

CARICA PAPAYA MEDIATED GREEN SYNTHESIZED SILVER NANOPARTICLES

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Received: 21 Jan 2018, Revised and Accepted: 08 Mar 2018

ABSTRACT

Objective: The present study was designed to biosynthesize NPs from leaves to study the reducing Ag⁺ ions and stabilizing the particles and confirm AgNP synthesis by using various spectroscopy and microscopic methods.

Methods: Bio-inspired AgNPs were rapidly synthesized at room temperature using fresh aqueous leaf extract of *Carica papaya*. A green and low-cost synthesis was effective in the formation of stable crystalline NPs in the solution. Amine, alkene and alkyl halides groups present in the *Carica papaya* leaf extract functioned as reducing as well as a stabilizing agent to produce shape controlled AgNPs.

Results: SPR confirmed the formation of AgNPs in UV-Visible spectra at 445.7 nm. The XRD result also showed the presence of elemental Ag⁰ as a crystalline nature. Study the functional groups responsible for the bio reduction of Ag⁺. HE-TEM and FE-SEM with EDX image showed spherical crystalline AgNPs.

Conclusion: Hence, the plant-based bio AgNPs could be used in biomedical applications.

Keywords: *Carica papaya*, Biosynthesis, AgNPs, HE-TEM, XRD, FE-SEM and EDX

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DOI: <http://dx.doi.org/10.22159/ijcpr.2018v10i3.27221>

INTRODUCTION

Nanomedicine is an emerging field expanding rapidly because of the development and incorporation of new nanocomposites into a range of products and technologies. In recent years, the application of nanoparticles (NPs) in medicine has increased and expanded to the fields of molecular imaging [1], drug delivery [2], diagnosis and treatment of cardiovascular diseases [3], wound healing [4] and development of materials and medical devices with antimicrobial properties [5].

New applications of NPs and nanomaterials are emerging rapidly in biomedical sciences [6]. This decade has witnessed the inception of new significant technological products particularly based on nanotechnology; NPs synthesis is being widely explored since they exhibit unique size and shape dependent properties for applications in optics, electronics, catalytic systems, magnetic and biomedical fields such as HIV inhibition, cancer cell cytotoxicity and genotoxicity [7]. Apart from this, recently the anti-tumour effect of AgNPs has been reported against different cancerous cell lines [8]. NPs with the size range between 1 and 1000 nm are mainly explored for the diagnosis and treatment of human cancers, which led to the new discipline of nano-oncology [9].

There are a number of methods used for the synthesis of silver nanoparticles (AgNPs) including physical and chemical methods [10-13], electrochemical reduction [14, 15], photochemical reduction [16] and thermal evaporation [17-18]. However, a rapid and green synthesis method using plant extract has developed an enormous interest in AgNPs synthesis due to green chemistry approach. Moreover, it is simple, cost-effective, eco-friendly, easily scaled up for large-scale synthesis, without using toxic and redundant chemicals in solid, liquid and gaseous form [19]. Indeed, a number of bacteria [20], fungi and yeast have been well-known for the synthesis of non-toxic noble NPs [21]. However, the microbial-mediated synthesis of NPs is not industrially feasible as it requires expensive medium and maintenance of highly aseptic conditions [22].

In this context, plant-mediated NPs synthesis seems to be a cost-effective as well as eco-friendly method. Moreover, NP synthesis from plants with medicinal properties proves to be beneficial in

treating various ailments in a better and easy way. On such plant is Papaya, a tropical fruit, often seen in orange-red, yellow-green and yellow-orange hues with a rich orange pulp. Whole plant parts, fruits, roots, bark, peel, seeds and pulp are known to have medicinal properties. It has been used for the treatment of numerous diseases like warts, corns, sinuses, eczema, cutaneous tubercles, blood pressure, dyspepsia, constipation, amenorrhoea, general debility, expel thread worms and stimulate reproductive organs [23]. It also effectively treats and improves all types of digestive and abdominal disorders [24-27]. Leaves of papaya, one of the plant part with numerous medicinal value have the history of streaming and eating with spinach in Asia [23]. It has found to have a significant effect on various tumor cell lines and the tea extract of leaves found to have antimicrobial, ease menstrual pain, relieve nausea, increase the appetite and antispasmodic activities [23]. Phytochemical investigations were carried out on this plant by several authors, which revealed the presence of carbohydrates, phenols, flavonoids, sterols, alkaloids, saponins, cardiac glycosides, ecdysterone *etc.*, from different parts of this plant. Survey of the literature revealed that NPs synthesis from *Carica papaya* leaves is scanty. In view of this, the present study was designed to biosynthesize NPs from leaves to study the reducing Ag⁺ ions and stabilizing the particles and confirm AgNP synthesis by using various spectroscopy and microscopic methods.

