

INVESTIGATION OF BIOGENIC SILVER NANOPARTICLES GREEN SYNTHESIZED FROM *CARICA PAPAYA*

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ABSTRACT

Objective: This work mainly focuses on the investigation of optical, structural and morphological characteristics of Silver nanoparticles (Ag-NPs) synthesized using Papaya (*Carica Papaya*) leaf extract and to study the mechanisms involved in the formation.

Methods: Ag-NPs were synthesized using colloidal method from silver nitrate using aqueous leaf extract of Papaya as reducing agent. Spectral analysis of Ag-NPs was done using UV-Vis spectroscopy and optical characteristics were studied. Fourier Transform Infrared (FTIR) analysis was done to investigate the mechanisms involved in the reaction. A detailed study of structural and morphological properties was done using X-Ray Diffraction (XRD) and Atomic Force Microscopy (AFM).

Results: Spherical shaped Ag-NPs of Face Centered Cubic (FCC) structure were formed and the average particle size was in the range 25-35 nm.

Conclusion: Papaya is a good reducing agent for the synthesis of Ag-NPs. Since it is a plant with medicinal values, thus synthesized Ag-NPs can be used for medicinal implications.

Keywords: Silver nanoparticles, Papaya, Atomic Force Microscopy, Green synthesis.

INTRODUCTION

Nanoparticles are now a significant interest owing area due to the unique, interesting and new or improved properties compared to their bulk counterparts. These superior properties arise mainly due to the large surface to volume ratio with decreasing particle size and the increased number of particles available at the grain boundaries [1, 2]. These materials are widely being used in the areas of medicine, catalysis, water treatment and solar energy conversion [3, 4].

The attractive physiochemical properties like anti-bacterial activity, catalytic activity, chemical stability and good conductivity makes Ag-NPs an attractive focus of study [5]. It is a safe and nontoxic inorganic antibacterial agent which can destroy disease-causing microorganisms [6]. So Ag-NPs are essential candidates in the field of nanotechnology and nanomedicine [7]. There are different physical, chemical and biological methods for the synthesis of Ag-NPs. Most of the physical and chemical methods involve toxic chemicals, high energy requirements etc. Biological methods for synthesis of Ag-NPs are ecofriendly alternative to physical and chemical methods. Biosynthesis methods are non-toxic, rapid, environmentally regime and comparatively inexpensive. These methods include use of microorganisms, fungi, enzymes and plant extracts as reducing and stabilizing agents for the synthesis of nanoparticles [8, 9]. Eliminating the need of cell cultures is one of the major advantages of using plant extracts and it is also a rapid method [10]. Many studies have been done on the green synthesis of Ag-NPs from plant extracts. From ancient cultures, people have a tradition of using plants medicinally and as a biological agent [11]. Some of the widely used medicinal plants are Aloe Vera [12], Tulsi [13], Nilgiri (*Eucalyptus chapmaniana*) [14], Amla (*Emblicaofficinallis*) [15], Ashwagandha (*Withaniasomnifera*) [16], TecomaStans [17], Rosa rugosa [18], Hibiscus rosasinensis [19] etc. *Carica Papaya* is also a plant with good medicinal values [20]. So for synthesis of Ag-NPs for medical applications plant extracts of Papaya can be used as reducing and stabilizing agent.

Synthesis of Ag-NPs from papaya was first reported by Namrata et al. using Callus extract of papaya as reducing agent. Spherical Ag-NPs of 60-80 nm size range was reported [21]. Papaya fruit extract for Ag-NPs synthesis was reported first by Devendra J et al. and that was the first Ag-NPs synthesis from any plant fruit extract [22]. Later, Farooqui M et al. have demonstrated the synthesis of Ag-NPs which

are highly toxic against different multidrug resistant human pathogens using papaya fruit extracts [23]. Ratika K et al. reported the synthesis of 2-5 nm sized Ag-NPs from papaya leaf extract; in which leaf extraction involves hot percolation and cold percolation treatments. Ag-NPs were of spherical shape, which was found to possess antimicrobial activity against the human pathogenic bacteria [20]. Though works on papaya plant extracts for Ag-NPs have been reported, a detailed investigation on the properties of Ag-NPs from the leaf extract of papaya and mechanism involved in the reaction has not been performed. In this work we present a detailed study of optical, structural and morphological characteristics of Ag-NPs and the mechanisms involved in the formation of Ag-NPs using *Carica Papaya* leaf extract. In the present work, we have demonstrated the preparation of Papaya leaf extract using a much simpler and faster method.

