GREEN SYNTHESIS OF SILVER NANOPARTICLES FROM THE LEAF EXTRACT OF *SANTALUM ALBUM* AND ITS ANTIMICROBIAL ACTIVITY

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Silver nanoparticles are most efficient antimicrobial agents used in the modern medical implications and medicines. In recent years nanoparticles are synthesized through different modes- chemical, physical and biological. Biological modes of nanoparticles synthesis are the most effective and non-toxic. In this paper we have elaborated our research work on the green synthesis of nanoparticles from the leaf extract of *Santalum album* a very effective plant used in Ayurveda to treat diabetes. The synthesized nanoparticles were characterized by UV-visible spectroscopy, atomic force microscopy, scanning electron microscopy. Antimicrobial activity of biosynthesized Silver nanoparticles was evaluated by means of inhibition zone analysis through well diffusion method.

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Keywords: Nanoparticles, Antimicrobial activity, *Santalum album*, atomic force microscopy (AFM), scanning electron microscopy (SEM).

1. Introduction

Synthesis of silver nanoparticles from the green suspensions such as leaf, root, bark and other plant parts green extracts is the most significant method and it is an non- toxic to the environment when compared to the conventional method of silver nanoparticles synthesis. Nanotechnology is the most active developing research area in advanced material sciences [1]. Nanotechnology is concerned with the synthesis of different types of silver nanoparticles with various sizes, shapes and application of these nanoparticles having potential use in the human welfare [2]. Nanotechnology with a considerable domain of physics, chemistry, electrical and material science has been a considerable progress in the visualization, construction and manipulation of materials at nano scale [3].

From the pre historic period silver has a major role as an antimicrobial agent with profound application in the medicine and medical appliances. These silver nanoparticles are lethal to almost all types of bacteria, virus and some types of eukaryotes. Nanoparticles don't show any side effect and toxicity when they are accumulated by human beings at low concentration [4, 5]. Nanoparticles have a wide range of application not only in the field of medicine but also precise application in the field of textile industries, house hold appliances, food storage containers etc., [6]. Nanocrystalline silver particles have tremendous application in the high sensitivity Bio-molecular detection, therauptics and diagnostics [7, 8, 9, 10].

Green synthesis of silver nanoparticles is the most effective and non toxic method to the environment. The slow kinetics and stabilization of the reaction of the silver salts reduction and it is the better method for manipulation and control over crystal growth and stabilization [11].

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2. Materials and methods

2.1 Preparation of plant extract

Fresh leaves were collected from the plant and there are kept in the hot air oven for one hour to completely dry then leaves were weighed 25gms and made into fine powder by using motor and pestle.This powder was dissolved into 100 mL of double distilled water and boiled at 60 ^{O}C for one hour and this mixture is filtered by using vaccum filter and the extract was collected into the conical flask and kept at 4 ^{O}C.

2.2 Synthesis of silver nanoparticles from the plant extract

To synthesize the silver nanoparticles the substrate of 1mM silver nitrate solution was prepared freshly. 90mL of 1mM silver nitrate solution was taken in a conical flask to this add 10mL of the silver nitrate solution Then the solution was incubated at room temperature.

2.3 UV-Visible spectroscopy

Characterization of the nanoparticles is preliminarily monitored by the UV-Visible spectroscopy .The solution containing the silver nanoparticles was measured at 200 to 800 nm to get the spectrum. The UV-Visible spectrophotometer here we used is LABINDIA UV 3000+.

2.4 Atomic force microscopy

AFM is an advanced method of characterization of silver nanoparticles. For this the sample was prepared by taking the sample solution about 1µL in an eppendroff tube and kept in a sonicator at room temperature about 15 minutes. After this the sample was taken out from the sonicator, one drop of the sample solution was dispersed on a mica based substrate into a thin layer and dried and it was viewed using AFM (NT-MDT).

2.5 Scanning electron microscopy

The surface, shape and size of the nanoparticles can be viewed certainly by using the advanced microscopic technique called SEM. The sample is prepared for this microscopy was by sonication the sample solution for about 15 min at the room temperature. The sample solution is dispersed on a clean glass slide. Then this coated by gold-titanium screen. Then the sample is viewed under the SEM ZEISS EVO®HD.

2.6 Antibacterial activity

Silver nanoparticles have a specific property to inhibit the growth of almost all the bacteria. This property can be monitored by preparing the plates of nutrient agar. Then the cultures of bacteria including the *Escherichia coli, staphylococcus aureus, Pseudomonas aeruginosa, Azotobacter chroococcum, Bacillus licheniformis* 9555, by the well plates method at different concentrations such as at lower concentrations including 2, 5,10 and 15 µL and at higher concentrations including 25,50,75 and 100 µL are used for the antimicrobial activity of the above bacterial species.

