



# Direct Investigation of Synthesis of Gold Nanoparticles Using *Polyscias scutellaria* Leaf Extract in the Hexane–Water System Using the Centrifugal Liquid Membrane–Spectrophotometry Method

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<https://doi.org/10.14710/jksa.23.7.255-260>

## Article Info

### Article history:

Received: 13<sup>th</sup> May 2020

Revised: 4<sup>th</sup> July 2020

Accepted: 17<sup>th</sup> July 2020

Online: 31<sup>st</sup> July 2020

### Keywords:

Green Synthesis; Centrifugal Liquid Membrane–spectrophotometry; Au Nanoparticles; Hexane–Water System; *Polyscias scutellaria*

## Abstract

The Centrifugal liquid Membrane (CLM) method, which provides an ultra-thin two-phase liquid membrane system in a rotating glass cell, was successfully applied to Green Synthesis from *Polyscias scutellaria* (PS) capped gold nanoparticles (AuNPs–PS) using a Mangkokan leaf (*Polyscias scutellaria*) extract as a reducing agent and stabilizer in the hexane–water system. PS extract in hexane fraction as the organic phase has a UV absorption spectrum at the maximum wavelength,  $\lambda_{\max}$  of 220 nm, while the precursor of HAuCl<sub>4</sub> solution as an aqueous phase has an  $\lambda_{\max}$  of 214 nm. Investigation of AuNPs–PS formation was carried out at various concentrations of Mangkokan leaf extract concentration; i.e., 0.001 0.003; 0.005; 0.007 and 0.009%, while the reaction was carried out at various rotational speeds of 5,000–9,000 rpm. The formation and stability of AuNPs–PS were observed from the phenomenon of surface plasmon resonance (SPR) and absorbance changes as measured by a UV–Vis spectrophotometer. The results of measurements using CLM–Spectrophotometry shows the formation of AuNPs–PS in the hexane–water system at  $\lambda_{\max}$  of 534 nm.

## 1. Introduction

Nanoparticles are materials that have a size on a nanometer scale ranging from 1–100 nm. Many researchers are interested in studying nanomaterials because, with nano-size, the material properties will be more advantageous than larger sizes [1]. Nanoparticle material engineering is the tailoring of size, shape, morphology, and arrangement of materials at the nanometer scale, which will determine the characteristics of nanoparticles synthesized [2].

Gold nanoparticles have various forms, including Au nanosphere, Au nanorods, Au nanoshells, Au nanocubes, and Au nanocages [3, 4, 5]. Synthesis of gold nanoparticles is generally carried out by the Turkevich method, namely by using chloroauric acid (HAuCl<sub>4</sub>) with a reducing agent sodium citrate. The primary reaction is

the reduction of Au salt (HAuCl<sub>4</sub>) to form a gold metal. This method converts the synthesis of Au<sup>3+</sup> ions into Au<sup>0</sup>. Sodium citrate is often used in the synthesis of gold nanoparticles. Besides acting as a reducing agent, it is also a stabilizer that can prevent the aggregation of gold nanoparticles [6].

The public knows Mangkokan (*Polyscias scutellaria*) as a medicinal plant. The roots, stems, leaves, or fruits of this plant contain bioactive compounds that can treat various types of diseases. In general, medicinal plants have flowers containing substances from the flavonoid, aurone, chalcone, anthocyanin/anthocyanidin groups, which in this study are used to reduce and stabilize agents [7, 8, 9].

The Centrifugal Liquid Membrane (CLM) method can measure reaction rates at the interface area in

seconds. This technique is a direct measurement of chemical species that are adsorbed at the liquid-liquid interface [10, 11]. The use of CLM is by injecting a small amount of reagent volume into a cylindrical glass. Then a high-speed rotation is carried out, which produces a centrifugal force forming a phase layer on the cylinder glass wall [12, 13]. The cylindrical glass cell is then connected to a photomultiplier from the UV-Vis spectrophotometer. In this study, the CLM method is used to see AuNP colloid systems with a water dispersing medium.

