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# BIOSYNTHESIS , CHARACTERIZATION & ANTIMICROBIAL ACTIVITY OF SILVER NANOPARTICLES FROM *SARACA ASOCA*

**Merli Maman [1] , Anchana devi C [1*] & Pushpa N [2]**

[1] PG & Research Department of Biotechnology, Women's Christian College, Chennai, India
2 Department of Microbiology, Cauvery College for Women, Trichy
* Corresponding author : dr.anchanababu@gmail.com

## ABSTRACT

Silver nanoparticle was prepared by green synthesis method using the powdered leaves of *Saraca asoca.* The resulted particle was characterised by Fourier Transformed Infrared Spectroscopy (FTIR) and Scanning Electron Microscopy (SEM). The antimicrobial activity of the resulted nanoparticle was tested against various bacterial and fungal species. The organisms used were *Escherichia coli, Staphylococcus sp , Streptococcus sp , Pseudomonas sp, Proteus sp, Bacillus sp, Aspergillus sp and Candida sp.* The size of the particle was proved to be in nano range by characterisation. *Streptococcus sp* (20 mm) among the bacterial species and *Aspergillus sp* (30 mm) and *Candida sp* (30 mm) among the fungal species showed good antimicrobrial activity.

**KEY WORDS:** Silver nanoparticle, *Saraca asoca*, Antimicrobial activity, FTIR, SEM.

## Introduction

*Saraca acoca*, a potential candidate having a significant antimicrobial activity and widely used in the field of ayurvedic medicine. (Pradhan .P *et al.,* 2009). especially, used in treatment of infertility ( *Rozati .R et al.*, 2006). The nanoparticles of silver ranges from 10-100nm and they have very high surface to volume ratio. They have very good solubility,

stability and also excellent optical, electrical and thermal properties. Silver nanoparticle are off different size and shape depending on the method of fabrication. As the size reduces, the area exposed is higher and thus the exposed atoms communicates well with its surroundings, this forms the basis for its excellent antimicriobial activity.

The physical and chemical method of nanoparticle is expensive due to its high power and chemicals consumption and its eco-unfriendly due to the deposition of nanoparticle waste to the environment. Thus, green synthesis of nanoparticles came to light where the bulk particles are reduced to nano size with the help of biological materials like plant or microorganisms. The silver nitrate is heated along with the powdered leaf sample of interest, as the size reduces, the colour of the particle changes from greyish white to dark brown (Banerjee .P *et al.*, 2014). Silver nanoparticle can also be synthesized with bacteria, fungi like yeast and also algae. This type of preparation of nanoparticle does not deposit its waste to the environment and does not utilize any chemicals. Thus it is considered to be the eco-friendly method of preparation of nanoparticles.Silver nanoparticleis characterized by UV spectrophotometer, FT-IR and scanning electron microscope(SEM).

Silver nanoparticle has gained much attention in the medical field for its excellent antimicrobial activity. Also when the medically important plant based green synthesis of silvernanoparticle is performed, the activity seems to be higher than what is expected due to the activeness of both the plant and the nanoparticle *(Tripathi R.M et al.,2014)* This can be studied further for their activity against bacteria and fungi for the treatment of various diseases.

Study area

The area of study is mainly the silver nanoparticle which was prepared by the leaves of *Saraca asoca*. The study differs from the other green synthesis methods of silver nanoparticles from the type of leaves used for the reduction of bulk silver to the nano size. And also the study of antimicrobial activity of the obtained nanoparticle against various organisms.

## Materials and methods

### Collection of leaf samples

*Saraca asoca* leaves were collected from Kollam(Kerala).The leaves were shade dried and powdered well.

