Crecimiento de núcleos de nanovarillas de oro para aplicaciones de diagnóstico

Seed-mediated Growth of Gold Nanorods for Diagnostic Applications

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RESUMEN

En el siguiente trabajo se describen las propiedades de nanovarillas de oro sintetizadas utilizando un método de semillas. La sal de oro primero se reduce empleando un agente reductor fuerte, el $NaBH_4$, para producir las partículas de semillas. En presencia de bromuro de cetiltrimetilamonio (CTAB), empleado como surfactante, se produce una reducción de la sal del metal con un agente reductor más débil (ácido ascórbico) para formar las nanovarillas de oro. Se investigan varios parámetros: concentración de oro, concentración de CTAB y concentración de $AgNO_3$. La morfología y distribución de las partículas se caracterizó mediante microscopia electrónica de transmisión (TEM) y mediante dispersión dinámica de luz (DLS). Se empleó un espectrofotómetro UV-Visible (UV-Vis) para observar las propiedades de dispersión de las muestras. El resultado muestra que la concentración de oro no tiene un impacto significativo en la formación de nanovarillas. Además, se forman nanoesferas de oro con desigual tamaño y distribución para todos los casos, como muestran los ensayos mediante TEM y DLS. Por otra parte, la forma alargada de algunos de ellos podría ser causada por una insuficiencia en la concentración de CTAB para ayudar en la formación de nanovarillas de oro y por tanto favorecen la formación de nanoesferas de oro. El papel de CTAB es dirigir el crecimiento mediante el bloqueo del eje más extenso de la cara del cristal y promover el crecimiento de oro en el eje corto para formar las nanovarillas. Se observa que se requiere CTAB 1,0 M para formar las nanovarillas. Cuando la cantidad de CTAB es mayor, las partículas se transforman hacia la formación de rectángulos. El análisis mediante UV-Vis muestra que las nanovarillas presentan señales alrededor de los 540 nm y 720. El estudio sobre el efecto del $AgNO_3$ muestra que al aumentar la concentración de $AgNO_3$ aumenta el largo de las partículas. Sin embargo, la forma de bastón se transforma en una forma rectangular utilizando una concentración elevada de $AgNO_3$. La formación de nanovarillas es altamente dependiente del CTAB y de la concentración de $AgNO_3$ a temperatura ambiente, pero también depende de la concentración de los precursores de oro en la solución de crecimiento. El largo de las nanovarillas de oro puede ajustarse variando los parámetros de la síntesis.

Palabras claves: Nanopartículas de oro; Nanovarillas; Crecimiento por semillas; Diagnóstico.

ABSTRACT

This work describes properties of gold nanorods synthesized using a seed-mediated method. Gold salt is first reduced using a strong reducing agent which is $NaBH_4$ to produce seed particles. In the presence of cetyltrimethylammonium bromide (CTAB) as a surfactant, subsequent reduction of more metal salt with a weak reducing agent (ascorbic acid) formed gold nanorods. Several parameters are investigated; gold concentration, CTAB concentration and $AgNO_3$ concentration. Morphology and distribution of particles are characterized using transmission electron microscope (TEM) and dynamic light scattering (DLS). UV-Visible (UV-Vis) spectrophotometer is used to observe scattering properties of the samples. The result shows that the amount of gold concentration does not have significant impact on nanorods formation. Gold nanospheres with uneven size and shape distribution are formed for all cases as observes using TEM and further proves using DLS. Moreover, some of them have elongated shape that could be caused by insufficient CTAB concentration to aid in the formation of gold nanorods and thus favour the formation of gold nanospheres. The role of CTAB is to direct the growth by blocking the long axis crystal faces and promoting gold growth on the short-axis faces to form nanorods. It is observed 1.0 M

CTAB is required to form nanorods. When the amount of CTAB is further increased, the shapes transform into rectangle shape. UV-Vis analysis shows that nanorods have peaks at around 540 nm and 720 nm. The study on the effect of $AgNO_3$ shows that the aspect ratio increases with increasing $AgNO_3$ amount. However, the rod shape transform into rectangle shape at high concentration of $AgNO_3$. The formation of nanorods is highly dependent on CTAB and $AgNO_3$ concentration at room temperature, but least dependent on the gold precursor concentration in the growth solution. The aspect ratio of gold nanorods can be tuned by changing synthesis parameters.