Choiry [14] studied the direct measurement of the Au-mercaptosuccinic-Fe (II) nanoparticle complex with the centrifugal liquid membrane (CLM) method using Cr (VI) reducing agents which are harmful to the environment. Research on AuNP green synthesis using extracts (*Polyscias scutellaria*) with UV light reduction and methylene blue reduction catalysis studies is still preliminary. They have a disadvantage because UV light is unstable [15]. While in this study, it tried to use plant extracts (*Polyscias scutellaria*), which contain secondary metabolites of flavonoids, alkaloids, tannins, and terpenoids that are environmentally friendly, can be a capping agent for stable AuNP and able to reduce  $\text{Au}^{3+}$  to  $\text{Au}^0$  well. Also, the use of hexane-water interphase systems provides benefits, as it only requires a small sample with sensitive detection and direct measurement using a UV-Vis spectrophotometer.

The renewal of this research is that direct observation using UV-Vis spectrophotometer in the hexane-water system can also be determined by the CLM method. Samples of the hexane - water system are injected into a cylindrical glass tube. A rotation (centrifugal force) to 10000 rpm is obtained to obtain a thin membrane (212  $\mu\text{m}$ ) with a large surface area. The centrifugal liquid membrane method was successfully applied to the green synthesis of *Polyscias scutellaria*-capped gold nanoparticles using plant extracts to reduce and stabilize using the hexane-water system.

## 2. Methodology

This research was conducted at the Nano Interfacial Chemistry Laboratory, University of Indonesia. This research was conducted in 3 stages, i.e., (i) synthesis of gold nanoparticles (AuNP) with Mangkokan leaf extract (*Polyscias scutellaria*), (ii) synthesis of AuNP@Mangkokan leaf extract by Centrifugal Liquid Membrane method and (iii) variation of Mangkokan leaf extract concentration using CLM method.

### 2.1. Equipment and Materials

The equipment used in this study was measuring cups, measuring flasks, chalice cups, stirring rods, test tubes, drop pipettes, volumetric pipettes, spray bottles, analytical balance, evaporators, Buchner funnels. The instruments used were Shimadzu 2600 UV-Vis spectrophotometer, Centrifugal Liquid Membrane equipment - NSK Nakanishi high-speed NK260 Electer and Ono Sokki HT-431 Non-Contact Digital Photo Tachometer to monitor Centrifugal Liquid Membrane control.

The material used in this study was the Mangkokan leaf plant from a plantation in the Cilebut area, Bogor, and pure gold bar (99.99%) from PT. Antam. The chemicals used were technical methanol, technical *n*-hexane, technical ethyl acetate, concentrated HCl 32%, concentrated  $\text{HNO}_3$  65%, HCl 0.02 M, 0.02 M NaOH, Wagner reagents, acetic anhydride, concentrated  $\text{H}_2\text{SO}_4$ , chloroform, 50% methanol, Mg powders,  $\text{FeCl}_3$ , HCl, and Silica gel 60 PF 254 obtained from Merck.

### 2.2. Synthesis of Au Nanoparticles with Mangkokan Leaf Extract (MLE)

$\text{HAuCl}_4$  stock solution was prepared by weighing 0.234 g pure Au (99.99%) and dissolved in aqua regia ( $\text{HCl}:\text{HNO}_3 = 4:1$ ). The solution was evaporated by heating  $120^\circ\text{C}$  to form  $\text{AuCl}_4^-$  crystals. The crystal was washed twice and then dissolved using 118.8 mL of double-distilled water so that a 1.51% (m/v)  $\text{HAuCl}_4$  mother liquor was obtained. Then a solution of  $\text{HAuCl}_4$  was made by pouring 9 mL of stock solution of  $\text{HAuCl}_4$  1.51% (m/v) on a 100 mL volumetric flask and diluted with double distilled water to the mark. Then, a 9 mL  $\text{HAuCl}_4$  solution was reacted with 1 mL of Mangkokan leaf extract solution, followed by characterization using a UV-Vis spectrophotometer directly connected to the Centrifugal Liquid Membrane.

### 2.3. General procedures for using Centrifugal Liquid Membrane (CLM)

AuNP@MLE synthesis was carried out using the Centrifugal Liquid Membrane (CLM) method. The characterization tool used to observe the formation of AuNP@MLE directly is the UV-Vis Spectrophotometer connected to the Centrifugal Liquid Membrane (CLM) motor. AuNP synthesis was carried out in which the  $\text{HAuCl}_4$  solution as a blank was inserted into a cylindrical rotating glass cell, then added the *n*-hexane solution then connected to the CLM rotator at a speed of 10,000 rpm. After 20 seconds, the rotating glass cell was shot by UV-Visible light at a wavelength of 200–800 nm. The MLE solution is mixed into a cylindrical rotational glass cell that has previously contained  $\text{HAuCl}_4$ , then connected to the CLM rotator at a speed of 10,000 rpm. After 2 minutes, the glass cells rotate, and the ultrathin membrane was formed, then UV-Visible was shot at a wavelength of 200–800 nm, and changes are observed every time unit.

