

Green Synthesis and Antibacterial Activity of Silver Nanoparticles(AgNPs) using Psoraleacorylifolia Seed Extract



N. Thirumagal, A. Pricilla Jeyakumari

Abstract: Silver nanoparticles using psoraleacorylifolia seed extract has been synthesized by green synthesized method. The size of the nanoparticle was found to be 38nm by powder X-ray diffraction method. From the FTIR spectrum the vibrational frequency at 641cm^{-1} confirms the bioreduction of silver nitrate into Ag^+ ion and spherical morphology of silver nanoparticles were identified using SEM and TEM analysis. The optical properties were analysed and cut off wavelength at 429nm was found using UV-Vis spectrum. The surface potential of the green synthesized nanoparticles was calculated by Zeta potential method. Biosynthesized silver nanoparticles were further evaluated for their potential antibacterial activity and it may be used as nanomedicine for skin diseases.

Index term: Antibacterial activity, Green synthesis, Silver nanoparticles, Seed extract, XRD, Zetapotential.

I. INTRODUCTION

The green combinations of nanoparticles are of paramount importance in the domain of nanotechnology. Plant mediated synthesis of nanoparticles in a green chemistry method invariably links nanotechnology with plants. Metals of nanoparticles are used in medical fields as nanomedicines. For example Platinum, Gold, Silver, Copper and Zinc etc. The *Psoraleacorylifolia* are frequently found all over China. The seeds are hard and black in colour and bitter to taste. It contains enormous biological resources that help in the treatment of heterogeneous skin diseases. These seeds contain various chemicals that may include Isopsoralen, Posoralen, bavahalone, Dehydriopsoralidin, Methyl-4-hydrobenzoate, and Corylin Psoralidin. Another name for *Psoraleacorylifolia* is Bakuchi. It is a significant herb used in the Indian medicine system from ancient times. This plant is chemoprotective, anti-inflammatory, antioxidant and antimicrobial in nature. The inorganic nanoparticles prepared using biological methods from the plants like

Moringaoleifera[1], *MalusDomestica*[2], *Hibiscusrosa-sinensis*[3], *AzadirchataIndica*[4], *Alliumacepa*[5], *EllaatoriaCardamon am*[6], *Cadiosperumhelicocacabu*[7], *Allphobiahirta*[8], *Eriobotryajaponica*[9] and *Vitex negundo*[10] are biocompatible and ecofriendly and used in biomolecular detection and used as catalysis, biosensor and medicine. When aqueous solution is added to *Psoraleacorylifolia* seed extract a chemical reaction takes place for which the biomolecules of *Psoraleacorylifolia* seed act as an agent and in the result of this process AgNPs are acquired. Imbued by the aforementioned facts, the present investigation is on the green synthesis of silver nanoparticles from *Psoraleacorylifolia* seeds in pursuit of antibacterial properties.

II. MATERIALS AND METHOD

A. Sample preparation

5g of *Psoraleacorylifolia* powder was added to 100ml of distilled water and the mixture was boiled for 2min at 100°C in a water bath, and then passed through Whatmann No.1 filter paper and also the mixture were collected in a beaker. 5mM aqueous solution of Silver Nitrate (AgNO_3) is prepared. 10ml of seed extract was added into 50ml of aqueous solution of AgNO_3 dropwise with constant stirring at 60°C and colour changes is observed that pointed out the formation of AgNPs (Ag^+). The beaker was incubated at 4°C for 24hrs and then centrifuged at 10,000rpm. The supernatants were collected, dried and then used for characterization

B. Characterization and synthesized AgNPs

i. Powder X-ray diffraction (XRD) method

The purified AgNPs were freeze dried and analyzed to evaluate their Crystalline structure and govern the mean size in powder X-ray diffractometer (Bruker D8 Advance, Germany) equipped with $\text{CuK}\alpha$ ($\lambda=1.5418\text{\AA}$) x-ray radiation source in the range from 10° - 80°C .

ii. UV-Visible spectroscopy

Biosynthesized AgNPs were examined by using UV-Vis spectrum ($\lambda=35$). In UV-Vis spectral study the wavelength was noticed in the range of 300-1100nm.

iii. FT-IR analysis

FT-IR analysis was brought to a conclusion by FT-IR spectrometer in the range of 4000 - 400cm^{-1} .