MATERIALS AND METHODS

Preparation of papaya leaf extract

The fresh leaves of papaya were collected from the *Carica Papaya* trees which are located in the VIT University Campus.

Table 1: Different experimental conditions

S. No.	Sample conditions
1	Fresh 1:1 Centrifuged.
2	Fresh 1:5 Centrifuged.
3	Fresh filtered 1:1
4	Fresh filtered 1:5



Fig. 1: Fresh Papaya leaves

Fig. 1 shows the freshly collected Carica leaves picture. These were washed with DI (De-ionized) water three times and kept for drying. For the experiments about 25 gm of the leaves were used. Then they are cut into pieces and mixed with the required amount of DI water. Further the mixture was kept for boiling for about 20 minutes [20] to obtain the leaf extract.

Synthesis of Silver nanoparticles

For each experimental set up two flasks were taken. The plant extract obtained from the above mentioned method is further added to the AgNO_3 solution. The different conditions in which the synthesis of Ag-NPs was carried out are mentioned in the table 1.

Fig. 2 shows the extracts obtained from the papaya leaves. fig. 2a is the fresh extract obtained immediately after boiling the leaves. Fig. 2b and 2c show extract mixed with AgNO_3 in 1:1 and 1:5 proportions respectively. In the former case a color change from dark brown to light brown color is observed while in the latter case the color changed from dark brown to faint brown respectively. These 1:1 and 1:5 proportion solution was further divided into two parts, one was centrifuged and the other was filtered as described in table 1.

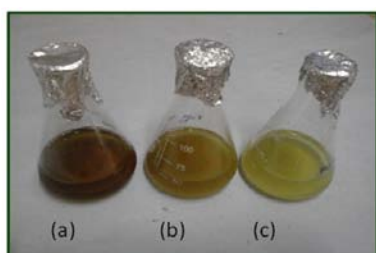


Fig. 2: [a]The fresh extract obtained immediately after boiling [b]1:1 reaction condition and [c]1:5 reaction condition respectively

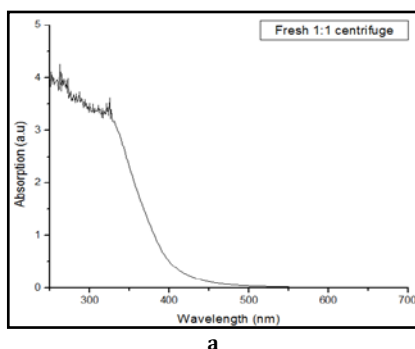
Characterization

Characterization of Ag-NPs

Powder X-Ray Diffractometer (Model No. D8 Advance, BRUKER, Germany) was used for the structural analysis of synthesized Ag-NPs. For all the samples with different conditions the UV-Vis spectra were recorded using the UV-VIS Spectrophotometer Analytik Jena Specord 210 plus from 250 nm to 700 nm range. For the morphological investigation of centrifuged samples, AFM analysis was carried out using the Nanosurf easy scan2 AFM instrument. FTIR spectroscopy of papaya leaf extract and synthesized Ag-NPs were recorded using ALPHA-T, Bruker FTIR spectrometer instrument.

RESULTS AND DISCUSSION

Reduction of silver ions present in the aqueous solution of silver complex, occurred during the reaction with the ingredients present in the papaya leaf extract. The formation of Ag-NPs and the involved mechanisms was studied from the data obtained from different characterization techniques.



XRD analysis

The formation and crystalline structure of Ag-NPs synthesized were confirmed from the obtained XRD pattern shown in [fig. 3]. Diffraction peaks were obtained at 2θ values 37.8° , 44.18° , 64.39° and 77.36° corresponding to Bragg reflections at (111), (200), (220) and (311) respectively. The diffraction pattern shows the formation of silver with the face centered cubic lattice in reference to JCPDS 89-3722 data.

