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RESEARCH ARTICLE!!!

**PLANT MEDIATED SYNTHESIS OF SILVER NANOPARTICLES AND
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green synthesis,
sadabahar, plantain
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India.**ABSTRACT**

Biological methods of synthesis have paved way for "greener synthesis" of nanoparticles and these have proven to be better methods due to slower kinetics, they offer better manipulation and control over crystal growth and their stabilization. This has motivated an upsurge in research on the synthesis routes that allow better control of shape and size for various nanotechnological applications. In this work, we have explored an inventive contribution for synthesis of silver nanoparticles using *catharanthus roseus* (Sadabahar), *Musa paradisiacal* (Plantain flower) and *Polygonum odoratum* (Coriandium) leaf extract. Synthesized nanoparticles were characterized by various methods such as UV-Vis spectroscopy, SEM and XRD. In addition, antibacterial activity of the synthesized silver nanoparticles was also determined. This new method is rapid time scales for biosynthesis of metallic nanoparticles using environmentally benign natural resources as an alternative to chemical synthesis protocols as reductant for synthesizing silver nanoparticles.

INTRODUCTION:

Green nanoscience/nanotechnology involves the application of green chemistry principles to the design of nanoscale products, the development of nanomaterial production methods, and the application of nanomaterials. The approach aims to develop an understanding of the properties of nanomaterials, including those related to toxicity and ecotoxicity, and to design nanoscale materials that can be incorporated into high performance products that pose little hazard to human health or the environment. It strives to discover synthesis methods that eliminate the need for harmful reagents and enhance the efficiency of these methods, while providing the necessary volume of pure material in an economically viable manner. It also provides proactive design schemes for assuring the nanomaterials produced are inherently safer by assessing the biological and ecological hazards in tandem with design. Finally, it seeks applications of nanoscience that maximize societal benefit while minimizing impact on the ecosystem. In this way, green nanoscience guides materials development, processing, and application design throughout the life cycle, starting with raw material selection through end-of-life [1].

The use of environmentally benign materials like plant extract, bacteria, fungi and enzymes for the synthesis of silver nanoparticles offers numerous benefits of eco-friendliness and compatibility for pharmaceutical and other biomedical applications as they do not use toxic chemicals for the synthesis protocol. Chemical synthesis methods lead to presence of some toxic chemical absorbed on the surface that may have adverse effect in the medical applications. Green synthesis provides advancement over chemical and physical method as it is cost effective, environment friendly, easily scaled up for large scale synthesis and in this method there is no need to use high pressure, energy, temperature and toxic chemicals [2].

Silver nanoparticles (AgNPs), among noble metal nanoparticles, have attracted great interest because of the large number of applications, such as in biolabelling, as intercalation materials for electrical batteries as optical receptors, in nonlinear optics, as a catalyst in chemical reactions and as antibacterial capacities [3]. The medicinal and preservative properties of silver have been known for over 2,000 years. Silver based compounds have been widely used in bactericidal applications, in burn, wound therapy and healthcare product [4]. They have been widely used as disinfectants that inhibit bacteria growth by inhibiting the essential enzymatic functions of the microorganism and attribution to reactive oxygen species mediated bactericidal activity [5]. Currently silver nanoparticles are widely used as antibacterial and antifungal agents in a diverse range of consumer products: air sanitizer

sprays, detergents, soaps, shampoos, toothpastes and washing machine. Nanoparticles exhibit new or improved properties depending upon their size, morphology and distribution [6]. Many techniques of silver nanoparticles synthesis are extremely expensive and also involve the use of toxic, hazardous chemicals, which may pose potential environmental and biological risks. Bioinspired synthesis of nanoparticles provides advancement over chemical and physical methods as it is a cost effective and environment friendly and in this method there is no need to use high pressure, energy, temperature and toxic chemicals [7]. The plant contains a variety of phytochemical compounds such as phenols, amino acids, flavones and these molecules are expected to self assemble and cap the metal nanoparticles formed in their presence and thereby induce some shape control during metal ion reduction [8]. Thus, it is of interest to study the use of plant extracts for synthesis of silver nanoparticles in order to search for the high efficiency natural source for silver nanoparticles preparation. In addition, antibacterial activity of the synthesized silver nanoparticles was also determined and reported.