3. Results and discussion

The synthesis of silver nanoparticles can be observed by the change in the color of the solution from pale yellow color to dark brown color after seven hours of incubation. It was indicated in Fig.1 that the silver nitrate has been reduced into silver nanoparticles during the reaction of the plant extract. The dark color of the solution indicates the reduction of the silver nitrate solution.

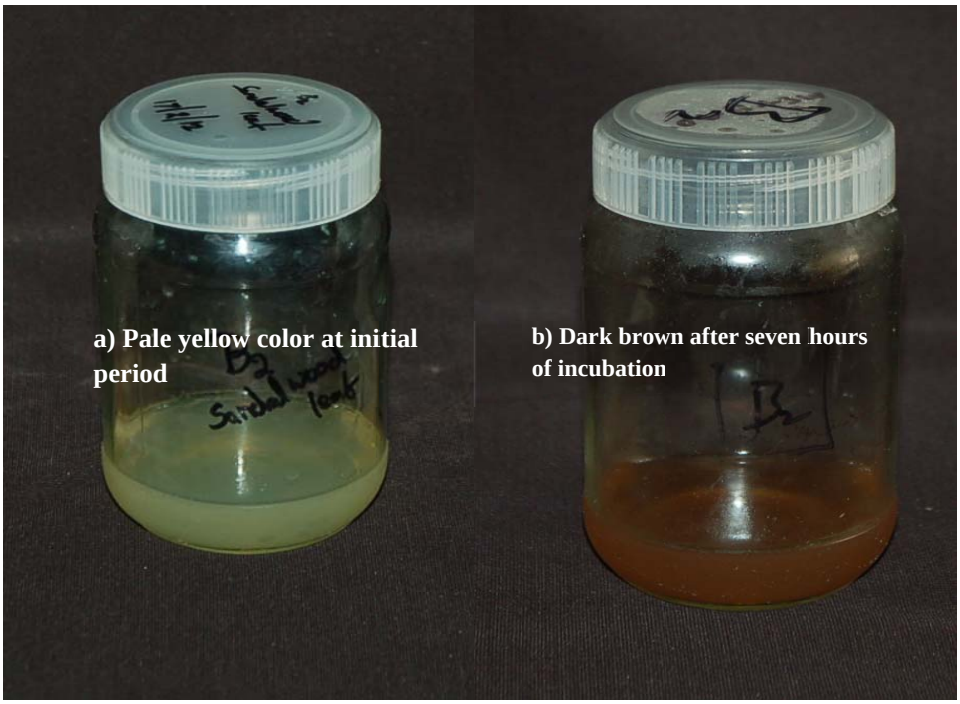

Fig. 1 Change in the color of the solution from pale yellow to dark brown color.

The pH of the solution has changed from the initial pH of 6.59 to the final pH of 7.10. The change of the pH towards alkaline condition of the solution favors the synthesis of silver nanoparticles.

UV-Visible spectrograph (Fig.2) indicates that at 423 nm, a maximum absorption has taken place which indicates the presence of colloidal silver components; the graph has taken the percentage of absorptions at different wavelengths as a function of time, as the reaction time increases the percentage of the absorption along with in the meantime.

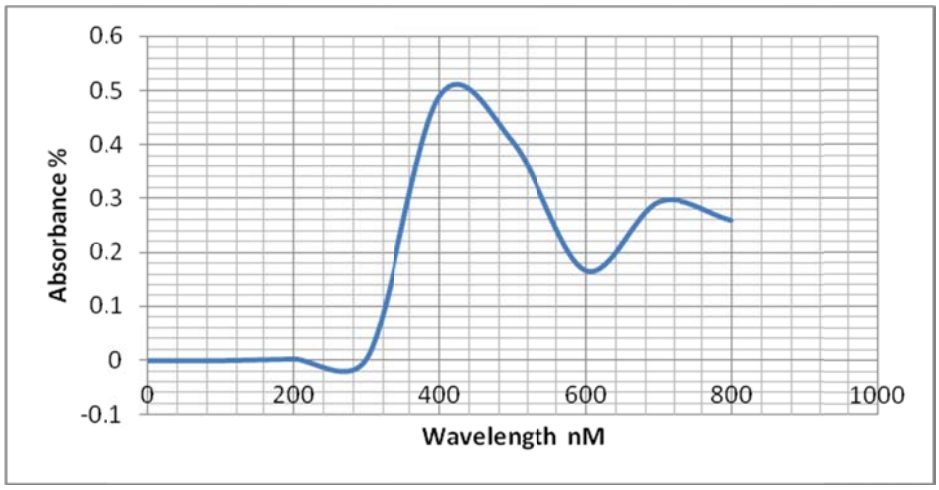

Fig. 2 UV-Visible spectrum of silver nanoparticles solution of Santalum album.

