SYNTHESIS OF GOLD NANOPARTICLES USING CHICK PEA LEAF EXTRACT USING GREEN CHEMISTRY

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Metallic nanoparticles are traditionally synthesized by wet chemical techniques, where the chemicals used are quite often toxic and flammable. Hence, in the present study, we describe a cost effective and environment friendly technique for green synthesis of gold nanoparticles from 0.1 mM HAuCl₄ solution through the extract of chickpea leaf as a reducing agent as well as capping agent. Nanoparticles were characterized using UV–Vis absorption spectroscopy, SEM and TEM. The most important outcome of this work will be the development of value-added products from *Cicer arietinum* L. for biomedical and nanotechnology based industries.

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1. Introduction

One of the most active areas of research now a day is the field of nanotechnology. Nanoscience involves the study of materials on the nanoscale level between approximately 1 and 100 nm [1]. The demand of nanoparticles is increasing due to their wide application in various areas.

The use of gold nanoparticles dates back to the 16th century, for both medical and staining purposes. Thereafter, gold nanoparticles have found application in analytical methods, such as colorimetric techniques for the determination of heavy metal ions in aqueous solutions [2]. One of the other major applications of nanotechnology is in biomedicine. Gold nanoparticles have been investigated in diverse areas such as in vitro assays, *in vitro* and *in vivo* imaging, cancer therapy, and drug delivery [3]. Nanoparticles can be further designed into multi-functional delivery systems with a tumor-specific targeting moiety, therapeutic payload, and diagnostic tool (imaging or biochemical sensor) that enables monitoring of therapeutic efficacy [4, 5, 6].

Nanoparticles synthesized by chemical methods are energy intensive processes and a large amount of capital is involved. The use of hazardous chemicals makes the method non eco-friendly. Thus, there is a need for 'green chemistry' that includes a clean, nontoxic and environment-friendly method of nanoparticle synthesis [7]. Moreover nanoparticles produced by these methods exhibit aggregation with time, there by compromising with the size factor upon storage [8, 9]. As an alternative to conventional methods, biological methods are considered safe and ecologically sound for the nanomaterial fabrication [10].

Nanoparticles have tremendous application including their use in highly sensitive diagnostic assays [11], thermal ablation and radiotherapy enhancement, respectively [12, 13], in drug and gene delivery [14] as antimicrobial agents [15] and in wastewater treatment [16].

Green method was used for the synthesis of gold nanoparticles [17] using the aqueous extract of rose petals. Plant extract mediated synthesis of silver and gold nanoparticles and its antibacterial activity against clinically isolated pathogens [15] was also studied.

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In present study synthesis of gold nanoparticles was done through a simple green route using the chick pea leaf extract as reducing as well as capping agent. The synthesis was confirmed using UV–Vis spectroscopy, TEM and SEM analysis respectively.

2. Materials and method

2.1 Plant Material and preparation of extract

Green chick pea leaves were used to make the aqueous extract. Leaves weighing 5g were thoroughly washed in distilled water, cut into fine pieces and were boiled into 100 ml sterile distilled water and filtered through Whatmann No.1 filter paper (pore size 25 μ m).

2.2 Preparation of Reagent

HAuCl₄ solution was prepared at concentration of 10⁻⁴M in Milli Q water.

2.3 Synthesis of nanoparticles:

5 ml of aqueous plant extract was added to 40 ml of 0.1mM HAuCl₄ solution and kept at room temperature. After 24 hrs., 2ml of the sample was taken in an eppendorf and centrifuged at 10000 rpm for 10 minutes at 15 °C. Supernatant was discarded and weight of the pellet was recorded and was given two washings at 10000 rpm for 2-3 minutes at 15°C. The pellet obtained was finally dissolved in 100µl of distilled water and vortexed.

2.4 Change in colour

Gold nanoparticles exhibit purple color in aqueous solution due to excitation of surface plasmon vibrations in gold nanoparticles.



