PAPAYA FRUIT EXTRACT: A POTENT SOURCE FOR SYNTHESIS OF BIONANOPARTICLE

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ABSTRACT

The development of rapid and reliable processes for the synthesis of nano sized materials is of great importance in the field of nanotechnology. Synthesis of silver nanoparticles using fruit extracts have been reported. There is an increasing commercial demand for nanoparticles due to their wide applicability in various areas such as electronics, catalysis, chemistry, energy and medicine. Metallic nanoparticles are traditionally synthesized by wet chemical techniques, where the chemicals used are quite often toxic and flammable. In this work, we describe a cost effective and environment friendly technique for green synthesis of silver nanoparticles from 1mM AgNO₃ solution through the extract of papaya fruit as reducing as well as capping agent. Nanoparticles were characterized using UV is Absorption Spectroscopy and FTIR. Further these biologically synthesized nanoparticles were found to be highly toxic against different multi drug resistant human pathogens.

Key Words: Silver nanoparticles, Papaya extract, Antibacterial activities, UV-Vis Absorption Spectroscopy, FTIR

INTRODUCTION

Nanomaterials are at the leading edge of the rapidly developing field of nanotechnology. Then nanoparticles have diverse applications.¹ The field of nanotechnology is one of the most active areas of research in modern materials science. Nanoparticles exhibit completely new or improved properties based on specific characteristics such as size, distribution and morphology. New applications of nanoparticles and nanomaterials are emerging rapidly². Nano crystalline silver particles have found tremendous applications in the field of high sensitivity biomolecular detection and diagnostics, antimicrobials and therapeutics³. Catalysis and micro-electronics. However, there is still need for economic, commercially viable as well environmentally clean synthesis route to synthesize silver nanoparticles. AgNPs are currently being studied extensively because of several properties that can be used in science and technology. AgNP preparation is also being experimented upon and studied to find the mostcost-efficient and effective way to synthesize them. Certain metals, including copper and silver, have long been known to possess broad-spectrum antimicrobial activities in ionic or macro metallic forms.⁴ It has been known that in nature a variety of nanomaterials are synthesized by biological process. A number of approaches are available for the synthesis of silver nanoparticles for example, reduction in solutions, chemical and photochemical reactions in reverse micelles, thermal decomposition of silver compounds, radiation assisted, electrochemical, sonochemical, microwave assisted process and recently via green chemistry route.⁵-⁷ The use of environmentally benign materials like plant leaf extract⁶, bacteria⁷, fungi¹⁰ and enzymes¹¹ 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. Silver has long been recognized as having inhibitory effect on microbes present in medical and industrial process. The most important application of silver and silver nanoparticles is in medical industry such as topical ointments to prevent infection against burn and open wound. Most of the scientific research on nanosilver has investigated its effects on bacteria. Nanosilver is highly toxic to several strains of bacteria including so called gram positive bacteria such as Staphylococcus aureus and streptococcus pneumonia and gram negative bacteria including E.coli and Pseudomonas aeruginosa, which is responsible for infections that resist treatment by conventional antibiotics. 

Here in, we report for the synthesis of silver nanoparticles, reducing the silver ions present in the solution of silver nitrate by the cell free aqueous extract of papaya fruit. Further these biologically synthesized nanoparticles were found highly toxic against different multi drug resistant human pathogens.

**MATERIAL AND METHODS**

**Plant material and preparation of the extract**

Green unripe Papaya (Carica papaya) fruits were used to make the aqueous extract. Unripe papaya fruit weighing 25g were thoroughly washed in distilled water, dried, cut into fine pieces and were crushed into 100 ml sterile distilled water and filtered through Whatman No.1 filter paper (pore size 25 μm). The filtrate was further filtered through 0.6 μm sized filters. Similarly fully riped papaya fruits and green leaves were also used to prepare the extract.

**Synthesis of silver nanoparticles**

1mM aqueous solution of Silver nitrate (AgNO₃) was prepared and used for the synthesis of silver nanoparticles. 10 ml of papaya fruit extract was added into 90 ml of aqueous solution of 1 mM silver nitrate for reduction into Ag⁺ ions and kept at room temperature for 24 hours.

**UV-Vis Spectra analysis**

The reduction of pure Ag⁺ ions was monitored by measuring the UV-Vis spectrum of the reaction medium at 5 hours after diluting a small aliquot of the sample into distilled water. UV-Vis spectral analysis was done by using double beam UV-Vis spectrophotometer jasco company 600.

**FTIR analysis of dried biomass after bioreduction**

To remove any free biomass residue or compound that is not the capping ligand of the nanoparticles, the residual solution of 100 ml after reaction was centrifuged at 5000 rpm for 10 min and the resulting suspension was redispersed in 10 ml sterile distilled water. The centrifuging and redispersing process was repeated three times. Thereafter, the purified suspension was freeze dried to obtain dried powder. Finally, the dried nanoparticles were analyzed by FTIR 4100,made by jasco company (Japan).