The surface of the nanoparticles and their sizes are measured by using Atomic Force Microscopy. Fig. 3 shows the dark and white spots which are the nanoparticles on the mica substrate. The thin layer of the sample on the mica substrate shows the nanoparticles of maximum

average size about 100 to 140 nm. This is estimated by the histogram (Fig. 3) obtained during the assay. The 3D image shows different sizes of nanoparticles with sharp peaks.
In (Fig. 4) are the SEM images of nano particles of different sizes ranging from 80nm to 200 nm which are dispersed in the form of different aggregates and particles. The nanoparticles were not in direct contact even within the aggregates, indicating stabilization of the nanoparticles by capping agents.

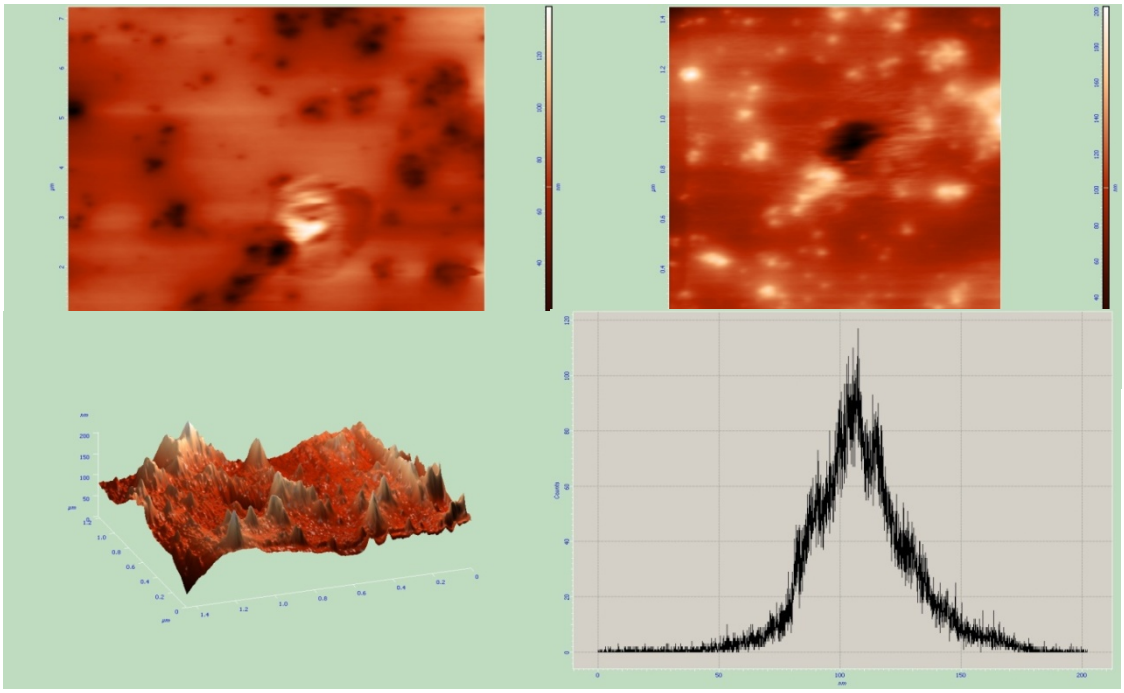

Fig.3 Atomic Force Microscopic images including inverse image, direct image, 3D image and Histogram