MATERIALS AND METHODS

Collection and identification

Fresh leaves of *Carica papaya* were collected from Presidency College, Chennai, Tamil Nadu, India, and were authentically identified by Dr. P. Jayaraman, Chennai, India, *Carica papaya* as (Caricaceae) with voucher specimen no: 123/PARC/2017.

Preparation of leaf extract and synthesis of AgNPs

Twenty grams of fresh leaves were washed thoroughly in tap water and distilled water for 30 min. in order to remove the debris. The aqueous extract was prepared by taking 25 g of washed and finely chopped leaves in 100 ml conical flask along with 100 ml of distilled water and the mixture was boiled at 45°C for 30 min. This aqueous extract was filtered through Whatmann No. 1 filter paper and was used for the synthesis of AgNPs. 10 ml of this aqueous leaf extract

was added to 90 ml of 1 mmol aqueous silver nitrate solution for the synthesis of AgNPs. A control setup silver nitrate was also maintained without *Carica papaya* extract.

Qualitative phytochemical analysis

Qualitative phytochemical analysis of aqueous extract was carried out by the method of Harborne (1973) [28].

Characterization of silver NPs

UV-Visible spectra were recorded as a function of the reaction time on PG Instruments spectroscopy. The studies on size, morphology and composition of the NPs were performed by means of HR-TEM (PHILIPS TECNAI 10) and FE-SEM with EDX (Carl Zeiss). The purified AgNPs were examined for the presence of biomolecules using FTIR analysis. Briefly, the spectrum obtained from the dried sample was recorded on FT-IR spectrum (Perkin-Elmer, USA) in the diffuse reflectance mode at a resolution of 4 cm⁻¹ in KBr pellets. Particle size analyzer was done dynamic light scattering (Malvern, MAL 1062727, UK). Crystalline AgNPs were determined by XRD. Briefly, the biosynthesized AgNPs were laid onto glass substrates on

Phillips PW 1830 instrument operating at a voltage of 40 kV and current of 30 mA with Cu K α 1 radiation.

RESULTS AND DISCUSSION

The present study was aimed to identify the phytochemicals present in *Carica papaya* and to synthesize AgNPs from the aqueous extract of *Carica papaya* leaves which is distributed as a weed throughout India. Phytochemical study of *Carica papaya* leaf extract shows the positive results for carbohydrates, tannins, saponins, phenols, flavonoids, alkaloids, anthocyanin, terpenoids and triterpenoids. On the other hand coumarins and steroids were absent in the aqueous extract (table 1).

The bioactive compounds such as polyphenol, carbohydrates, vitamin and trace elements present in the leaf extract plays an important role as an antioxidant, anticancer, antitumor, anti-inflammatory, anti-obesity, anti-helminthic, analgesic, antipyretic, anti-nociceptive, anti-hepatitis, hepatoprotective, cardiac, Diuretic agent, attributing platelet augmentation, anticancer property, anti-acne activity, easing menstrual pain and relieving nausea. [31-34].

Table 1: Qualitative phytochemical analysis of *Carica papaya* aqueous extract

S. No.	Secondary metabolites	Aqueous extract
1	Carbohydrate	+++
2	Tannins	+
3	Saponins	+++
4	Flavonoids	+++
5	Alkaloids	+++
6	Anthocyanin	+
7	Quinones	+
8	Glycosides	-
9	Cardiac glycosides	++
10	Terpenoids	+
11	Triterpenoids	++
12	Phenols	+++
13	Coumarins	-
14	Acids	+
15	Protein	+
16	Steroids	-

++Strongly positive++Positive+Trace-Not detected

UV-vis analysis of green phyto-synthesized solutions

The aqueous extracts of papaya leaves (*Carica papaya*) when mixed in an aqueous solution of silver ion complex, the reduction of pure Ag⁺ ions to Ag⁰ was monitored by measuring

UV-vis spectrum of the reaction media. The reduction of silver nitrate into AgNPs during exposure to plant extract is followed by a gradual increase in colour development from clear to dark brown colour, as a result of the surface plasmon resonance phenomenon (fig. 1).