Table. 2: XRD Analysis

Diffraction angle (degrees)	Crystal plane
37.8	111
44.18	200
64.39	220
77.36	311

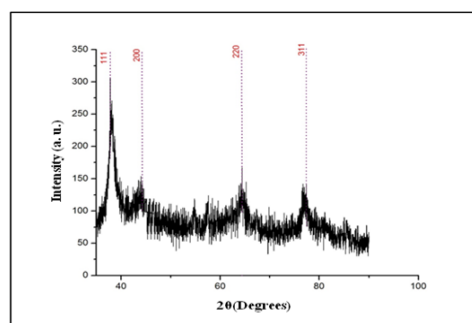
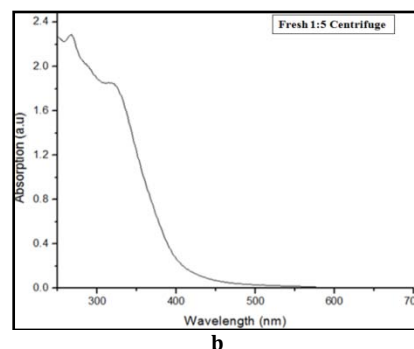


Fig. 3: XRD pattern of synthesized Ag-NPs

UV-Vis analysis

The mixing of Papaya leaf extract with AgNO_3 solution resulted in a color change as shown in [fig. 2] which is due to the Surface Plasmon Resonance (SPR) of Ag-NPs. This color change indicates the formation of Ag-NPs and the different colors obtained for centrifuged as well as filtered samples can be due to the difference in average particle sizes. A similar color change was reported with papaya fruit and leaf extracts which is due to the formation of Ag-NPs [20-23]. For further investigation on the optical properties of Ag-NPs, UV-Vis analysis was done for all the experimental conditions as described in table 1. The spectra in [fig. 4] shows the same. The UV-Vis spectrum was taken within 30 minutes after the reaction. In all the four conditions SPR of Ag-NPs resulted in absorption ranging from 250 nm-500 nm. Absorption intensities of Ag-NPs obtained were 4, 2.3, 3.6 and 1.6 a. u. for 1:1 centrifuged, 1:5 centrifuged, 1:1 filtered and 1:5 filtered respectively. For centrifuged samples the intensity was found to be more than that of filtered samples. This indicates the absorption of 1:1 proportion is greater than that of 1:5 proportions. Maximum absorption of 4 a. u. was obtained for 1:1 centrifuged extract. The broad absorption peak indicates that the particles are polydispersed.



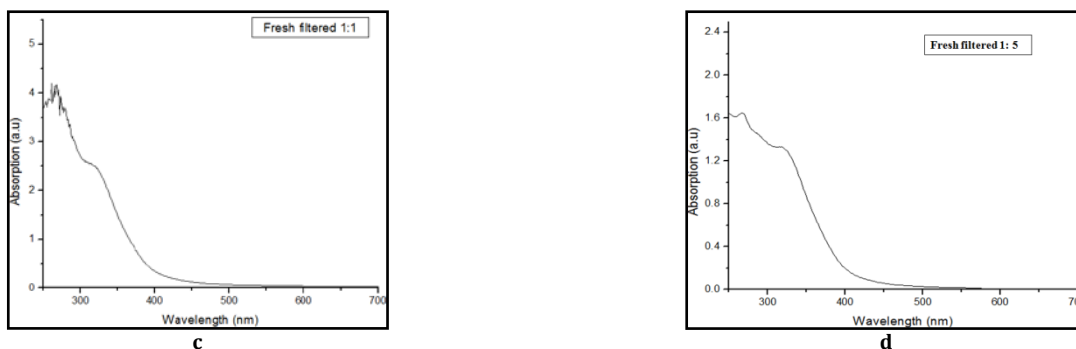


Fig. 4: UV-Vis spectra of Ag-NPs formed from [a] Centrifuged 1:1 [b] Centrifuged 1:5 [c] Filtered 1:1 and [d] Filtered 1:5 condition

AFM analysis

Centrifuged samples showed maximum optical absorption, hence the morphology of only those samples were studied further. AFM examination of Ag-NPs from papaya leaf extract is done for the first time. AFM images of Ag-NPs synthesized from centrifuged 1:1 and 1:5 conditions are shown in [fig. 5a and 5b] respectively.