Keywords: Gold nanoparticles; Nanorods; Seed-mediated growth; Diagnostic.

INTRODUCTION

The field of nanotechnology is undergoing speedy developments in many fronts. It is a field of science that deals with the control and manipulation of particles at the length scale between 1 to 100 nm and the exploitation of novel properties and phenomena developed at that scale. The creation of new properties in nano-scale is resulted from three size-related factors which are quantum confinement, large surface to volume ratio and force or energy changes landscape with the shrinking scale (1). In addition, a wide variety of materials have been formed into nanoparticles (AuNPs). Besides the most common spherical shape (AuNS), various shapes such as cube, rod, ring, wire, tetrapod, and triangular prism type of nanoparticles have been synthesized. As the nanoparticle synthesis technology has been advanced, the application of nanoparticle has been extended further into various fields especially in biomedical applications such as X-ray contrast agents, diagnosis of diseases, photothermal therapy, drug delivery and biological sensors (2).

Gold nanoparticles (AuNPs) have attracted significant interest among scientists for generations as a novel platform for biotechnology and biomedicine due to their amazing chemical and physical properties at nanoscale level. In this work, gold nanorods (AuNRs) were synthesized using seed-mediated approach. AuNRs differ from spheres since they have a particular aspect ratio in which the material is elongated along a single dimension, keeping the other two dimensions approximately equal. It is essential to limit growth direction to a single axis by using surfactant in order to synthesize AuNRs (3). AuNRs have two peaks in their absorbance spectra that correspond to plasmon resonances: one at approximately 520 nm from plasmon oscillations in the shorter transverse direction and a second at longer wavelengths (typically between 700 nm and 1300 nm) from longitudinal oscillations (4). Apart from the obvious advantages of AuNP that do not photobleach or blink, chemically inert and biologically compatible, AuNRs added extra dimension as an attractive candidates for biomedical applications since their position of the longer wavelength plasmon peak can now be tuned to near infrared wavelengths which penetrate deep into cells and tissue by varying the AuNRs aspect ratio.

In this work, AuNRs have been synthesized using a seed-mediated approach. The effect of synthesis parameters on the properties of AuNRs formation was studied. It was observed that the formation of AuNRs were highly dependent on CTAB and silver nitrate concentrations at room temperature, but least dependent on concentration of gold in the growth solution.

MATERIALS AND METHODS

Seed mediated approach was used to synthesis gold nanorods (AuNRs). Seed solution was prepared by mixing Hexadecyltrimethylammonium bromide solution (CTAB) (5 mL, 0.20 M) with 5.0 mL of 0.00050 M gold (III) chloride. 30 mL of ice-cold 0.010 M sodium borohdyride was added to the stirred solution which resulted in the formation of a brownish yellow solution. The seed solution was vigorously stirred for 2 min and kept at

25°C. The growth solution was prepared by dissolving CTAB (5 mL, 0.20 M) in 20 mL of 0.0040 M silver nitrate solution at 25°C. To this solution, 5.0 mL of 0.0010 M gold (III) chloride was added followed by the addition of 10mL 0.0788 M ascorbic acid. Ascorbic acid as a mild reducing agent changed the growth solution from dark yellow to colorless. After that, 50mL of the seed solution was added into the growth solution. This solution was maintained at 25°C without stirring overnight. The color of the solution gradually changed within 10-20 min. AuNPs were then washed three times by centrifugation at 10000 rpm with deionised water (DI) to remove excess CTAB and re-dispersed in DI water. Several parameters were studied including the effect of gold concentration in the growth solution (0.0005M-0.003M), CTAB concentration (1.0M-1.5M), silver concentration (0.003M-0.007M) and reaction temperature (25-70°C).

Shape, size and distribution of the AuNPs were observed using Philips 420T transmission electron microscope (TEM). Absorbance properties of AuNPs were analyzed using the ultraviolet-visible spectrophotometer (UV-Vis). Particle size distribution AuNPs was determined by using Zetasizer NanoZS.

RESULTS AND DISCUSSION

Gold chloride concentration of growth solutions was varied; 0.0005M, 0.001M, and 0.002 M and 0.003 M. From the result of UV-visible absorbance spectra (Fig.1), obviously there was only one absorbance peak at ~540 nm for all cases indicates AuNSs were formed because AuNRs should exhibit two absorbance peaks as opposed to only one peak at ~520 nm for AuNSs. However, for 0.003 M $HAuCl_4$, the absorbance peak cannot be seen clearly because the concentration of solution is too dilute and hence affect the absorption of light.