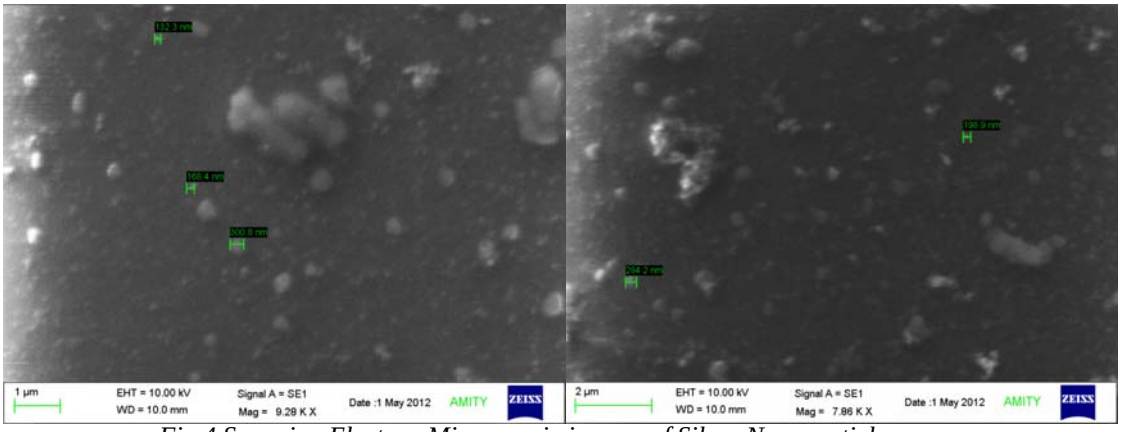

Fig.4 Scanning Electron Microscopic images of Silver Nanoparticles

Anti-microbial activity

Silver nanoparticles are the most effective inhibitors of the bacterial growth by binding on the surface of the bacteria which causes high PMF on the outside of the cell wall [12]. The inhibition of the bacteria were observed at very high diameter of inhibitions at higher concentrations and the diameter of inhibition is very low or sometimes negligible due to minimal inhibition of concentrations of silver nanoparticles [13,14,15,16]. The images from (Fig.5) show different bacteria with different concentrations of nanoparticles and their inhibitions.

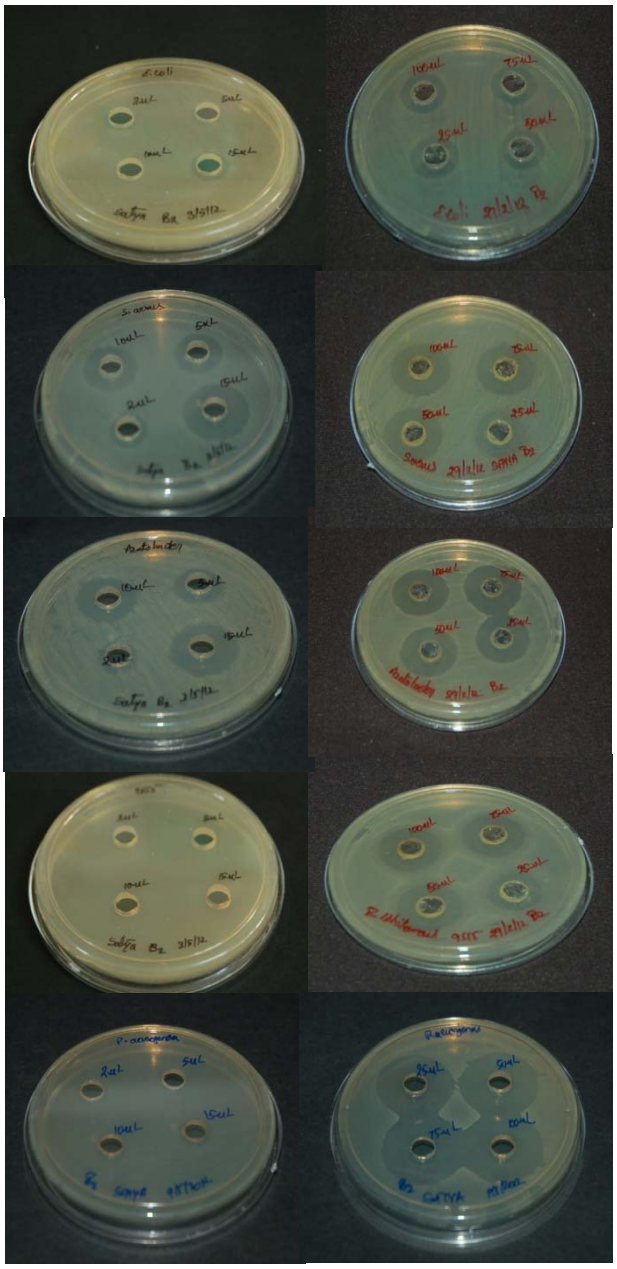

Fig. 5 Antimicrobial activity of the nano particles at different concentrations ranging from 2 µL, 5 µL, 10 µL, 15µL ,25 µL, 50 µL, 75 µL, 100µL with the pathogenic and soil born bacteria Escherichia coli, Staphylococcus aureus, Azotobacter chroococcum, Bacillus licheniformis, and Pseudomonas aeruginosa

