCTION

Nanotechnology is the precise and most advanced field of study in the modern material sciences[1]. Its major application in the field of different applied sciences including biology, physics, and chemistry creates a major interest of research. Nanoparticles exhibit completely new or improved properties based on specific characteristics such as size, distribution and morphology2. Chemical and physical synthesis of nanoparticles production leads to the synthesis of environmentally toxic byproducts but production of silver nanoparticles from the green extracts including bark, leaf, root, flower, fruit extracts of plants produces environmentally non-toxic and ecofriendly nanoparticles with effective stability [2,3,4]. Biological syntheses of nanoparticles slower enzyme kinetics for catalytic activity of plants extracts and also offer better manipulation, control over the crystal growth and stability[5]. Nanoparticles had wide range of application in different fields of sciences including medicine[10], pharmacology, electronics, cosmetics [6,7], Drug delivery systems[8],Biosensors[9], interaction with Biomolecules, Cancer therauptics[10]. Silver nanoparticles shows a wide range of antimicrobial activity at minimal concentrations, which has the potential application as anti-infection and anti-inflammatory medicines for the burning skins treatments and it has major application in the coating of the medical devices[10,11,12].

MATERIALS AND METHODS

Preparation of plant extract

Syzgium cumini leaves were collected from tropical forest, Kurnool district, Andhra Pradesh, India. These leaves were dried in hot air oven at 60 ^{0}C overnight. The dried leaves were grinded well with the help of mortar and pestle and mixed with measured volume of double distilled water. This solution was boiled in the water bath at 60^{0}C to 80^{0}C for one hour and after cooling at room temperature, the solution was filtered through Whatman filter paper no.1. Filtrate was collected and was stored at 4 °C for further analysis.

Synthesis of silver nanoparticles

$AgNO_3$ was purchased from Qualigens fine chemicals, Mumbai, India. 10^{-3} M Silver nitrate solution was prepared and stored in brown bottles. 10 mL of leaf extract was added to 90 mL of 10^{-3} M $AgNO_3$ solution for bioreduction process at 30 °C. The color change of the leaf extract from pale yellow to dark brown indicated that the silver nanoparticles synthesis.

Characterization of nanoparticles by UV-Visible spectroscopy

The resonance plasmon of the silver nanoparticles was studied by the UV-Visible spectroscopy in the wavelength range of 200 to 800 nm. The samples were prepared by the dilution in small aliquots, left for 5 hours and monitored through the above wavelength range by UV-160A (Shimadzu Corporation Kyoto, Japan) double-beam spectrophotometer.

Atomic Force Microscopy

Atomic force microscopy was used to identify the size, shape and dispersion of the samples in the range of nanometers. The samples were prepared on the mica based slides and viewed under Model NT-MDA Solver.

Scanning Electron Microscopy

The samples were prepared by sonicating the liquid sample solutions for 15 min at 4^{0}C, and were coated on the surface of glass slides with gold-platinum interface. The samples were viewed under SEM ZEISS EVO®HD.

Antimicrobial activity of Silver nanoparticles

The potential applications of silver nanoparticles were tested for antibacterial activity. The antibacterial activity of silver nanoparticles was studied by disc diffusion method and inhibition zone measurement against *Escherichia coli* MTCC 1302*, Staphylococcus aureus* MTCC

740, *Pseudomonas aeruginosa* MTCC 2295, and *Bacillus licheniformis* MTCC 9555. Starting from lower concentrations including 2, 5, 10 and 15 µL to higher concentrations including 25, 50, 75 and 100µL with volume made up to 100 µL with water wherever needed. The inhibition zone gradually increased with increasing amounts of nanoparticles for the *E. coli, S. aureus,*and *P. aeruginosa*

RESULTS AND DISCUSSION

Initially the nanoparticles synthesis could be observed by the change in the color of the solution from pale yellow to dark brown and the intensity of color increased with the time of incubation (Fig. 1). It took eighteen hours at the room temperature for the color change along with the acidification of the solution from 6.6 to 5.2.