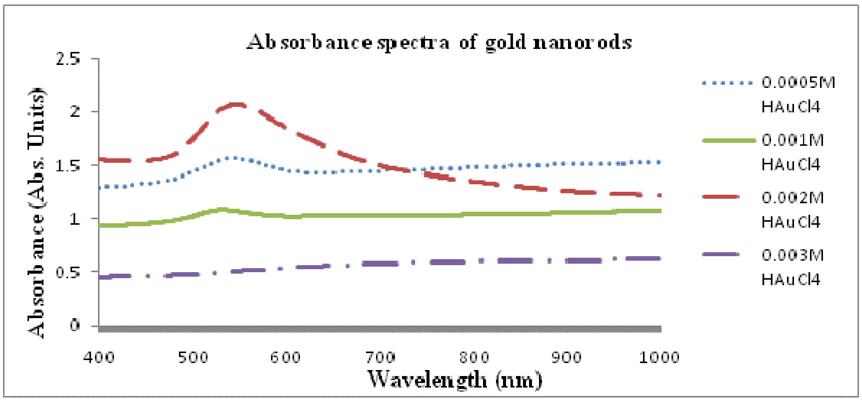

Fig.1. UV-Vis spectra of AuNRs synthesized using different gold chloride concentration.

Transmission electron microscopy (TEM) images (Fig. 2) further confirmed that all gold concentration did not form AuNRs. Also, the shapes of AuNSs were found to be slightly elongated rather than entirely spheres. Therefore, the amount of CTAB added was expected to be insufficient in inducing AuNRs formation. Moreover, bromide or silver bromide could be the initial species that deposit on the seed through chemisorb on gold crystal faces. However, the affinity of bromide or silver bromide for gold is modest. Hence, high concentrations of bromide will be required to drive the ions onto the gold surface. The low amount of CTAB will lower the Br concentration and hence favor spheres (5). Hence, the effect of CTAB concentration was subsequently studied.

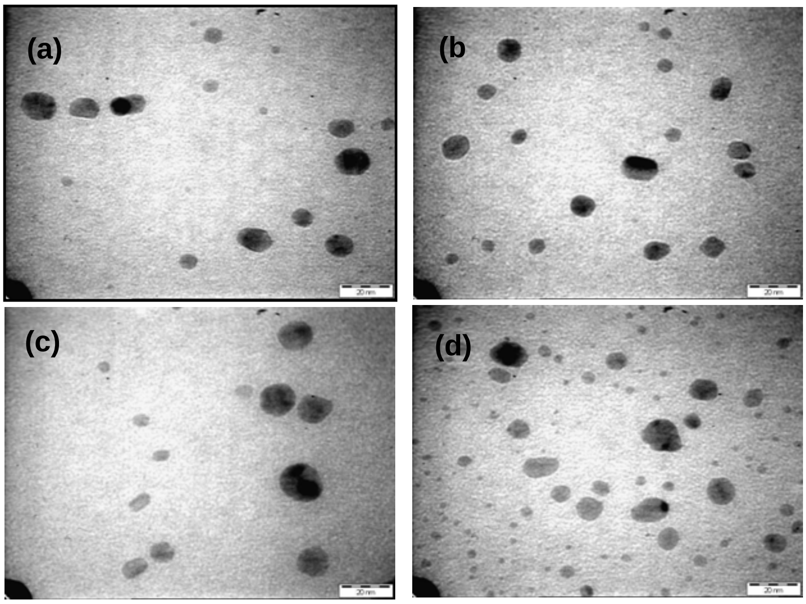

Fig.2. TEM images of AuNPs synthesized with varying $HAuCl_4$ concentration in growth solution; (a) 0.0005 M, (b) 0.001 M, (c) 0.002 M and (d) 0.003 M.

