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Effective utilization of biological catalyst for synthesis gold (Au) nanoparticles

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ABSTRACT

Gold nanoparticles were prepared by reduction of Chloroauric acid ($HAuCl_4$) using enzymes of Glycyrrhiza Glabra root extract as a reducing agent and also used as stabilizer. UV–visible spectroscopy was used for quantification of Gold nanoparticle synthesis. The synthesized gold nanoparticles were characterized with Transmission electron microscopy (TEM), X-ray Diffraction (XRD) and Fourier transform Infrared Spectroscopy (FTIR).The results of TEM, XRD, and UV–vis absorption spectra show that the Gold nanoparticles have a narrow size distribution.

Introduction

Biomolecules conjugated to metal nanoparticles (NPs) attract substantial recent interest in the rapidly developing area of nanobiotechnology [1]. The similar dimensions of metal NPs and biomolecules such as enzymes, antigens–antibodies or nucleic acids suggest that the integration of biomolecules with metal particles could yield new hybrid systems that combine the unique optical or electronic properties of metallic NPs with the selective and catalytic functions of biomolecules.

Specifically, the integration of enzymes with metallic NPs could lead to new NP–enzyme biocatalytic conjugates where the metallic NPs control, or direct, the enzyme functions. Alternatively, the enzymes activities may regulate the optical or electronic features of the NPs, thus enabling the development of new bioelectronic or sensor systems. Biomolecule–metal NP hybrids are used as tags for optical sensing, and as catalytic labels for the amplified electrical, electrochemical or microgravimetric analysis of biorecognition events [2]. Nanoparticles are often referred to as clusters, nanospheres, nanorods and nanocups are just a few of the shapes at the small end of the size ranges from 1 to 100nm. Nanoparticles exhibit a number of special properties relative to bulk material and often have unique visible properties because they are small enough to confine their electrons and produce quantum effects [3].

Experimental

Glycyrrhiza Glabra root was washed several times with de-ionized water. 50 g of the root was finely cut and stirred with 150 mL de-ionized water at 300 K for 5 min and filtered to get the extract. The filtrate is used as reducing agent and stabilizer. To a vigorously stirred 30 mL aqueous solution of $HAuCl_4 \cdot 3H_2O$, Chloroauric acid (50 mg dissolved in 240 mL water) were procured from Sigma–Aldrich. 7 mL extract is added to a vigorously stirred 100 mL aqueous solution of $HAuCl_4 \cdot 3H_2O$ (1×10^{-3} M) and stirring continued for 10 min. Reduction takes place slowly at 300 K and is complete in 30 min as shown by stable dark purple colour of the solution giving colloid. It is found to be stable for more than 10 months, showing no precipitation or colour change.

Spectra analysis

The UV–vis spectra were recorded using, ELCO 150 UV-visible spectrophotometer with samples in quartz cuvette. X-ray diffraction pattern of dry nanoparticle powder was obtained using Siemens D5005 X-ray diffractometer with CuKα radiation (λ = 0.1542 nm). The FTIR spectra were obtained on a Nicolet 5700 FTIR instrument with the sample as KBr pellets. The morphology of the nanoparticles was analysed using the high-resolution images obtained with a JEOL 3010 transmission electron microscope.

XRD analysis of silver nanoparticles

The Gold nanoparticle solution thus obtained was purified by repeated centrifugation at 10000 rpm for 20 min followed by re-dispersion of the pellet of Gold nanoparticles into 10 ml of deionized Toluene. After freeze drying of the purified Gold particles, the structure and composition were analyzed by XRD (Rigaku RINT 2100 series). The dried mixture of Gold nanoparticles was collected for the determination of the formation of Au nanoparticles by an X'Pert Pro x-ray diffractometer operated at a voltage of 40 kV and a current of 30 mA with Cu Kα radiation in a θ- 2 θ configurations [4]. The crystallite domain size was calculated from the width of the XRD peaks, assuming that they are free from non-uniform strains, using the Scherrer formula.

$D = 0.94 \lambda / \beta \cos \theta \rightarrow (1)$

where D is the average crystallite domain size perpendicular to the reflecting planes, λ is the X-ray wavelength, β is the full width at half maximum (FWHM), and θ is the diffraction angle.

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To eliminate additional instrumental broadening the FWHM was corrected, using the FWHM from a large grained Si sample.

β corrected = $(FWHM^2sample - FWHM^2si)1/2 \rightarrow (2)$

This modified formula is valid only when the crystallite size is smaller than 100 nm [5]

Results and discussion

It is reported that aqueous extract of Glycyrrhiza Glabra root extract is composed of a variety of The herb contains glycyrrhizin, glycyrrhetinic acid, flavonoids, asparagine, iso-flavonoids, and chalcones [6] and these may be responsible for reduction of metal ions and efficient stabilization of nanoparticles.

UV–vis, and TEM analysis

UV–vis spectroscopy is an important technique to ascertain the formation and stability of metal nanoparticles in aqueous solution Fig. 1. Figure 1a shows the UV–vis spectra of the Gold.. Colour of Gold colloid is attributed to surface plasmon resonance (SPR) arising due to the collective oscillation of free conduction electrons induced by an interacting electromagnetic field [7]. SPR bands of the colloids are centered at 540 nm. The bands are broad and the intensity increases indicating increase in production of nanoparticles. The TEM images of the colloids are shown in (Fig. 2).