Materials and Methods

Preparation of the plant extract

Fresh leaves of *catharanthus roseus* (Sadabahar), flowers of *Musa paradisiacal* (Plantain flower) and seeds of *Polygonum odoratum* (Coriandium) are collected from Periyakulam. 25gms of collected green leaves and flowers were thoroughly washed with tap water and then with distilled water, cut into fine pieces, and boiled in 100ml of distilled water, for half an hour. The aqueous extract thus obtained was filtered through whatman No.1 filter paper to obtain a clear extract. The extract was collected in cleaned and dried 100ml beaker. Then, the filtrates were collected and refrigerated for further experiments.

Synthesis of Silver Nanoparticles using Plant extracts

Aqueous solution of silver nitrate (AgNO_3) at concentration of 0.02 mmol/ml was prepared and used for the synthesis of silver nanoparticles. The 10ml of the above prepared plant extract under normal condition was taken in test tube and 2ml of the silver nitrate solution is added. The change of colour takes place within few minutes and the precipitate is formed. The precipitate is separated with the help of whatman No.1 filter paper.

Antibacterial activity

Antibacterial activity was assayed using standard well diffusion method against human pathogenic bacteria *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Agar (NA) was prepared for cultivation of the bacteria. 100 μ l of fresh overnight grown

cultures of the bacteria were spread on Nutrient Agar containing Petri plates. With a sterile borer 1mm holes were punched in the medium. 100 μ l of the solution containing nanoparticles was inoculated in this hole and the plates were incubated at 37⁰C overnight. The next day, zone of inhibition in the bacterial mat was measured [9].

Results and Discussion

Three different plant extracts were used to produce silver nanoparticles, the reduction of silver ions into silver nanoparticles occurred after mixing silver nitrate with different plant and spices extract, followed by colour change of the solutions due to reduction of silver ion, which may be the indication of formation silver nanoparticles. The synthesized silver nanoparticles were characterized by UV-Visible Spectroscopy, XRD and SEM. The important parameters, viz. FWHM (full width half maximum), particle size, structure of compounds, the value of energy gap, presence of the compounds were discussed.

UV-Visible Spectroscopy

The bioreduction of silver in aqueous solutions was monitored by periodic sampling of aliquots of the mixture and subsequently measuring UV-Vis spectra. UV-Vis spectral analysis was done by using ShimadzuUV-1800 double beam spectrophotometer. The absorption peaks are measured in the range of 200-800 nm [10].

UV-Vis spectroscopy analysis showed that the wavelength of silver nanoparticles synthesized using *Musa paradisiacal*, *Polygonum odoratum* and *catharanthus roseus* extract centered at 400-500 nm due to the excitation of surface plasmon vibrations in the silver nanoparticles.