CTAB concentration was studied from 1.0 M, 1.5 M and 2.0 M. From UV-Vis absorbance spectra (Fig. 3), two absorbance peaks were observed for all cases, which was the characteristic of AuNRs. One of the absorbance peaks is at ~520 nm and the other at ~700-1300 nm. Besides, it can be seen that longitudinal peaks and transverse peaks shifted to longer wavelength first and then followed by shifting to shorter wavelength. Longitudinal peak was attributed to the length of AuNRs, while transverse peak represented the width of AuNRs. In other words, length and width of AuNRs increased when CTAB concentration increased from 1 M to 1.5 M. However, as CTAB concentration increased to 2 M, both length and width of AuNRs decreased. This was due to the shape changes of the rod shape. Additionally, the intensities of absorbance increased with increasing CTAB concentration since the concentration of AuNRs solution to absorb light increased.

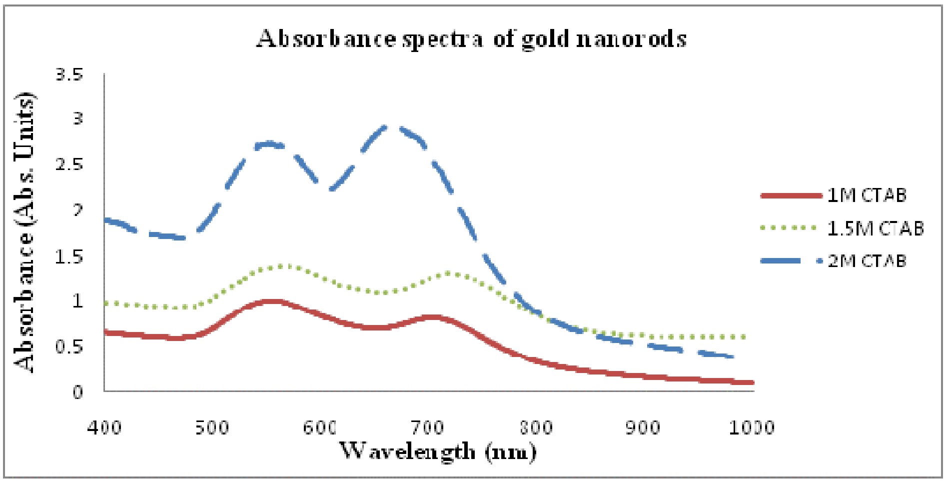

Fig.3. Absorbance spectra of AuNRs with various CTAB concentrations.

Formations of AuNRs for all cases were confirmed by TEM observation (Fig.4). This proved that a high concentration of CTAB was required for AuNRs formation. Nonetheless, a small amount of AuNSs was present as a byproduct. The role of CTAB was to aid in anisotropic growth of AuNRs by preferentially bound to {110} side face of gold. As a result, gold could only grow along front face {100} direction since side face was blocked by CTAB. From the TEM images in Fig. 4, it was observed that the shape of AuNRs formed with 1.5 M and 2 M CTAB were slightly different compared to AuNRs formed using 1 M CTAB. With 1 M CTAB, well-shaped of AuNRs were produced (Fig. 4 (a)). While with 1.5 M and 2 M CTAB, the ends of AuNRs becomes flattened rather than rounded. In other words, the shapes of rods had transformed into dogbones. Also, a large number of amorphous and unshape particles are present (Figs. 4 (b) and (c)). The result could be caused by the excessive amount of CTAB on the {110} side face that prevented more gold atoms from depositing on this plane, and more gold atoms deposited on the {100} front face with some preference. This could lead to gold nanodogbones formation. This condition was also observed in the studies conducted by Wang et al. (6) and Gou and Murphy (7).

Fig. 5 shows the particles size distribution of AuNRs with different CTAB concentration using Zetasizer. It can be seen that, size distribution for all samples have two distinct peaks. The first peak attributes to the width of AuNRs while the second peak represents the length of AuNRs. These results further confirmed that width and length of AuNRs increase with increasing CTAB concentration from 1 M to 1.5 M. On the other hand, width and length of AuNRs decrease as concentration of CTAB increase to 2 M.