**Antibacterial assays**

The antibacterial assays were done on human pathogenic Escherichia coli and Pseudomonas aeruginosa by standard disc diffusion method. Briefly Luria Bertani (LB) broth/agar medium was used to cultivate bacteria. Fresh overnight cultures of inoculum (100 μl) of each culture were spread on to LB agar plates. Sterile paper discs of 5mm diameter (containing 50mg/litre silver nanoparticles) along with four standard antibiotic containing discs were placed in each plate.

**RESULTS AND DISCUSSION**

It is well known that silver nanoparticles exhibit yellowish brown color in aqueous solution due to excitation of surface Plasmon vibrations in silver nanoparticles. As the papaya fruit extract was mixed in the aqueous solution of the silver ion complex, it started to change the color from watery to yellowish brown due to reduction of silver ion (Fig.1), which indicated formation of silver nanoparticles. The bio reduction of Ag⁺ ions in the solution was monitored in the aqueous component and the spectrum of the solution measured through UV spectrophotometer. It is
generally recognized that UV–Vis spectroscopy could be used to examine size- and shape-controlled nanoparticles in aqueous suspensions. Fig. 2 shows the UV-Vis spectra recorded from the reaction medium after 24 hours. Absorption spectra of silver nanoparticles formed in the reaction media has absorbance peak at 430 nm, broadening of peak indicated that the particles are polydispersed.\textsuperscript{14,15}

Fig. 1: Digital photographs of (A)1 mM AgNO\textsubscript{3} without papaya extract (B) papaya fruit extract (C)1Mm AgNO\textsubscript{3} with papaya extract after 24 hrs of incubation

FTIR analysis was used for the characterization of the extract and the resulting nanoparticles (Fig. 3). FTIR absorption spectra of water soluble extract before and after reduction of Ag ions are shown in Fig. 3. Absorbance bands in Fig. 3(a) (before bioreduction) are observed in the region of 500–2000 cm\textsuperscript{-1} are 1697, 1618, 1514, 1332, 1226 cm\textsuperscript{-1}. These absorbance bands are known to be associated with the stretching vibrations for –C C–C O, –C C– [(in-ring) aromatic], –C–C– [(in-ring) aromatic], C–O (esters, ethers) and C–O (polyols), respectively. In particular, the 1226 cm\textsuperscript{-1} band arises most probably from the C–O group of polyols such as hydroxyflavones and catechins. The total disappearance of this band after the bioreduction (Fig.3(b)) may be due to the fact that the polyols are mainly responsible for the reduction of Ag ions, whereby they themselves get oxidized to unsaturated carbonyl groups leading to a broad peak at 1650cm\textsuperscript{-1} (for reduction of Ag). Further the nanoparticles syntheses by green route are
found highly toxic against multi drug resistant human pathogenic bacteria at a concentration of 50 ppm. Silver nanoparticles exhibited antibacterial activities against E. coli and Pseudomonas aeruginosa as it showed a clear inhibition zone where as the standard antibiotics like Ampicillin, Tetracycline, Cefixime and Rifampicin does not shown any inhibition zone. Antibacterial effects of Ag nanoparticles obeyed a dual action mechanism of antibacterial activity, i.e., the bactericidal effect of Ag⁺ and membrane-disrupting effect of the polymer subunits.

Reduction of silver ions present in the aqueous solution of silver complex during the reaction with the ingredients present in the papaya fruit extract observed by the UV-Vis spectroscopy revealed the presence of silver nanoparticles may be correlated with the UV-Vis spectra. UV-Vis spectroscopy is well known to investigate shape and size controlled of nanoparticles. FTIR analysis confirmed that the Bioreduction of Ag⁺ ions to silver nanoparticles are due to the reduction by capping material of plant extract. The Silver nanoparticles synthesized via green route are highly toxic to multidrug resistant bacteria hence has a great potential in biomedical applications. The present study showed a simple, rapid and economical route to synthesized Silver nanoparticles.

CONCLUSION
In conclusion, the bio-reduction of aqueous Ag⁺ ions by the fruit extract of the papaya plant has been demonstrated. The reduction of the metal ions through leaf extracts leading to the formation of silver nanoparticles of fairly well-defined dimensions. But the capabilities of the other plant part such as fruit as a capping and reducing agent is not tested and not well defined. In the present study we found that fruits can be also good source for synthesis of silver nanoparticles. This green chemistry approach toward the synthesis of silver nanoparticles has many advantages such as, ease with which the process can be scaled up, economic viability, etc. Applications of such eco-friendly nanoparticles in bactericidal, wound healing and other medical and electronic applications, makes this method potentially exciting for the large-scale synthesis of other inorganic materials (nanomaterials).

REFERENCES
If all mankind were to disappear, the world would regenerate back to the rich state of equilibrium that existed ten thousand years ago. If insects were to vanish, the environment would collapse into chaos.

Edward O. Wilson