Fig. 1(A-C) color change during formation of nanoparticles

2.5 UV-Vis spectra analysis:

The reduction of pure Au+ ions was monitored by measuring the UV-Vis spectrum of the sample after diluting a small aliquot of the sample into Milli Q water. UV-Vis spectra analysis was done by using UV-Vis spectrophotometer, UV-2450 (Shimadzu) and nanodrop. The sample was scanned for determining the wavelength which shows maximum absorbance. Milli Q water was taken as reference.



Fig. 2. UV-Vis absorption spectra of gold nanoparticles synthesized by chick pea leaf extract

2.6 Transmittance Electron Microscopic (TEM) analysis of Gold Nanoparticles:

Samples for Transmittance Electron Microscopic (TEM) analysis were prepared by drop coating biologically synthesized gold nanoparticles on to carbon coated copper TEM grids. The films on the TEM grid were allowed to stand for 2 minutes, following which extra solution was removed using a blotting paper and grid allowed to dry prior to measurement. TEM analysis was done using Joel TEM machine at Plant Virology Unit, Division of Plant Pathology, Indian Agricultural Research Institute, New Delhi. TEM analysis is done to find out the shape and size of gold nanoparticles.



Fig 3 (A-D): TEM analysis of Gold Nanoparticles

2.7 Scanning electron microscopy (SEM) of Gold Nanoparticles

The gold nanoparticles were characterized with scanning electron microscopy. The sample was prepared by placing a drop of very fine suspension of nanoparticles (in water) over a cut glass and allowed to air dry. The glass was placed over an aluminium stub, with previously pasted carbon tape. The stub was gold coated using a Gold coater. To ensure high conductivity, silver paint was applied at the periphery of the stub SEM analysis was done at Advanced Instrumentation Research Facility (AIRF), JNU, Delhi.



 100 nm
 EHT = 20.00 kV
 Date :22 Jun 2010
 AIRF, JNU
 ZEINN

 WD = 7.5 mm
 Mag = 255.84 K X
 AIRF, JNU
 ZEINN

B

Fig 4 (A-B): SEM analysis of Gold Nanoparticles

3. Result and discussion

The excitation of surface plasmon vibrations of gold nanoparticles exhibits chamge in color which makes it easy to follow the formation of gold nanoparticles in the aqueous solution [18].

As the plant extract (Fig. 1 a) was mixed in the aqueous solution of the gold ion (Fig. 1 b) complex, it started to change the color from yellowish to purple due to reduction of gold ion; which indicated formation of gold nanoparticles. Color changes from yellowish to purple within 45 minutes (Fig. 1 c). After 24 hours dark purple color was obtained, which indicated that the total reduction of gold ion and formation of gold nanoparticles. It is generally recognized that UV–Vis spectroscopy is used to examine size and shape-controlled nanoparticles in aqueous suspensions. Absorption spectra of gold nanoparticles formed in the reaction media has absorbance peak at 550 nm (Fig. 2), broadening of peak indicated that the particles are polydispersed. TEM analysis of gold nanoparticles formed by the chickpea leaf extract is done and found that nanoparticles are of spherical, pentagonal and triangle shape. TEM is done at the magnification of 200000X having a scale of 50 nm. In which particles are found in different size 30 nm to 80 nm (Fig 3 A-D). The

SEM image (Fig. 4 A and B) showed the high density gold nanoparticles synthesized by the chick pea leaf extract. SEM image at the magnificence of 25000 showed different shapes of nanoparticles.

4. Conclusions

The reduction of the metal ions through leaf extracts leading to the formation of gold nanoparticles of fairly well-defined dimensions. This eliminates the need of toxic chemicals for the synthesis of nanoparticles. Green chemistry used includes a clean, nontoxic and environmentfriendly method of nanoparticle synthesis. Hence, it can be concluded that other than toxic chemical synthesis, plants (chick pea) also have ability to perform synthesis of nanoparticles which is cost effective and environment friendly technique. These nanoparticles which are synthesized using green chemistry can be used in the future for a variety of applications.

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