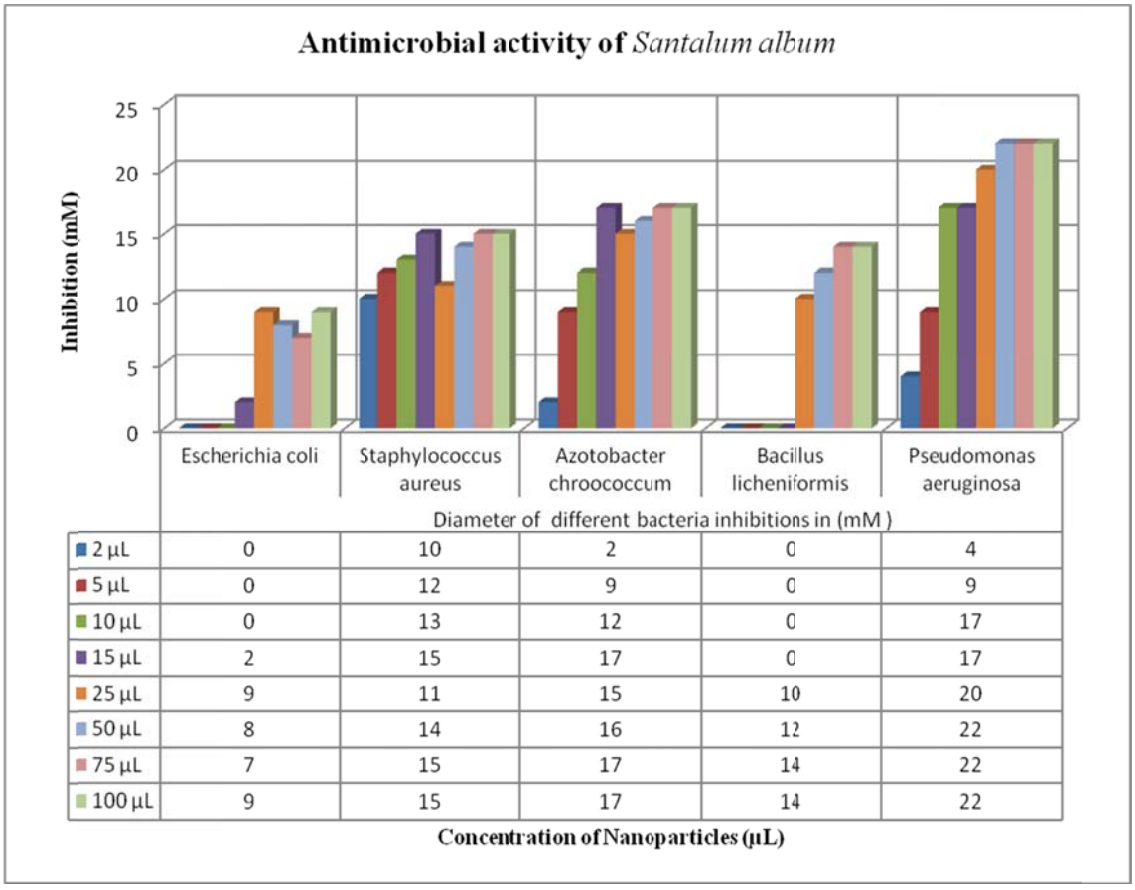

Fig .6 Antimicrobial activity of the nano particles at different concentrations ranging from 2 µL, 5 µL, 10 µL, 15µL ,25 µL, 50 µL, 75 µL, 100µL with the pathogenic and soil born bacteria Escherichia coli, Staphylococcus aureus, Azotobacter chroococcum, Bacillus licheniformis and Pseudomonas aeruginosa

4. Conclusion

Silver nanoparticles have profound major applications in the medical, electrical and other major fields. In the present study we made a contribution towards the synthesis of silver nanoparticles, which are characterized by different techniques which provide the size, morphology and dispersity of nanoparticles. The synthesized nanoparticles are spherical, polydispersed and having a size ranges from 80nm to 200nm. This also shows antimicrobial activity at both in lower and higher concentrations, these synthesized nanoparticles are environmentally non-toxic and require very less time and man power. Moreover, the synthesis of nanoparticles with different sizes and shapes is the basic challenging task in the green synthesis of nanoparticles and this requires a basic understanding of the nuclei formation and the influence of reaction species in nuclei morphology [17].

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