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The synthesized AgNPs sample was dried and grinded with KB_r pellets and analysed . The various modes of vibrations were recognized to confirm the functional groups present in AgNPs.

iv. SEM with EDAX

A small amount of sample was placed on a carbon coated copper grid to prepare thin film of the sample and the extra solution was wiped off by blotting paper and empowered to dry under mercury lamp for 5minutes to look at the form of the particles.Biosynthesized AgNPs of *Psoraleacorylifolia* on carbon film werescrutinized using EDAX JEOL6360 TESCAN.

v. TEM analysis

The surface Morphology and particle size of the synthesized AgNPs were assessedby TEM (Tecnai spirit G² with 120KV) applying accelerating voltage fromLaB₆source.TEM samples were gradually developed by drop castinga small volume of AgNPs in water onto carbon-copper grid before examining inTEM. The sample wascontained to be in a thin film and made dried under a vacuumwith a mercury lamp for 30minutes. Then the shape andmagnitude of the particle were observed.

vi. Antibacterial activity

Antibacterial activity of synthesized AgNPs wasexperimented with *Staphylococcus aureus*, *Bacillus sp*, *Klebsiellasp* and *Escherichia coli* bacterium by agar disc diffusion technique

III. RESULTS AND DISCUSSION

A. POWDER X-ray DIFFRACTION

The fig.1reveals the X-ray diffraction design performed by the synthesized AgNPs after the reduction of *Psoraleacorylifolia*.The four peaks were obtained at $2\theta=38^\circ, 44^\circ, 64^\circ,77^\circ$ corresponding to the planes (111),(200),(220),(311) of face centered cubic crystal of AgNPs as per [JPDFS File No: 04-0783]. For cubic system, the lattice constant values($a = b = c=4.0677 \text{ \AA}$) and d-spacing value $2.3485 \times 10^{-10} \text{ m}$. These diffraction peaks match very well not only with the relative peaks position but also with this relative peak intensities. The average grain size of AgNPs was calculated from Debye Scherrer’s equation and mean crystalline size of the particles is found to be 38nm.Thiscorrelates with Peulmakumariponnaian.et al.

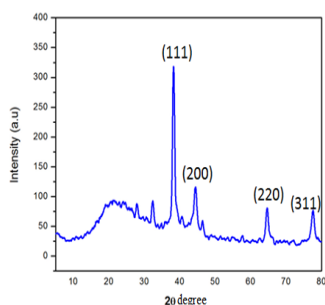


Fig. 1 XRD pattern of synthesized silver nanoparticles

B. FT-IR ANALYSIS

In the present investigation, the Fig.2a ostensibly shows the functional groups present in the *Psoraleacorylifolia* seed extract and Fig.2b shows the spectrum of synthesized AgNPs and it exhibits the transmission peaks were at 3405, 2956, 2856, 1632, 1516, 1384, 1102 and 622 cm^{-1} . Likewise, figure 2a conveys the transmission peaks were at 3293, 2958, 2826, 1654, 1541, 1450, 1384, 1101 and 641 cm^{-1} . Three absorption peaks at 834, 1107, and 1168 indicates –C-N stretching vibrations of amine, C-O-C- or C-O groups[12, 13], C-O-C- , C-O and –C=C- are obtained from heterocyclic compound. These above – mentioned compounds exist in *Psoraleacorylifolia* seed extract are capping agent of the formation of AgNPs[14] . The broad peaks at 3405, 2926and 2856 cm^{-1} weresuitable to OH amine (NH) group and aliphatic C-H of *Psoraleacorylifolia* seed extract respectively, 1632 indicates –C-OH stretching 1384 indicates methyl bending stretching respectively The stretching bond 3293,1654 and641 cm^{-1} disappears in Figure 2b due to the bioreduction confirms the silver nitrate into Ag^+ ion.The FT-IR bands of *Psoraleaorylifolia* seed extract and AgNPs can be compared and given in table 1.