Figure 1
Change in the color of the solution from pale yellow to dark brown

The characterization of the silver nanoparticles was done by the Plasmon resonance with the spectrum obtained in the range of 200 to 800 nm. The maximum absorbance was observed at 400 nm, a characteristic wavelength indicating the presence of silver nanoparticles (Fig. 2).

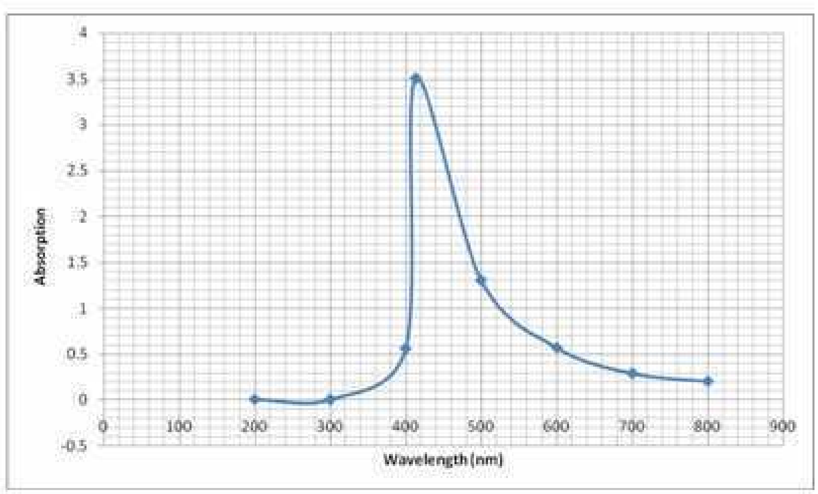


Figure 2
UV-Visible spectrum of silver nanoparticles from Syzgium cumini leaf extract

The dimensions of the nanoparticles including the size, shape, dispersion was observed by the atomic force microscopy. The AFM images showed the presence of bright spots on the surface indicating presence of the nanoparticles and the inverse showed the dark spots indicating size and dispersion of the nanoparticles (Fig. 3 a,b). The 3D image showed the sharp peaks with lower to higher sizes of the nanoparticles and the histogram of the AFM provides the basic information of the average size ranging from 100-130 nm (Fig. 3 c,d).