It shows the formation of Ag-NPs of spherical shape and the particle size in the range 25 nm-30 nm and 35 nm-50 nm for 1:1 and 1:5 conditions respectively. In literature, there exist reports for the AFM study of Ag-NPs from leaf extracts of other plants like Pimentadoica [24]. Our AFM data shows formation of spherical, more uniform and large number of Ag-NPs formed from papaya leaf extract. Results show that the nanoparticles obtained from 1:1 experimental condition were more uniform than that obtained from 1:5 conditions.

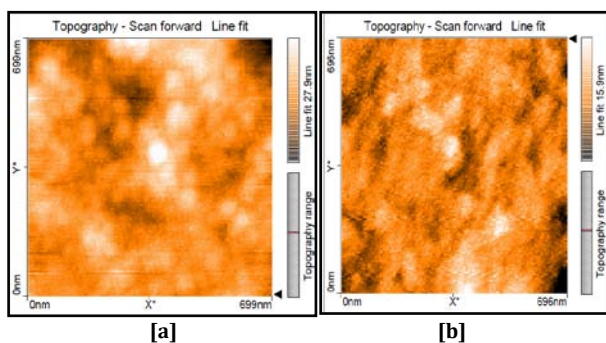


Fig. 5: AFM images of Ag-NPs from [a] Centrifuged 1:1 sample and [b] Centrifuged 1:5 samples

FTIR analysis

FTIR spectroscopy is helpful in examining the substance structure of the surface of the silver nanoparticles and the biomolecules responsible for the formation. Till date, no reports are available for the FTIR study of Ag-NPs from Papaya leaf extract. Hence FTIR analysis was done for our samples to study the compounds responsible for the formation of Ag-NPs. The spectra for papaya leaf extracted before and after bio reduction of Ag-NPs are shown in [Fig. 6a and 6b] respectively. Absorbance bands at 3417 cm⁻¹, 1620 cm⁻¹, 1535 cm⁻¹ and 1400 cm⁻¹ wavenumber have been observed. 3417 cm⁻¹ can be due to OH stretching in alcohols and phenolic compounds. In case of Grandiflora leaf extract, the FTIR analysis showed C-N stretching vibrations and amide-I bands occurring at 1628 cm⁻¹, hence in our case 1620 cm⁻¹ can be due to amide I bond of proteins arising from carbonyl stretching in proteins [25].

As a result carbonyl group of amino acids is acting as reducing and stabilizing agents for the formation of Ag-NPs similar to that seen in the reports for leaf extracts of Carob and Ocimum [26, 27]. FTIR spectroscopic study affirmed that the papaya leaf extract is capable of reduction and stabilization of Ag-NPs.

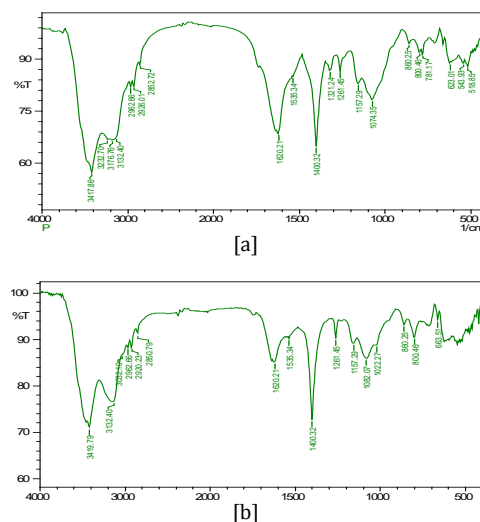


Fig. 6: FTIR spectrum of papaya leaf extract [a] before bioreduction and [b] after bioreduction

CONCLUSION

Silver nanoparticles were successfully synthesized from papaya leaf extract using simple and rapid green reduction method. The XRD pattern obtained was compatible with the standard pattern of Ag with FCC structure which confirms the formation of Silver. Ag-NPs synthesized from the centrifuged 1:1 condition shows good absorption when compared to other conditions. AFM data revealed that the Ag-NPs obtained are of spherical shape with particle size of 25 nm-30 nm. Papaya plant is having good medicinal value; hence Ag-NPs synthesized from this material can be used for various medical applications.

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CONFLICT OF INTERESTS

Declared None

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