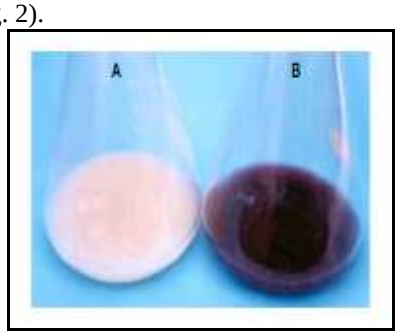

Fig. 1 Au nanoparticle formatiom

Colloids consist mainly of large nanoparticles having nearly spherical shape particles of size 4–20 nm. It is clear from the images (Fig. 2) that the particles in colloid are well-dispersed with a more uniform size 7 nm (average). A similar TEM image was observed for polydispersed Gold nanoparticles by Lee et al. [8] in chemical reduction and Huang et al. [9] in biosynthesis. It is evident from the TEM image that the shape and size of the silver nanoparticles could be controlled by adjusting the reaction mixture. The high resolution TEM images (Fig. 2) indicate good crystallinity of the nanoparticles.

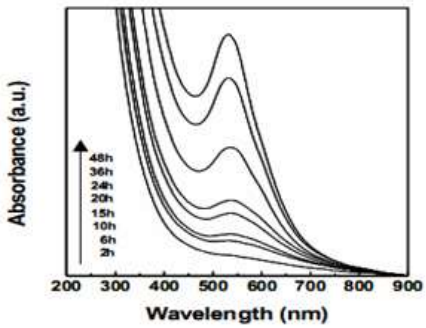

Fig. 1a. UV-VIS Spectra of Au nanoparticles

XRD and FTIR studies

The crystalline nature of Au nanoparticles was confirmed from the X-ray diffraction analysis. Fig 3. shows the XRD pattern of Au nanoparticles obtained using mushroom extract. The diffraction peaks at 38.1°, 44.5° and 64.6° correspond to the (1 1 1), (2 0 0) and (2 2 0) facets [10], [11], [12], [13] and [14] of the face centered cubic crystal structure, respectively. The peak corresponding to the (1 1 1) plane is more intense than the other planes. The ratio between the intensity of the (2 0 0) and (1 1 1) diffraction peaks is much lower than the usual value (0.52) suggesting that the (1 1 1) plane is the predominant orientation [15]. The width of the (1 1 1) peak is employed to calculate the average crystallite size using Scherrer equation. It is found that the calculated average size is ~7 nm that matches with the particle size obtained from TEM image of Au nanoparticles.

Fig. 2 TEM analysis of Au Nanoparticles

FTIR measurement was carried out to identify the possible biomolecules in Glycyrrhiza Glabra root responsible for capping leading to efficient stabilization of the Gold nanoparticles. Prominent IR bands (Fig.4) are observed at 2914, 2847, 1708, 1601, 1464, 1376, 1008, 880, 716 and 469 cm^{-1}. Most of the IR bands are characteristic of flavonoids and tepenoids present in the root. The sharp bands at 2914 and 2847 cm^{-1} arise from C–H stretching modes. The medium intense bands at 1708 and 1601 cm−1 are assigned to the stretching vibrations of C=O; and C=C, respectively [16] and [17]. The absorption bands located at 1376 and 1008 cm^{-1} may be attributed to –C–O and –C–O–C stretching modes. The medium intense band at 1464 cm−1arises from the C–N stretching mode of the aromatic amine group [18]. The vibrational bands corresponding to bonds such as –C=C, –C=O, –C–O, –C–O–C and –C–N are derived from the water soluble compounds such as flavonoids, terpenoids and thiamine present in Glycyrrhiza Glabra root. Hence, it may be assumed that these biomolecules are responsible for capping and efficient stabilization. The presence of reducing sugars in the extract could be responsible for the reduction of gold ions and formation of the nanoparticles [19, 20]. This rapid and environmentally benign method is a faster synthesis comparable to chemical reduction methods.

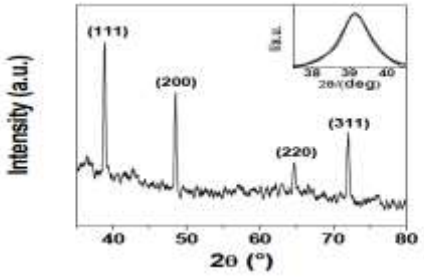

Figure3: XRD analysis of Au nanoparticles

Conclusions

Gold nanoparticles of are synthesized from silver precursor using aqueous extract of Glycyrrhiza Glabra leaf at room

temperature. The reduction is found to be accelerated by the change reaction mixture. The colloid obtained by rapid reduction is found to consist of well-dispersed nearly spherical particles having size around 7 nm. The nanoparticles are found to be highly crystalline as evidenced by the peaks in the XRD pattern corresponding to Bragg reflections from the (1 1 1), (2 0 0), (2 2 0), (3 1 1) and (2 2 2) planes of the fcc structure, and clear lattice fringes in the high resolution TEM image. The biomolecules responsible for efficient stabilization of the silver colloid is suggested by studying the FTIR spectrum.

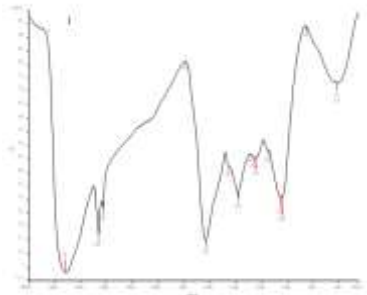

Fig. 4. FTIR analysis of Au Nanoparticles

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