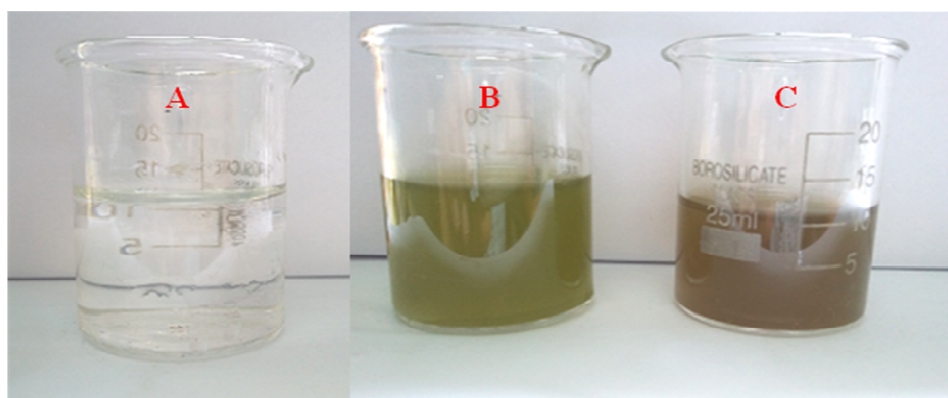


Fig. 1A: Color intensity of 1 mmol AgNO₃ solution, B. Aqueous extract of *Carica papaya* (pale light green colour), C. AgNPs synthesized at different hours with pale light green colour to dark brown colour

The UV-visible spectral analysis of (*Carica papaya*) without AgNO₃ did not show any change in colour. The UV-vis spectra of all the samples exhibited distinct surface plasmon resonance bands. The position of plasmon resonance band in UV-vis spectra is sensitive to particle shape, size, its interaction with the medium and the extent of charge transfer between medium and the particles. Here, the sharp bands were centered at 445.7 nm in (*Carica papaya*) AgNPs extract clearly indicate the presence of silver nanoparticles (fig. 2).

Similarly, AgNPs synthesized by using aqueous extract of *Sargassum polycystum* showed absorbance at 430 nm [35], *Sargassum longifolium* showed absorbance peak at 460 nm [36], *Eucalyptus hybrid* [37], *Acalypha indica* [38], *Solanum tarvum* [39], *Helianthus annus* [40], and *Cassia auriculata* [41], the absorbance peaks were between 400 and 450 nm. When compared with these plants and seaweeds, AgNPs synthesized from an aqueous extract of *Carica papaya* were active at the relatively lower wavelength.

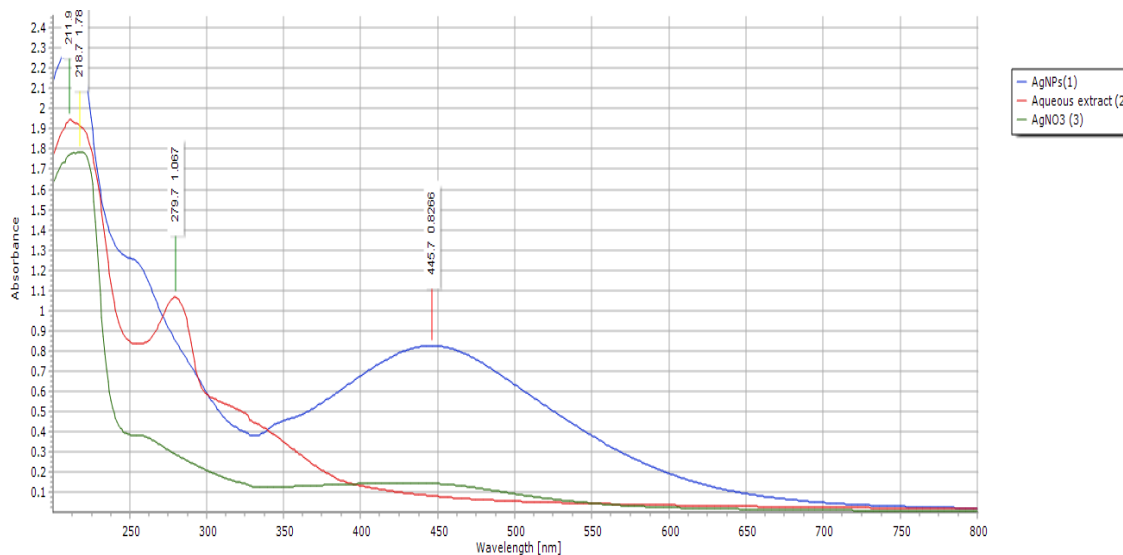


Fig. 2: UV-Vis spectral image of *Carica papaya* aqueous extract based synthesized silver nanoparticles