### Green synthesisof Silver Nanoparticles

50ml of 1mM silver nitrate was prepared.From the preparation, 20ml was taken in a beaker and heated on a hot plate with magnetic stirrer.1ml of aqueous dissolved *Saraca asoca* leaf powder (1mg/ml) was added to the silver nitrate solution for every 10 minutes in correct interval of time.The leaf powder was added till the colour of the solution changes to brown or dark brown.Once after the colour was obtained, the solution was taken and covered with aluminium foil and stored in dark overnight.The colour of the solution was observed next day and then it was stored in refrigerator.The bottle in which the nanoparticle was stored was fully covered with aluminium foil to prevent the photo activation of silvernanoparticles.(Figure 1) (Banu A and Rathod V . 2013), (Logeshwari P *et al., 2013).*

### Characterization of Synthesized silver nanoparticles

Ultraviolet-visible Spectroscopy (UV-Vis) was performed in Shimadzu UV-VIS Spectrophotometer. The studies of size, morphology and composition of the nanoparticles were performed by means of transmission electron microscopy (SEM) and Fourier transform infrared (FTIR) spectral measurements were carried out to identify the potential biomolecules in *Saroca asoca* leaf extract which is responsible for reducing and capping the bioreduced silver nanoparticles. Samples for SEM studies were prepared by placing drops of the silver nanoparticles solutions on carbon-coated SEM grids. (Okafor F *et al*., 2013)

### Antimicrobial activity of the Green Synthesized Nanoparticle

The authenticated organisms *Escherichia coli, Staphylococcus sp, Streptococcus sp, Pseudomonas sp, Proteus sp, Bacillus sp, Aspergillus sp and Candida sp.* used for testing were obtained from National Committee for Clinical Laboratories Standards (NCCLS), purchased from Institute of Microbial Technology, Chandigarh, India. Overnight cultures were prepared with nutrient broth.

**Preparation of plates and antimicrobial testing**

Muller- Hinton Agar (MHA) for antibacterial testing and SDA for antifungal testing were prepared ,autoclaved and the media was poured onto the petriplates and allowed to solidify, each organism was swabbed onto the plates under sterile conditions.The antimicrobial testing was done in well method where the samples were loaded in the well. The plates were incubated @ 37˚C for 24hr and the zone of inhibition was observed.The diameter of the zone was measured in mm.

## Result and Discussion

The results of the study are presented as tables and figure below

To 1mM silver nitrate solution the aqueous extract of *Saraca asoca* was added to prepare the green synthesis of silver nanoparticles (Figure 1)

**Figure 1:** Image showing the Green synthesis of Silver nanoparticle.


UV-visible spectroscopy is a technique to find the properties and formation of different nanoparticles where, the nanoparticles synthesized are influenced by their shape, size, free electron density , interparticle interactions and medium surrounding it. In the present study, reduction of silver ions present in the aqueous solution of silver nitrate during the reaction with the ingredients of *Saroca asoca* leaf extract has been seen by the UV-Vis spectroscopy

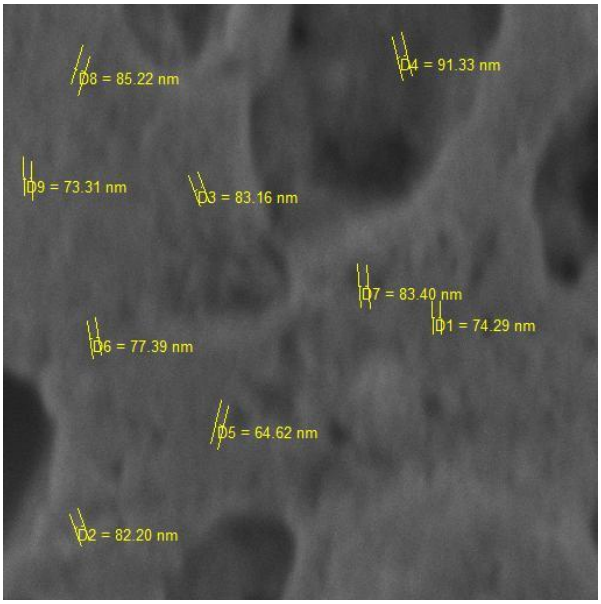
ranging from 300 to 600 nm. The maximum absorption was obtained at 434 nm which corresponds to surface plasmon resonance (SPR) of silver nanoparticles. (Table 1)

Table 1: Table showing the OD values of Nanopreparation.