The figures (1, 2, and 3) represent the UV-VIS spectrum of silver nanoparticle

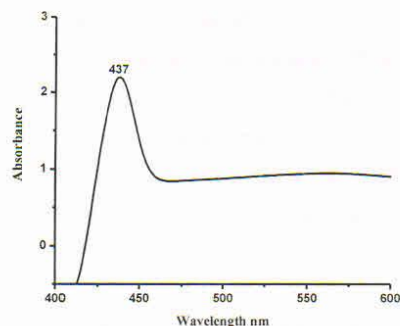


Figure 1. UV-VIS Spectrum of silver nanoparticles from *Musa paradisiacal*

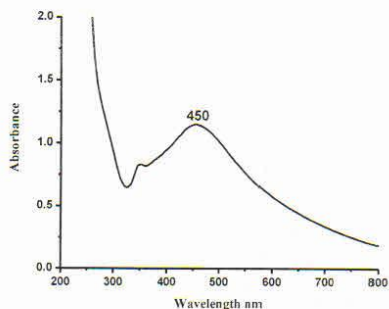


Figure 2. UV-VIS Spectrum of silver nanoparticles from *Polygonum odoratum*

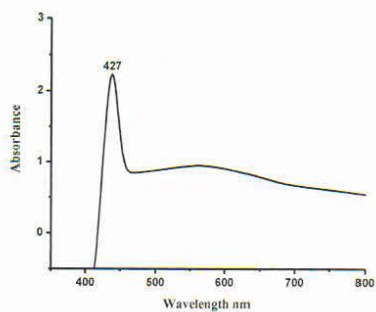


Figure 3. UV-VIS Spectrum of silver nanoparticles from *catharanthus roseus*

X-Ray diffraction studies

The silver nanoparticle solution obtained was purified by repeated centrifugation at 5000 rpm for 20 minutes followed by redispersion of the pellet of silver nanoparticles into 1ml of deionized water. After freeze drying of the purified silver nanoparticles, the structure and composition were analyzed by XRD. The crystallite domain size was calculated from the width of the XRD peaks, assuming that they are free from non-uniform strains, using the Scherer formula.

$$D = 0.94 \lambda / \beta \cos \theta$$

Figures (4, 5 and 6) show the XRD spectrum for silver nanoparticles [11]. The size of the silver nanoparticles thus estimated was found to be 8.5052nm for *Musa paradisiacal*, 8.5707nm for *Polygonum odoratum* and 3.6881 nm for *catharanthus roseus*.

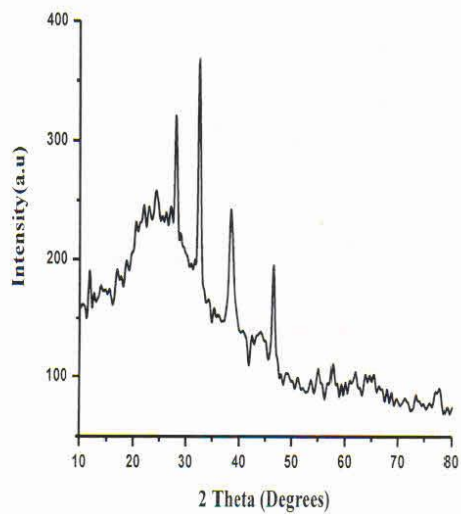


Figure 4. XRD Pattern of silver nanoparticles form *Musa paradisiacal*

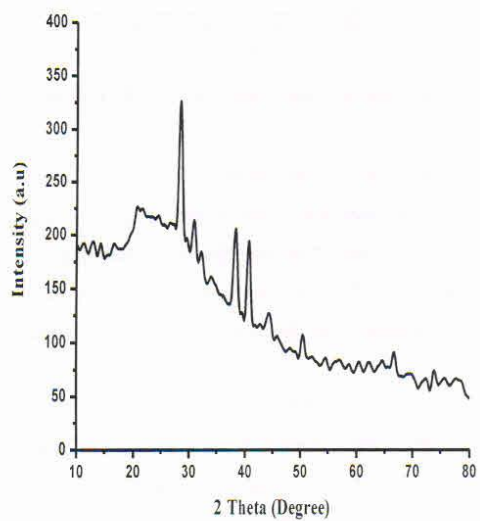


Figure 5. XRD Pattern of silver nanoparticles form *Polygonum odoratum*

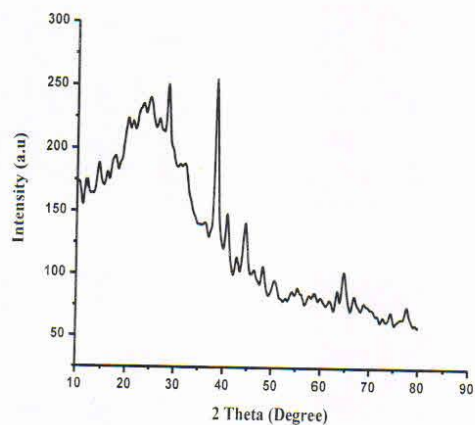


Figure 6. XRD Pattern of silver nanoparticles from *Catharanthus roseus*

Scanning Electron Microscopy

Size, shape and distribution of green synthesized silver nanoparticles were characterized by Scanning Electron Microscope. The particle morphology of the silver nanoparticles fabricated using *Musa paradisiacal* plant extract. Insert of Figure. 7 (a, b, c, d) shows the particle morphology of silver nanoparticles at low and high magnification respectively. The particle aggregation was clearly viewed and it indicates the uniform particle dimension of the nanoparticle synthesis process and no action of stabilization of nanoparticles. However, in the higher magnification, some smooth solid blocks are identified which may be due to very close packing of the silver nanoparticles [12].