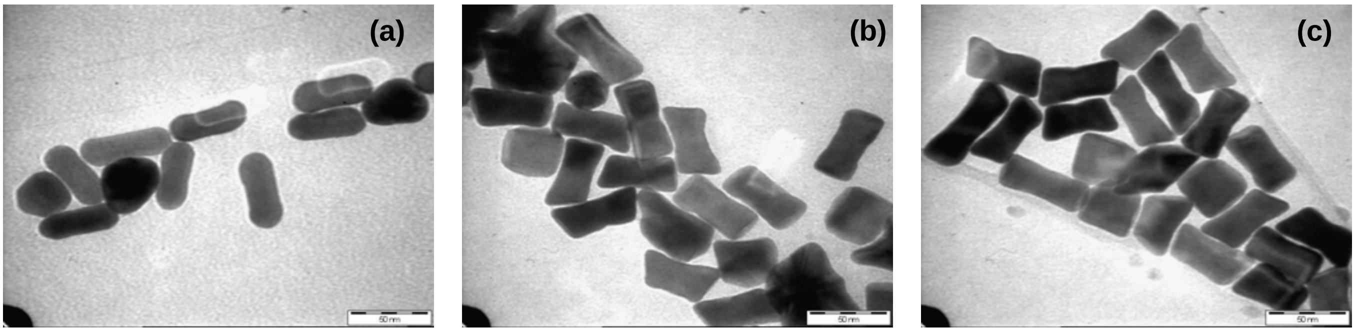

Fig.4. TEM images of AuNRs synthesized with varying CTAB concentration; (a) 1 M, (b) 1.5 M and (c) 2 M CTAB.

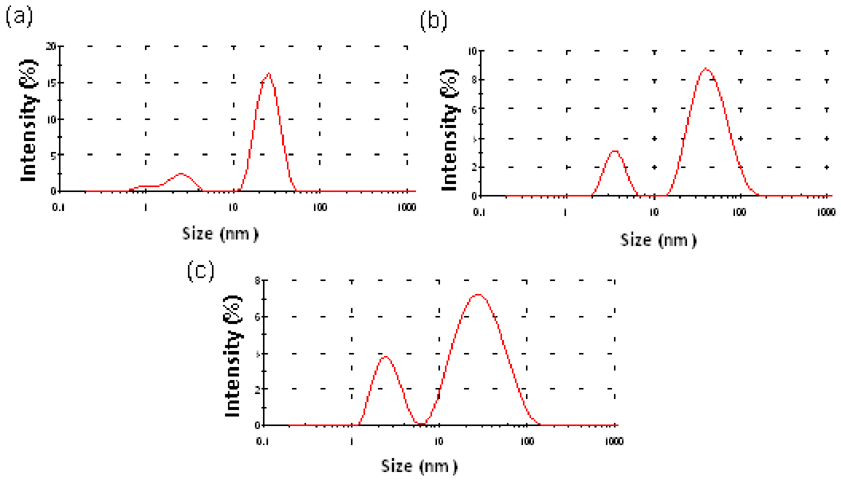

Fig.5. Size distribution by intensity for (a) 1 M, (b) 1.5 M and (c) 2 M CTAB.

The effect of silver concentration was studied by varying the concentration of silver nitrate; 0.003 M, 0.004 M, 0.005 M and 0.007 M. UV-Vis spectra for these four samples showed the presence of AuNRs with two absorbance peaks (Fig. 6). It can be seen that, both longitudinal and transverse peaks shifted to longer wavelength first, subsequently shifted to shorter wavelength and finally shifted to longer wavelength. This indicates that both length and width of AuNRs increase with increasing concentration of silver nitrate except at 0.0005 M of silver nitrate due to the shape changes and causes length and width of AuNRs reduce. The role played by silver nitrate is quite similar with CTAB molecule. Both of them have preferential binding on {110} side face of rods. Thus, the addition of silver content was used to grow and stabilize AuNRs to a desired aspect ratio. When silver nitrate concentration increases, more silver deposited on AuNRs {110} side face, resulted in AuNRs with higher aspect ratio (8).

Fig. 7 shows the TEM images of AuNRs synthesized by using different silver nitrate concentration. Well-shaped of AuNRs were produced with 0.004 M silver nitrate (Fig. 7 (b)). Meanwhile, AuNRs with 0.003 M, 0.005 M and 0.007 M of silver nitrate exhibit dogbones shape (Figs. 7 (a), (c) and (d)). Also, a higher concentration of silver nitrate resulted in lower rod yield.

Fig. 8 shows the particles size distribution of AuNRs with different silver nitrate concentration using Zetasizer. It can be seen that both peaks that represent width and length of AuNRs increased when silver nitrate concentration increased except at 0.005 M of silver nitrate, both length and width of AuNRs decreased.