XRD spectral analysis

Insert fig. 3 shows the XRD pattern of *Carica papaya* aqueous extract based synthesized silver nanoparticles which exhibits strong and broad diffraction peaks at $2\theta = 24.9, 41.30$. The four other characteristic peaks showed in the case of Ag⁰ nanocomposite hydrogel indicates the face-centred cubic (fcc) structure. The diffraction peaks at $2\theta = 38.9, 44.62$ and 64.9 corresponding to

the reflections of crystal planes (1 1 1), (2 0 0) and (2 2 0) respectively. This face-centred cubic (fcc) structure indicates that Ag⁰ nanoparticles were dispersed in the *Carica papaya* aqueous extract based synthesized silver nanoparticles by the bioreduction method. Similar results were reported by [42, 43], these author's biosynthesized AgNPs using *Cinnanonum camphora* and marine algae (*Sargassum polycystum* and *Sargassum longifolium*) extract compound respectively.

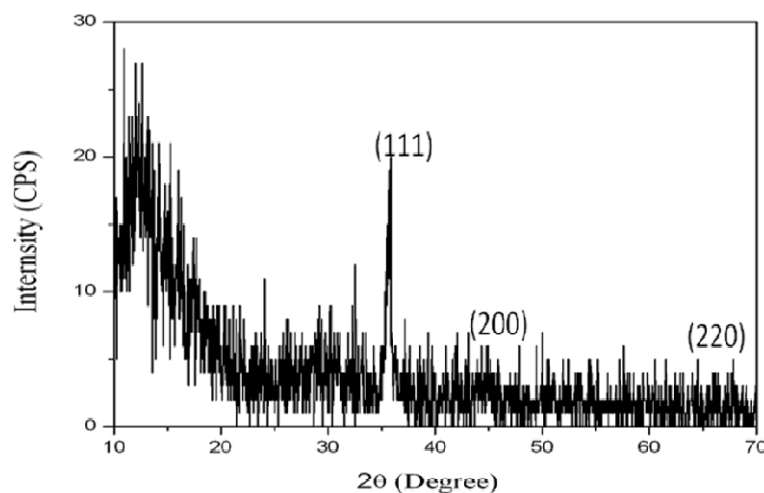


Fig. 3: XRD pattern of *Carica papaya* aqueous extract based synthesized silver nanoparticles

HR-TEM and FE-SEM analysis

HR-TEM analyses showed the particle size 10 nm with a spherical morphology and the SAED pattern reveals that nanoparticles are polydispersive in nature (fig. 4 A and B). The FESEM image of synthesized silver nanoparticles also showed the spherical shaped nanoparticles with the aggregation (fig. 5).

The EDX spectrum also confirms the bio reduction of silver nanoparticles with the peaks of silver and oxygen and this may be due to the surface plasmon resonance (SPR) (fig. 6). The FE-SEM image has clearly proved the bio reduction of Ag⁺ to Ag⁰ by the formation of spherical ovoid morphology. Overall the size and morphology distribution of AgNPs was found to be below 100 nm.