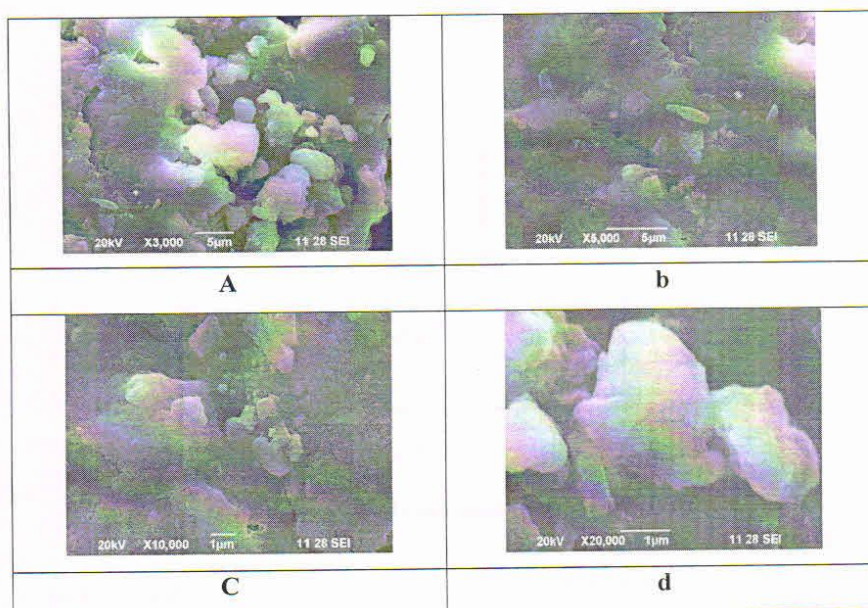


Figure 7 (a, b, c, d) SEM images of silver nanoparticles of *Musa paradisiacal* at low and high magnification

Antibacterial activity

The antimicrobial assay was done on human pathogenic *Escherichia coli*, *Pseudomonas aeruginosa* (Gram negative) and *Staphylococcus aureus* (Gram positive) by standard agar disc diffusion method. The pure cultures of bacteria were sub-cultured on nutrient agar medium. Each strain was swabbed uniformly onto the individual plates using sterile cotton swabs. Wells of 10 mm diameter was made on nutrient agar plates using gel puncture. Using a micropipette, 50 µl of nanoparticles solution was poured onto each well on all plates. After incubation at 37°C for 24 hours, the different levels of zone of inhibition were measured and it was tabulated in table I.

TABLE 1: ANTIBACTERIAL ACTIVITY FOR BIOSYNTHESIS OF SILVER NANO PARTICLES

Compounds	E.Coli (mm)	S.Aures (mm)	P.Seudomonous
Silver nano particles from <i>catharanthus roseus</i> (Sadabahar),	10	10	10
Silver nano particles from flowers of <i>Musa paradisiacal</i> (Plantain flower)	Resistant	10	14
Silver nano particles from seeds of <i>Polygonum odoratum</i> (Coriandium),	Resistant	12	Resistant
Amikacin (Standard)	18	18	18

Conclusion

A critical need in the field of nanotechnology is the development of reliable and eco-friendly processes for synthesis of metallic nanoparticles. Here, we synthesized a simple biological and low-cost approach for preparation of stable silver nanoparticles by reduction of silver nitrate solution with a bioreduction method using *catharanthus roseus* (Sadabahar), flowers of *Musa paradisiacal* (Plantain flower) and seeds of *Polygonum odoratum* (Coriandium) aqueous extract as the reducing agent. The results confirmed the reduction of silver nitrate to silver nanoparticles with high stability and without any impurity. The UV-visible spectra showed a broad peak located at 400 -500 nm for silver nanoparticles. X-ray diffraction pattern (XRD) revealed that the resultant nanoparticles were nanometric in size and the particle size was found to less than 20 nm. The SEM picture revealed the morphology of the particles. In the future, it would be significant to know the precise mechanism of biosynthesis and to technologically engineer the nanoparticles with the aim of attaining better control shape, over size and whole monodispersivity.

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