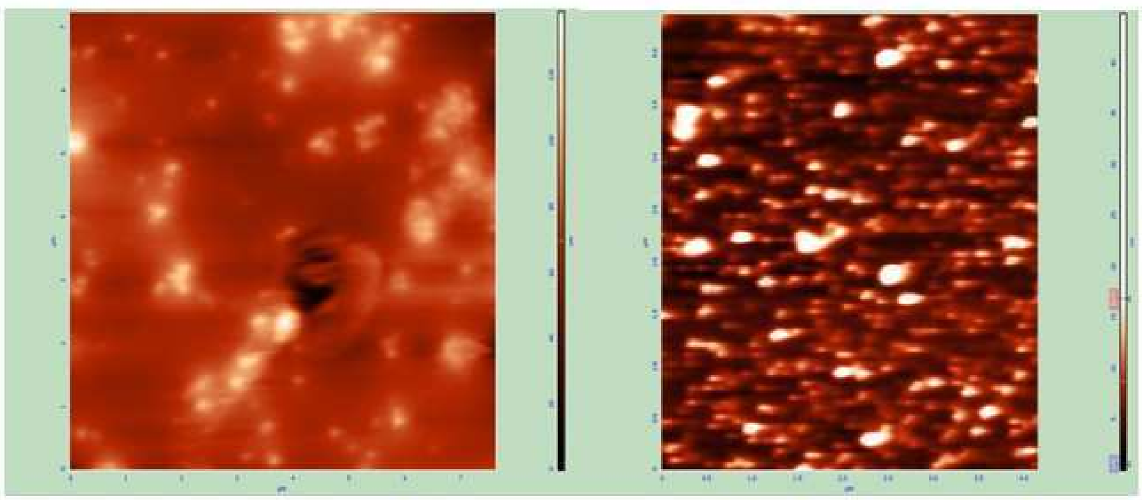


Figure 3A, B.
Direct and Inverse images of silver nanoparticles from AFM

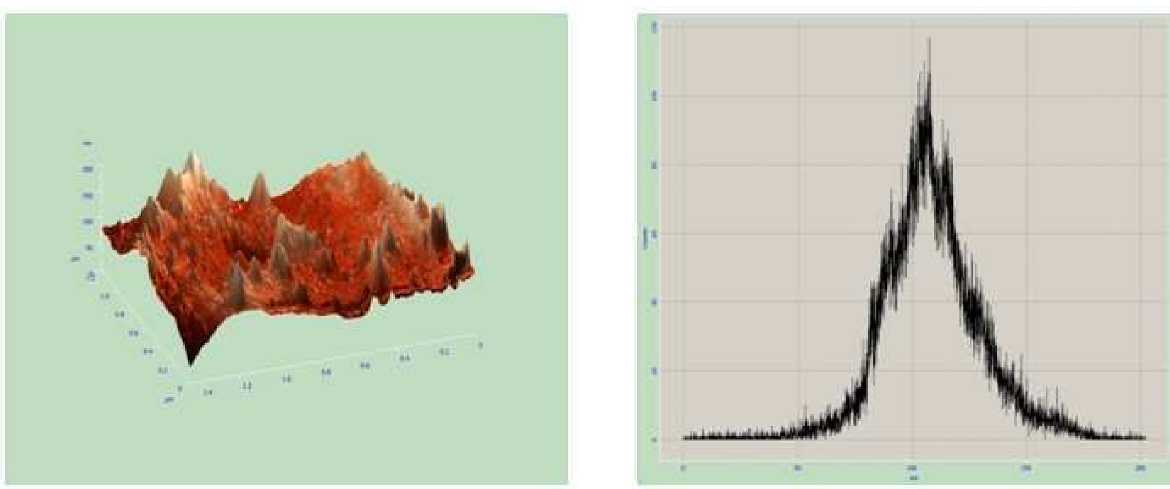


Figure 3C, D
3 D and Histogram images of silver nanoparticles from AFM

The Fig. 4 indicated scanning electron microscopic images showing the shape and the size of the nanoparticles. The surface images showed the small dots ranging from 100 to 160 nm. The nanoparticles were observed to be polydispersed with single and cluster forms.