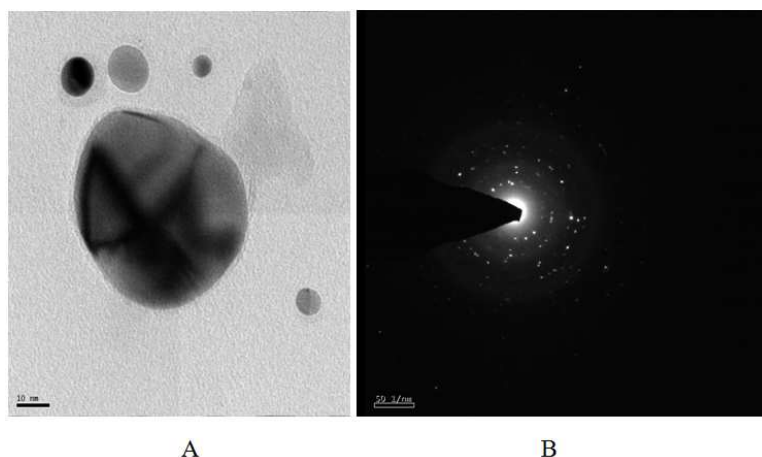


Fig. 4: A-HR-TEM image of *Carica papaya* aqueous extract based synthesized silver nanoparticles and the B-SAED pattern image of *Carica papaya* aqueous extract based synthesized silver nanoparticles

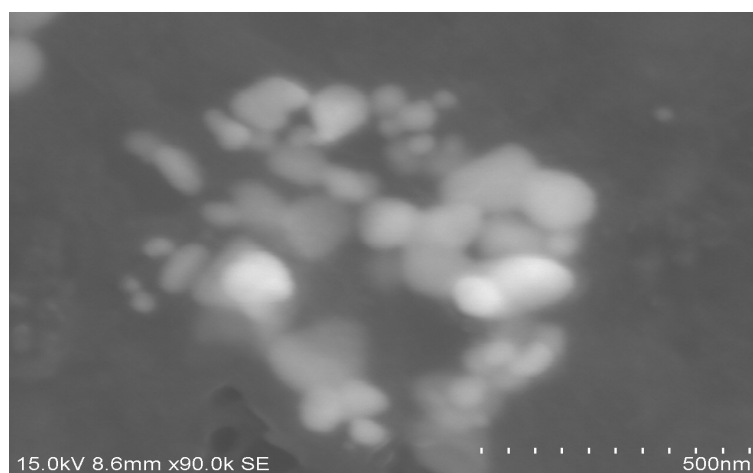


Fig. 5: FE-SEM image of *Carica papaya* aqueous extract based synthesized silver nanoparticles

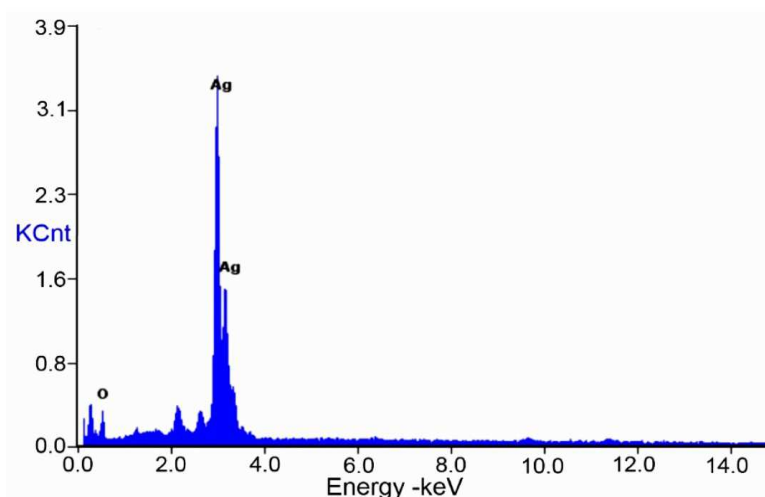


Fig. 6: EDX Characterization spectrum obtained for *Carica papaya* aqueous extract based synthesized silver nanoparticles

CONCLUSION

In this paper, we have reported cost-effective and eco-friendly bio-reducing method for synthesizing silver NPs using fresh leaves aqueous extract. These biologically synthesized silver NPs play a

crucial role in protecting our environment as green. UV-Vis spectroscopy revealed the surface plasmon property, while TEM and SEM with EDX image revealed the nano nature of the prepared sample. The structural analysis by XRD strongly suggests the formation of elemental silver NPs instead of their oxides in

biosynthesized NPs. The XRD structural analysis of AgNPs showed that they were crystalline in nature, which might be due to the presence of bio reduction of AgNPs. Therefore, the use of natural antioxidants for the synthesis of AgNPs seems to be a good alternative which could be attributed to its benign composition. The plant material responsible for the reduction and stabilization of nanoparticle needs further study including extraction and identification of the bioactive compounds presented in the extract.

AUTHORS CONTRIBUTIONS

All the author have contributed equally

CONFLICT OF INTERESTS

Declared none

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