### 2.4. Concentration Variation in Synthesis of AuNP-Mangkokan Leaf Extract (*Polyscias scutellaria*)

The variation of Mangkokan leaf extract concentration was 0.001%; 0.003%; 0.005%; 0.007% and 0.009%. 250  $\mu\text{L}$   $\text{HAuCl}_4$   $2.0 \times 10^{-4}$  M solution was injected into the glass cell as a blank, then add 200  $\mu\text{L}$  *n*-hexane and rotated at a rotational speed of 10,000 rpm. Then still in a non-rotating condition, the MLE solution was injected until the desired mixture concentration was obtained. At a rotation speed of 10,000 rpm, a  $\lambda_{\text{max}}$  measurement was performed using a UV-Visible spectrophotometer in the range of 200–800 nm after 2 minutes of rotation.

### 3. Results and Discussion

#### 3.1. Liquid - Liquid Interface Chemistry

Based on the UV-Vis absorption spectrum pattern, what water phase is at a wavelength of 220 nm, and the organic phase at 350 nm. The wavelength at 535 nm corresponds to the results of AuNP synthesis. Based on the phenomenon of liquid-liquid interface extraction, forming AuNP synthesis is drawn towards the water phase, indicating the dissolution process. The AuNP color is pink, distinctive color, and Figure 1 shows a wavelength of 535 nm corresponding to the surface plasmons resonance (SPR) of AuNP. Based on preliminary tests of liquid-liquid interface chemistry, it can be seen the phenomenon between the water-phase hexane phase. So, it can be concluded that AuNP can be synthesized by the centrifugal liquid membrane (CLM) method so that the concentration profile can be seen continuously at the interface and bulk phase.

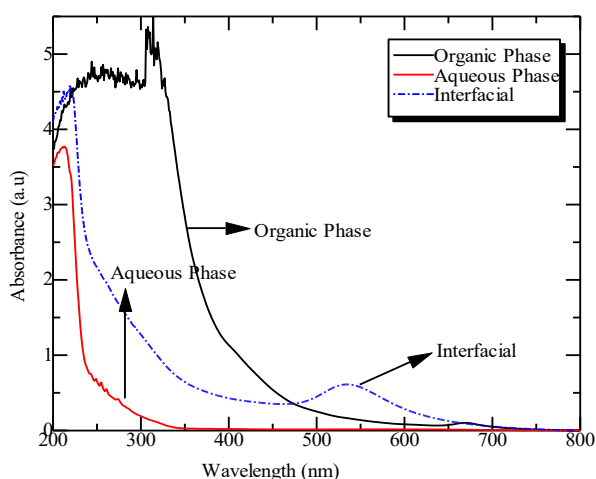


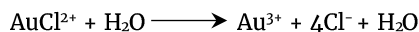
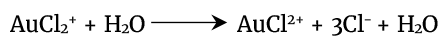
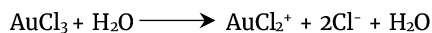
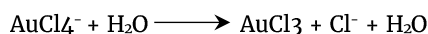
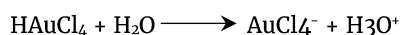
Figure 1. UV-Vis Absorption Spectra in Liquid-Liquid Interface Chemistry

#### 3.2. AuNP@MLE Synthesis with Centrifugal Liquid Membrane (CLM) Method

AuNP nanoparticles in this study were used as a chemical species detector. The AuNP@MLE synthesis process can be observed using the Centrifugal Liquid Membrane (CLM) method through a change in color from clear (colorless) to pink and measurements by UV-Vis spectroscopy at a wavelength of 520–580 nm which is the typical peak of AuNP. The pink color produced from AuNP arises from the plasmon band's frequency, which absorbs green light strongly and transmits red light.