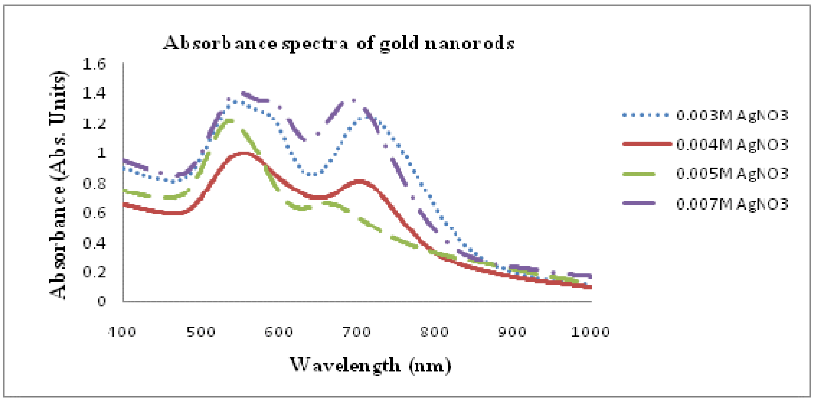

Fig.6. UV-Vis spectra of AuNRs synthesized with various silver nitrate concentrations.

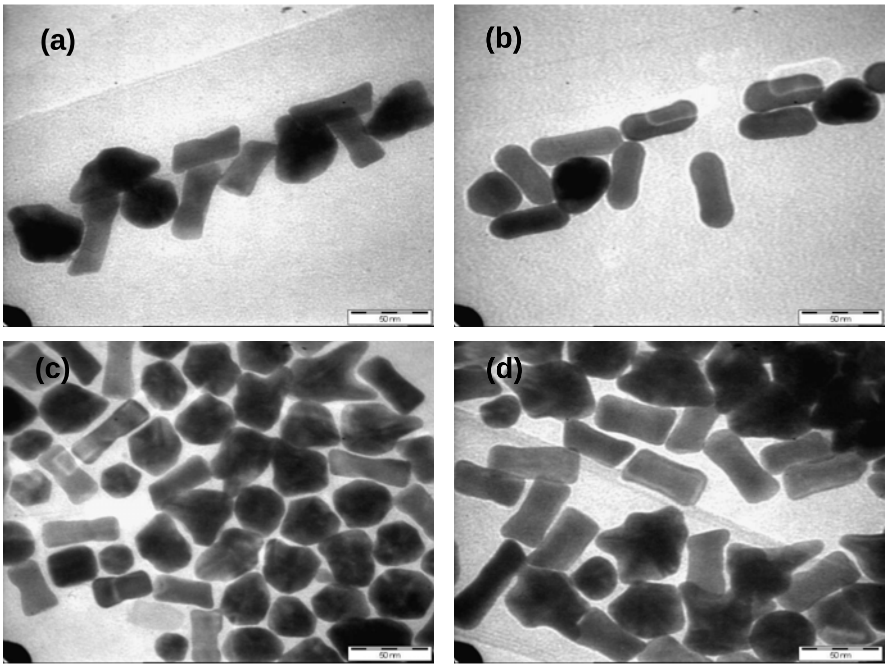

Fig.7. TEM images of AuNRs synthesized with varying silver nitrate concentration; (a) 0.003 M, (b) 0.004 M (c) 0.005 M and (d) 0.007 M.