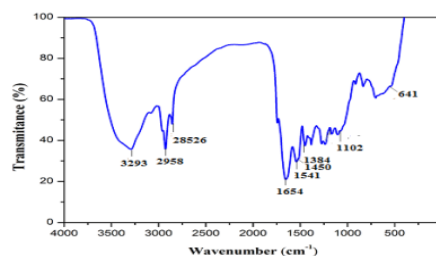


Fig. 2aSeedextract FTIR (spectrum)

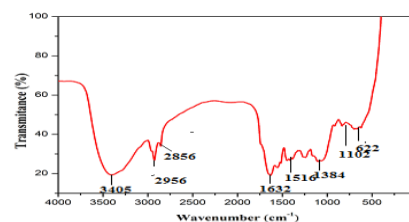


Fig. 2b synthesized AgNPs (FTIR spectrum)

Table1. Comparison of FT-IR bands

Vibrational frequencies (cm^{-1})		Assignments [15]
Synthesised AgNPs	Seed extract	
3405	3293	Water of hydration
---	3293	Asymmetric NH stretching
2926-2856	2958-2826	Symmetric NH stretching
1632	1654	C-OH Stretching
1516	1541	C=O unsaturated Ketones
1384	1384	methyl bending

1102	1107	C-O stretching
---	641	C-C stretching
622	--	Ag ⁺ ions

C. UV-Vis SPECTRUM ANALYSIS

The synthesized AgNPs by the reduction of *Psoraleacorylifolia* clearly exhibits the maximum absorbance at 429 nm due to the collective oscillation of surface electron. Henceforth, it has an excellent optical transparency in the complete visible region and the band gap energy calculated is 2.89 eV [fig. 3].

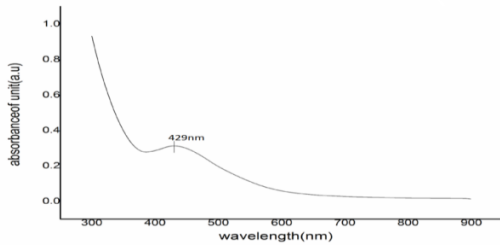


Fig.3 UV Spectrum of Silver nanoparticles synthesized by *Psoraleacorylifolia* seed extract

D. SEM WITH EDAX ANALYSIS

The SEM image of the sample in Fig. 4 gives direct facts of the magnitude and morphology of the unified AgNPs nanoscale particle. The SEM of AgNPs implies that it has a spherical form of morphology with wider particles dissemination. The agglomerates may be due to the drying process. Fig. 4 shows the presence of agglomerates bordering on 5-10 μm in diameter.

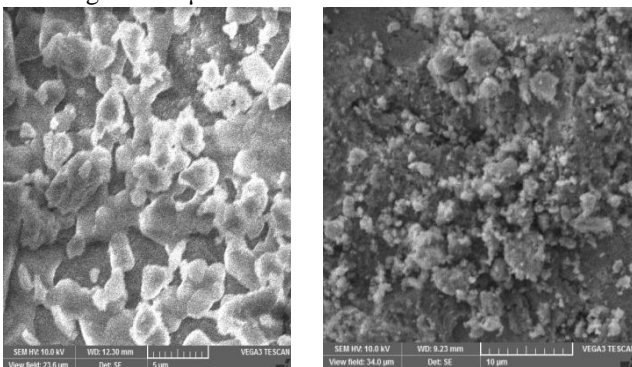


Fig. 4 SEM images of synthesized silver nanoparticles

The energy dispersive X-ray analysis (EDAX) reveals well built signal and confirms the formation of AgNPs. Thus, plain reaction in this preparation was between silver nitrate and *Psoraleacorylifolia* which led to Ag⁺ ions. The EDAX (fig.5) pattern shows the element present in the sample as shown in the table 2.