| Absorption in nm | OD value |
|---|---|
| **60** | **337** |
| **80** | 367 |
| **100** | 385 |
| **120** | 392 |
| **140** | 403 |
| **160** | 434 |

The SEM results reveal that the scale of size of the nanoparticle varies from 70nm to 90nm. Moreover preparation of uniform sized nanoparticle is a challenge in the field of nanotechnology. SEM analysis shows high-density AgNPs synthesized by *Saraca asoca* leaf extract (Figure 2). It was shown that relatively spherical and uniform AgNPs were formed with diameter of 70 to 90 nm. The SEM image of silver nanoparticles was due to interactions of hydrogen bond and electrostatic interactions between the bioorganic capping molecules bound to the AgNPs.

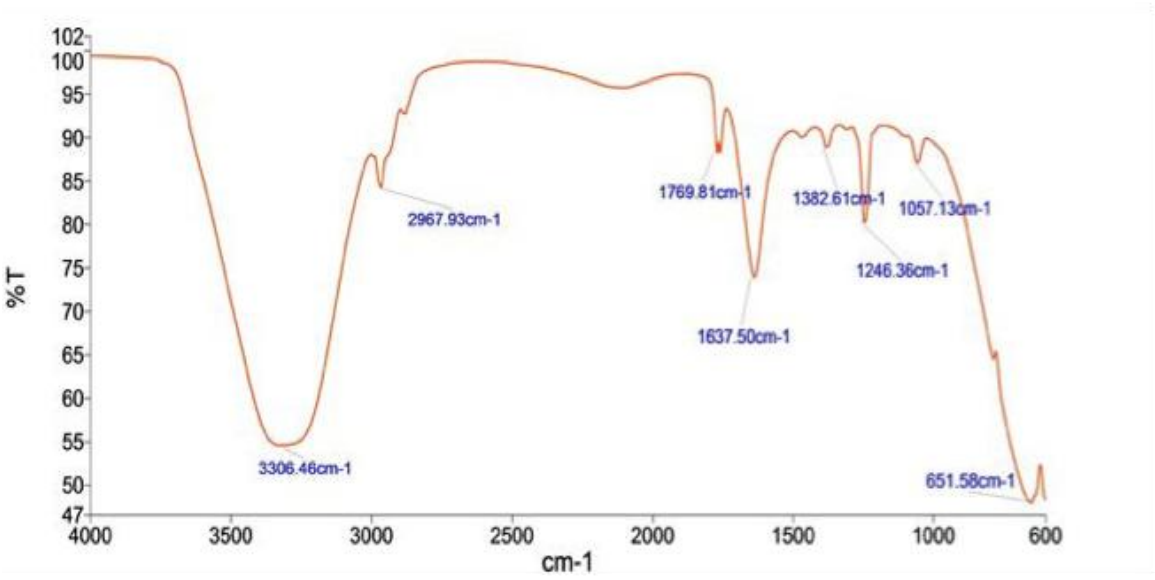
**Figure 2:** Image showing the SEM results of Siver nanoparticles




FTIR measurements identifies the possible biomolecules responsible for capping and efficient

stabilization of the metal nanoparticles synthesized by *Saraca asoca* leaf extract. Further the type nanoparticle was characterized by FTIR showed peaks at 2927, 1637 and 1382 reveals the presence of silver nanoparticle. (Figure 3) ( Devaraj . P *et al* ., 2013) **.**