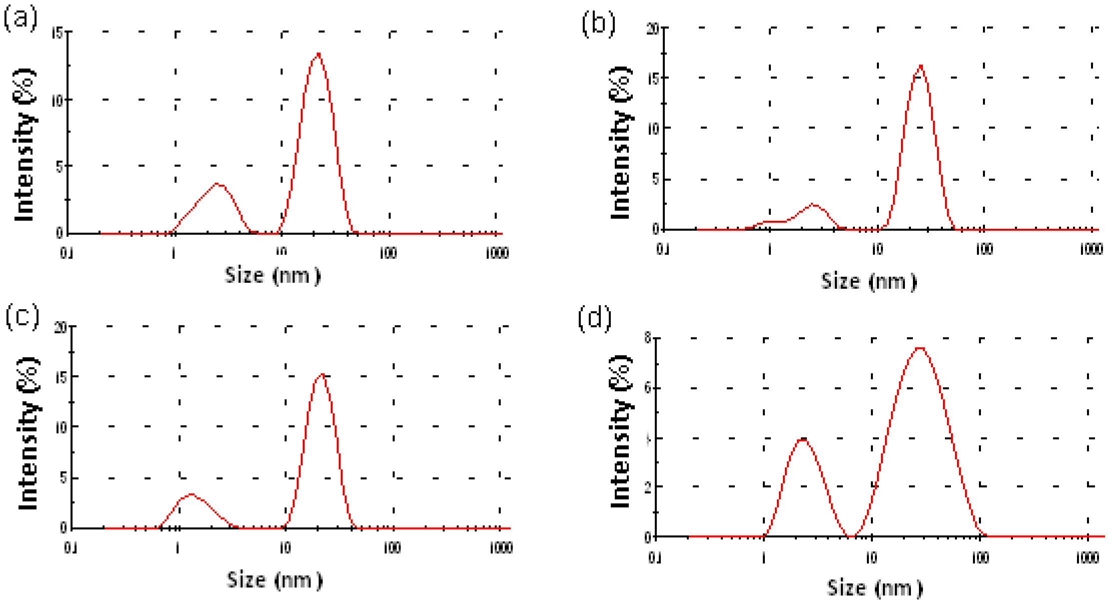

Fig.8. Size distribution by intensity for AuNRs synthesized with varying silver nitrate concentration; (a) 0.003 M, (b) 0.004 M, (c) 0.005 M and (d) 0.007 M.

The effect of temperature was studied at 25°C, 35°C and 70°C. UV-Vis spectra exhibited two absorbance peaks for samples synthesized at 25°C and 35°C but only one peak for 70°C (Fig. 9). The result suggested that AuNRs was formed up to 35°C. When temperature increased to 70°C, only AuNSs were produced. Furthermore, longitudinal and transverse peaks shifted to shorter wavelength as temperature increased from 25 C to 35°C. This indicated that both length and width of AuNRs decreased with increasing temperature. Besides, the intensity for AuNRs at 35°C was lower than 25°C, indicated that lower rods yield.

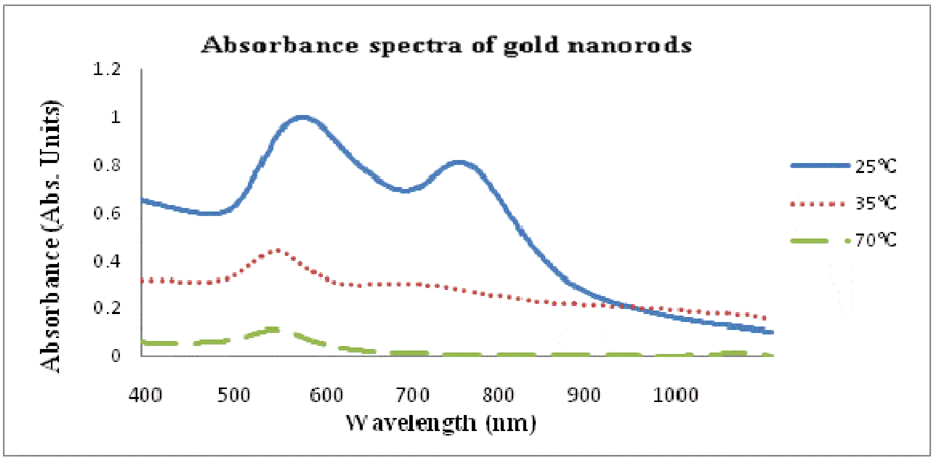

Fig.9. UV-Vis spectra of AuNRs synthesized at various temperatures.

TEM images in Fig.10 further confirmed AuNRs were produced at 25°C and 35°C whereas only AuNSs were formed at 70°C. When the solution temperature decreased, the growth rate of gold particles also decreased. Consequently, the gold intermediates remained in the solution for a relatively long time. Thus, the probability that CTAB adsorbed onto unstable facets of the gold intermediate increased at low temperature. Meanwhile, the growth rate of gold increased as the temperature increased hence the intermediate rapidly disappeared. As a result, intermediates did not adsorb by CTAB and formed spheres [9].