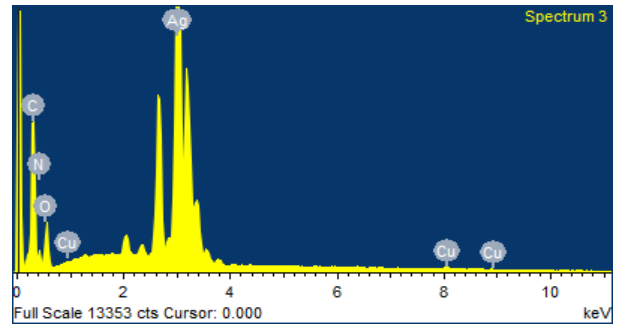


Fig. 5 EDAX of synthesized silver nanoparticle

Table 2 EDAX of synthesized *Psoraleacorylifolia* AgNPs

Element	Weight%	Atomic%
C K	15.04	30.35
N K	19.69	34.07
O K	16.16	24.49
Cu K	0.41	0.16
Ag L	48.70	10.94

E. TEM ANALYSIS

TEM technique was used to determine the accurate size and the morphology of AgNPs from *Psoraleacorylifolia* seed extract. TEM images (Fig.6) clearly show that the particles are round in structure and the bar marker representing the particle sizes as 20 nm-23 nm.

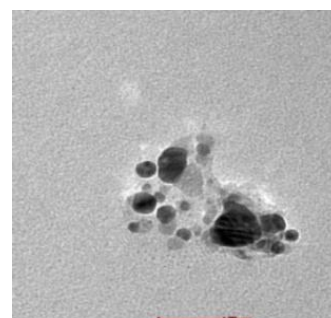
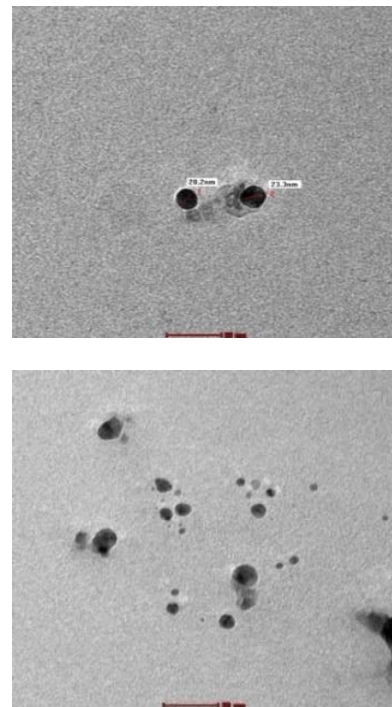


Fig. 6 TEM images of synthesized silver nanoparticles

F. ZETA POTENTIAL

Interestingly enough, A Zeta potential was employed to assess the surface potential of synthesized AgNPs solutions and its stability. The Zeta potential was quantified the electro kinetic potential of colloidal dispersion.

In the prepared sample zeta potential value of synthesized AgNPs was found to be -25.7mV with 100% intensity. Hence the AgNPs were well constructed in colloidal solution as shown in (fig.7). The calculated Zeta potential lies in the range (+25mv) (-25mv) made the nanomaterial so stable.

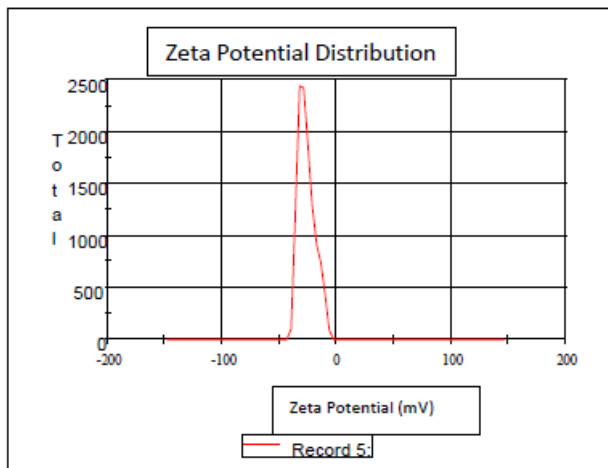
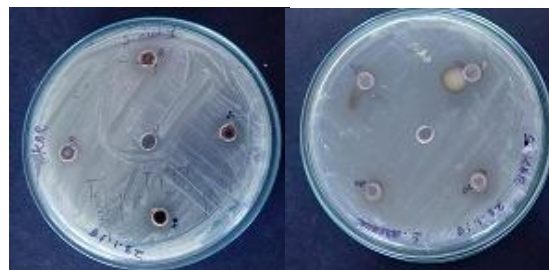