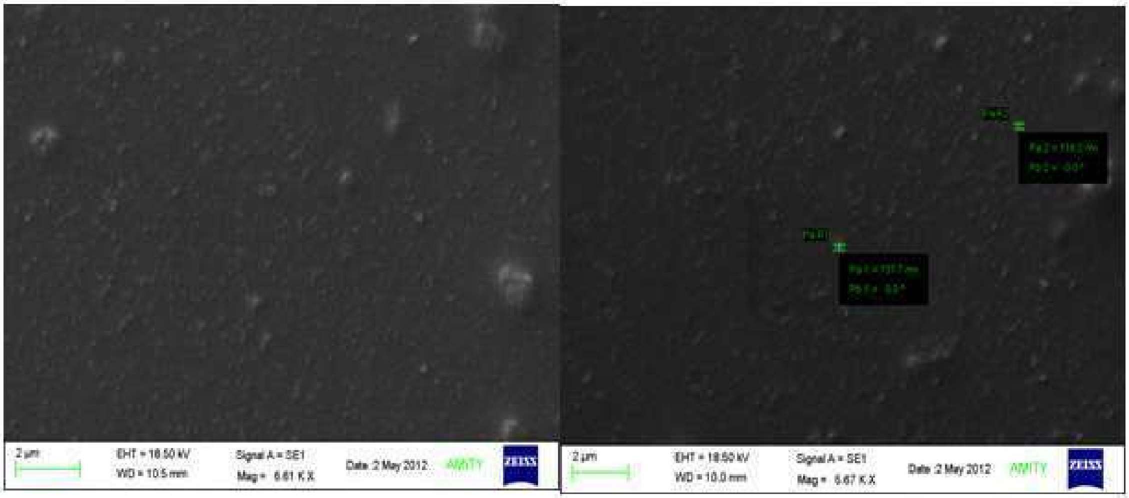
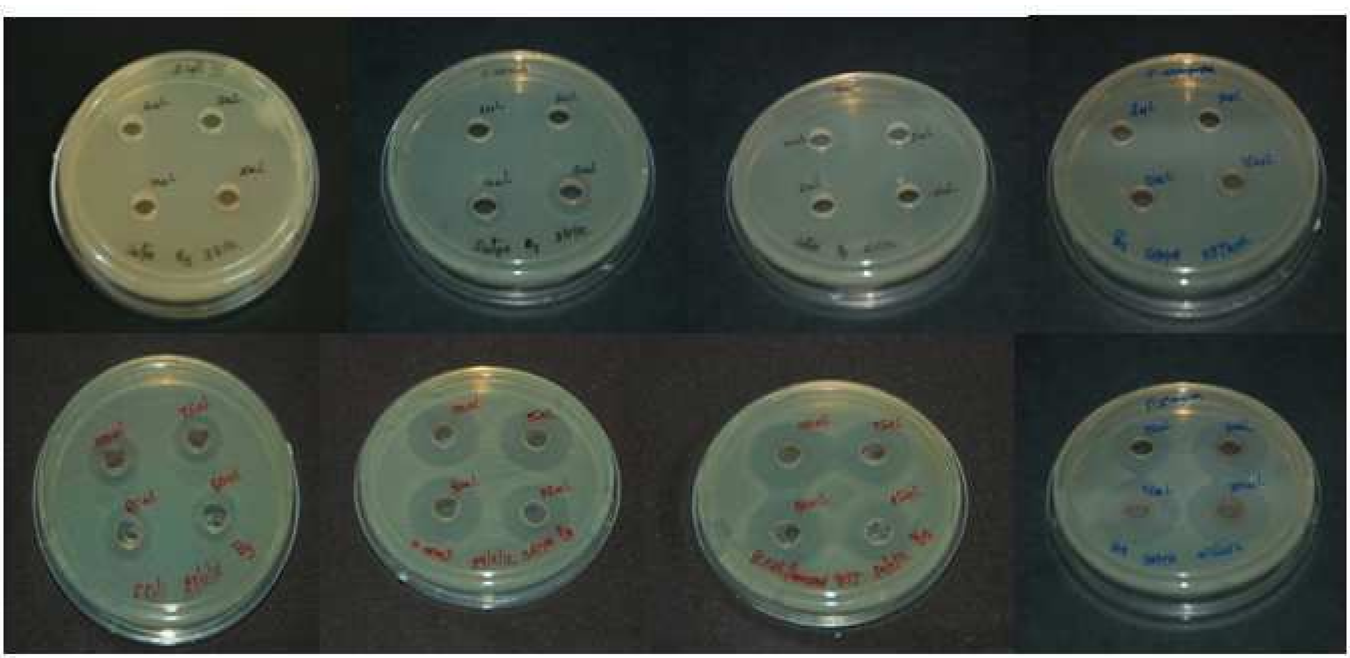
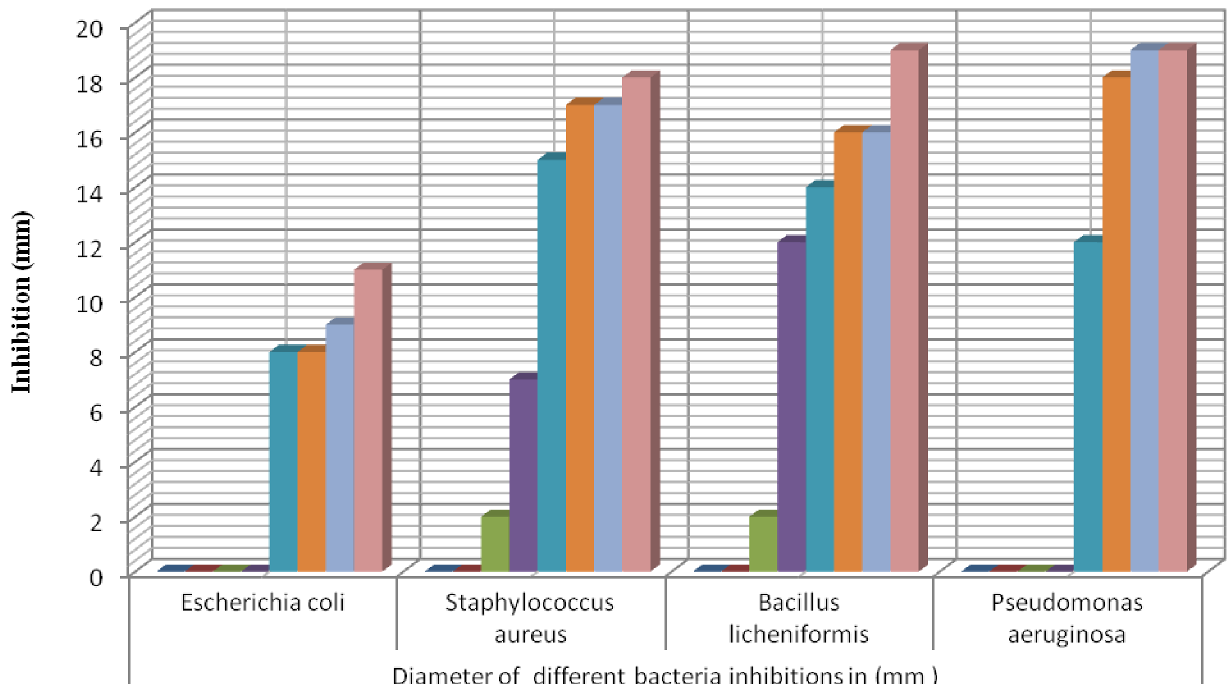