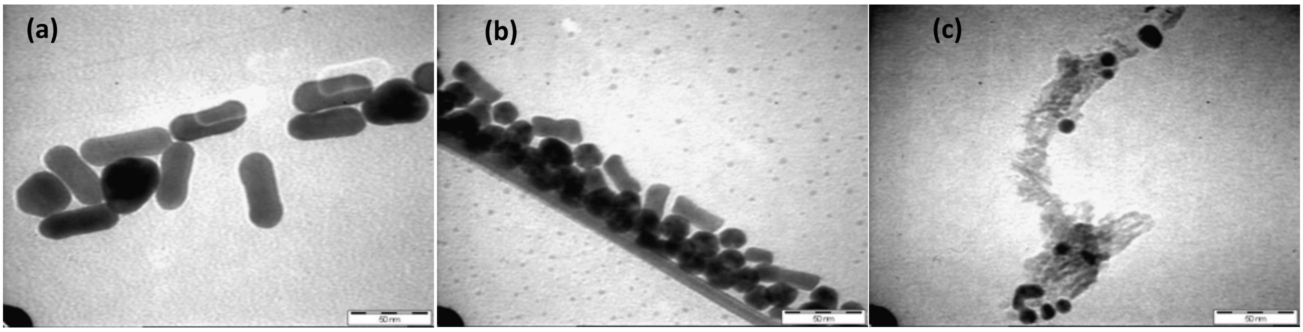

Fig.10. TEM images of AuNRs synthesized with varying temp: (A) 25°C (B) 35°C and (C) 70°C

Fig. 11 shows the particles size distribution of AuNRs with different temperature using Zetasizer. It can be seen that, both peaks that represented width and length of AuNRs decreased when temperature increased. The formation of shorter length AuNRs at higher temperature might be related to the number of nuclei with the addition of sodium borohydride. More nuclei were present when temperature increased caused less gold monomer available per nuclei. Consequently, shorter length of nanorods was formed [10]. AuNSs formed at 70°C shows well size distribution and indicated that AuNSs were almost similar in size. The Z-average size for samples synthesized at 70°C was 60.66 nm.

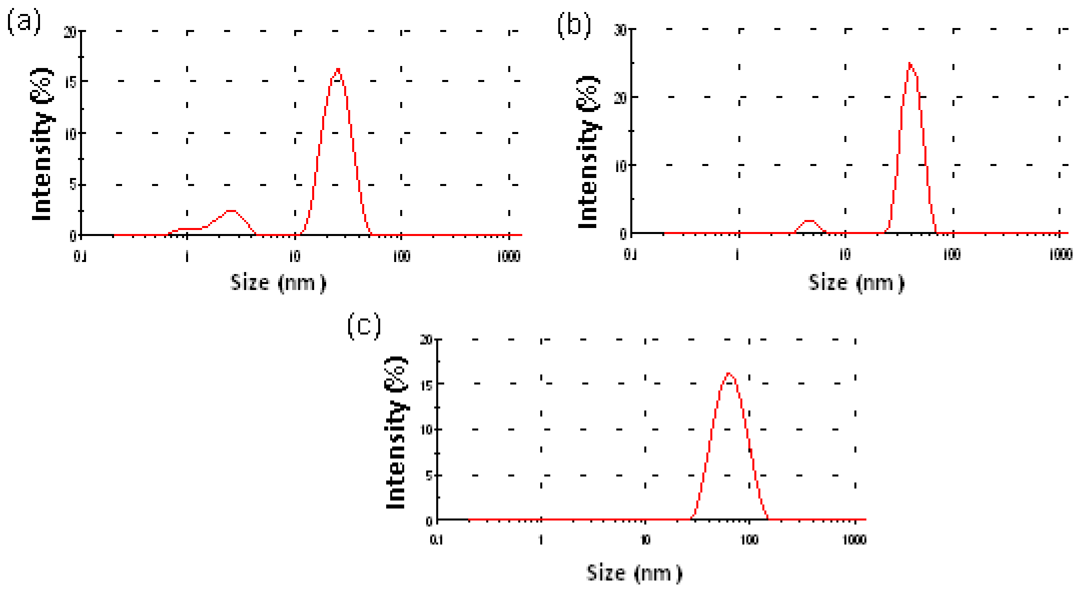

Fig.11. Size distribution of AUNRs synthesized with varying temperature, (a) 25°C, (b) 35°C and (c) 70°C.

CONCLUSIONS

Gold nanorods (AuNRs) have been successfully synthesized using the seed mediated approach. The formation of AuNRs is highly dependent on CTAB concentration and concentration of silver nitrate at room temperature and least dependent on gold concentration in the growth solution.

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