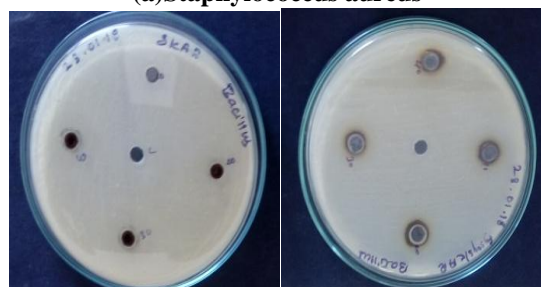
Fig. 7 Zeta potential of synthesized AgNPs using *PsoraleaCorylifolia* seed extract

G. ANTIBACTERIAL ACTIVITY

The antibacterial activity of synthesized Ag nanoparticles was determined using the agar disc diffusion assay method [16]. The antibacterial activity of seed extracts was evaluated against four bacteria species: *Staphylococcus aureus*, *Bacillus sp*, *Klebsiellasp* and *Escherichia coli* and the growth inhibition zones were shown in the fig.8. The Muller Hinton agar was prepared and poured into the sterile petriplates. The bacterial inoculums was invariably spread using sterile cotton swab on a Muller Hinton agar plate. Wells were made with the help of a cork borer (7 mm). Seed extracts were used in various concentrations of 10, 20, 30 and 40mg substances for each of the 4 wells respectively. One well control was maintained at 7 mm diameter holes cut in the agar gel, 20 mm apart from one another. The plate, in general, incubated for 24 h at 36°C ± 1°C, under aerobic conditions. After incubation, confluent bacterial growth was mentioned. Effect of seed extract synthesized AgNPs on pathogens was resolved by diameter of the zone of inhibition and the maximum zone inhibition was found against *Staphylococcusaureus* was 30mm as viewed in the table. 3. The Fig.9 obviously shows the distinction of zone inhibition of variegated micro organism by bar diagram method.



(a) *Staphylococcus aureus*



(b) *Bacillus sp*



(c) *Escherichia coli*



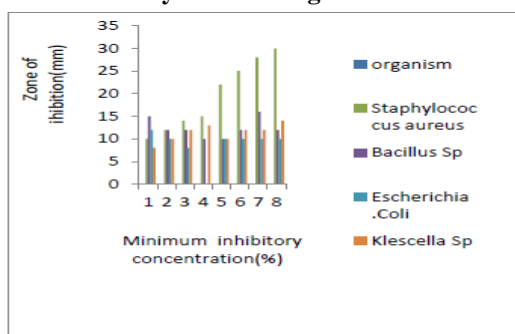
(d) *Klebsiella sp*

Fig.8 Zone inhibition of various bacterial in seed extract and synthesized AgNPs

Table3. Diameter of Zone of inhibition (mm) for Different concentration of different microorganism

Organisms	Zone of inhibition in Diameter (mm)							
	Different concentration							
	Seed extract				synthesized AgNPs			
	10 mg	20 mg	30 mg	40 mg	10 mg	20 mg	30 mg	40 mg
<i>Staphylococcus aureus</i>	10	12	14	15	22	25	28	30
<i>Bacillus Sp</i>	15	12	12	10	10	12	16	12
<i>Escherichia .Coli</i>	12	10	8	-	10	10	10	10
<i>Klebsella Sp</i>	8	10	12	13	10	12	12	14

Fig.9 The zone inhibition induced by seed extract and synthesized AgNPs



IV. CONCLUSION

The Silver nanoparticle was combined by green blended technique applying *Psoralea corylifolia* as diminishing agent. From the powder X-ray diffraction pattern the particle size was found to be 38nm. The optical property determined was confirmed the material is suitable for optoelectronic device fabrications. The components present were validated by EDAX. The structural morphology of the nanoparticles and size as substantiated by SEM and TEM. For the first time, the surface potential and stability for the synthesized nanoparticles by using *Psoralea corylifolia* as reducing agent was calculated (-25.7) and also the silver nanoparticle became widely known to control the most productive antibacterial activity in the face of *Staphylococcus aureus*. The result of research shows and confirms that the biosynthesized AgNPs using *Psoralea corylifolia* seeds can help to produce effective nanomedicine for skin diseases.

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