**Figure 3:** Image showing the FTIR results of silver nanoparticle

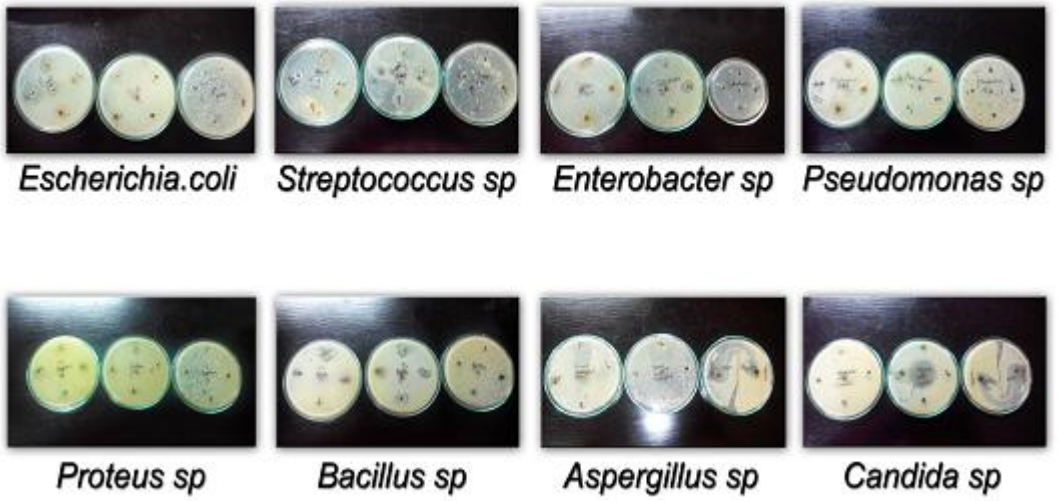



The green synthesized silver nanoparticles showed a significant antimicorbial actiivty when compared to the bioactive fractions of ethanol, Petroleum ether and aqueous extracts .The green synthesized silver nanoparticle showed good activity against *Streptococcus sp* (20 mm) and *Bacillus* sp(30mm) among the bacterial species and *Aspergillus sp* (30 mm) and *Candida sp* (30 mm) among the fungal species. ( Gahlaut .A *et al*., 2013), (Hawas U *et al*., 2013) The silver nano particles as antimicrobial agents is relatively new, because of their high reactivity due to increased volume to surface ratio, these nanoparticles play a significant role in inhibiting the microbial growth. (Balu S.S *et al*., 2012) . The strong interaction of silver with thiol groups present in key respiratory enzymes in bacteria is responsible for the inhibitory action of silver on bacterial cells. (Figure 4, Table 3) (Mohanty. S *et al.*,2013).

Table 2: Table showing the Antimicrobial activity of Silver nano particles and bioactive compounds from *S.asoca*

| Micro organism | Zone of inhibition in(mm) | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Crude sample | | | Peteroleum - ether fractions | | | Ethanol fractions | | | | Silver nanoparti-cle |
| | Ethanol crude | Pet - roleum ether crude | Aque-ous crude | F1 | F2 | F3 | F1 | F2 | F3 | F4 | |
| *Escherichia coli* | 25 | 20 | 20 | 15 | 10 | 10 | 0 | 0 | 0 | 0 | 0 |
| *Enterobactersp* | 20 | 15 | 15 | 15 | 20 | 15 | 0 | 0 | 0 | 0 | 10 |
| *Streptococcus sp* | 30 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 20 |
| *Pseudomonas sp* | 20 | 15 | 15 | 10 | 10 | 10 | 0 | 0 | 0 | 0 | 15 |
| *Proteus* | 20 | 20 | 15 | 25 | 20 | 30 | 0 | 0 | 0 | 0 | 10 |
| *Bacillus sp* | 30 | 15 | 0 | 25 | 20 | 20 | 0 | 0 | 0 | 0 | 30 |
| *Aspergillussp* | 5 | 10 | 20 | 40 | 20 | 10 | 0 | 0 | 0 | 0 | 30 |
| *Candida sp* | 10 | 10 | 50 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 20 |

Figure :4 Image showing the Antimicrobial actiivty of silver nanoparticles against microorganisms




## Conclusion

The leaf sample of *S.asoca* were collected , shade dried and powdered.Silver nanoparticle was prepared from 1 mM silver nitrate using the aqueous dissolved *S.asoca* by green synthesis method.The nanoparticles were characterized by UV spectroscopy, FTIR and SEM.The antimicrobial activity of the bioactive compounds were tested against various microorganisms and the diameter of the zone obtained were calculated and tabulated, which indicated that the bionanoparticles exhibited good inhibition when compared to the bioactive comounds from *S.asoca.*

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