Figure 4
Scanning electron microscopic images of nanoparticles

The silver nanoparticles, synthesized from the plant extract showed a wide range of antimicrobial activity with different concentrations on the different pathogenic bacteria. The inhibition of the bacteria was observed with high value of inhibitions zones at higher concentrations and the diameter of inhibition zone was very low or sometimes negligible due to minimal inhibition at lower concentrations of silver nanoparticles [13, 14, 15, 16]. The images from (Fig. 5) showed different bacteria with different concentrations of nanoparticles and their inhibitions.

Figure 5.
Anti-microbial activity of nanoparticles from Syzygium cumini leaf extract with different concentrations ranging from 2, 5, 10, 15, 25, 50, 75 and 100 µL





	Escherichia coli	Staphylococcus aureus	Bacillus licheniformis	Pseudomonas aeruginosa
2 µL	0	0	0	0
5 µL	0	0	0	0
10 µL	0	2	2	0
15 µL	0	7	12	0
25 µL	8	15	14	12
50 µL	8	17	16	18
75 µL	9	17	16	19
100 µL	11	18	19	19

Concentration of nanoparticles (µL)

Figure 6

Anti-microbial activity of nanoparticles from Syzygium cumini leaf extract with different concentrations ranging from 2, 5, 10, 15, 25, 50, 75 and 100 µL

CONCLUSION

The biogenic silver nanoparticles from the *Syzygium cumini* showed plasmon resonance at 400nm. The size of the nanoparticles ranges from 100 to 160 nm and they are poly dispersed free and clustered forms. These synthesized nanoparticles showed an effective antimicrobial activity against the pathogenic bacterial species. Nanoparticles had profound variety of applications, but still there is dormancy about the actual mechanism of the nanoparticles synthesis from the biological mode of synthesis.

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