The synthesis of gold nanoparticles uses the Brust method, while the application uses the CLM method. The combination of these methods aims to obtain the smallest AuNP size possible to enlarge the surface area that interacts with the analyte to be detected. This research has the advantage of measuring with CLM combined with UV-Vis spectrophotometry to measure it in situ. When the cell is rotated and shot directly by UV-Vis light, the wavelength can be adjusted to the desired  $\lambda_{max}$  until the formation of AuNP size is on the nanometer

scale. The AuNP stability analysis process that is formed is carried out directly. CLM rotation can accelerate the growth of AuNP and maintain uniformity of the size distribution formed. Proof based on observing the reaction with the formation of pink and UV-Vis spectra measurement results. The use of CLM as a detection system is due to the volume of media used is only about 0.2–1 mL, and this depends on the stability of the AuNP formed. AuNP synthesis research using the CLM method was carried out by injecting 250  $\mu$ L H<sub>AuCl<sub>4</sub></sub> solution into the glass cell as a blank, and the glass cell was rotated at 7,000 rpm. The H<sub>AuCl<sub>4</sub></sub> solution used has a concentration of  $2.0 \times 10^{-4}$  M. In water, the H<sub>AuCl<sub>4</sub></sub> solution ionizes to Au<sup>3+</sup> through the following steps:



Furthermore, 200 mL of hexane MLE fraction is injected then the glass cell is rotated, then another 50 mL of MLE water fraction is added to the speed of 10,000 rpm. Then  $\lambda_{max}$  is measured using UV-Vis spectrophotometry and the smoothness of the spectra pattern produced after 2 minutes of rotation is observed.

#### 3.3. Variation of Mangkokan Leaf Extract Concentration

The variation of Mangkokan leaf extract concentration in the CLM method aims to determine the ability to reduce MLE's active compounds, determine the smallest AuNP size, and the  $\lambda_{max}$  that appears in the UV-Vis absorption spectrum. The smaller the wavelength, the smaller the AuNP that is formed, while the higher the wavelength, the larger the AuNP that is formed.

AuNP@MLE synthesis was carried out by adding H<sub>AuCl<sub>4</sub></sub> with a fixed volume. This synthesis is carried out at room temperature in the CLM system. The results obtained show the smaller the concentration of the mixture, the smaller the  $\lambda_{max}$ , which means the smaller the particle size formed. At the same time, the optimum concentration is the concentration that gives the smallest wavelength in UV-Vis spectra.

AuNP growth was observed rapidly in 2 minutes using CLM. The centrifugal force possessed by the CLM system accelerates the formation of AuNP. Figure 2 shows the results of UV-Vis characterization in AuNP synthesis using the CLM method. It is seen that with variations in MLE concentrations, there are secondary metabolites of flavonoid compounds that play a role in the formation of AuNP by influencing the wavelength, absorbance, and spectral band shape of the AuNP formed. AuNP synthesis was carried out by varying the volume of the reducing agent, as presented in Table 1.

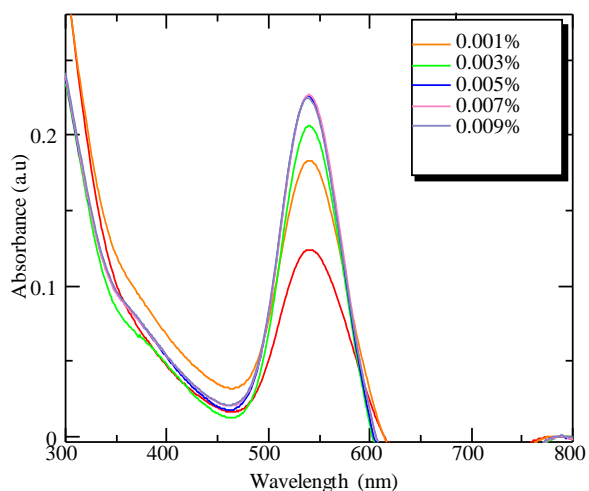
**Table 1.** Variation of concentration of Mangkokan leaf extract in reducing HAuCl<sub>4</sub> and λ<sub>max</sub> produced

Volume of HAuCl <sub>4</sub> (1.0 x 10 <sup>-4</sup> M) (mL)	Volume of Mangkokan leaf extract (1.0x10 <sup>-3</sup> M) (mL)	Mangkokan leaf extract concentration (%)	λ <sub>max</sub> (nm)
1	1	0.001 %	537
1	1	0.003 %	531
1	1	0.005 %	533
1	1	0.007 %	530
1	1	0.009 %	530

With a concentration of 0.001%, MLE gives noisy results with a wavelength of 537 nm and a rough spectrum and not yet pink. This indicates that AuNP has not formed completely. At MLE with a concentration of 0.003%, the absorption spectrum formed is still slightly rough, the size of the nanoparticles is still significant because λ<sub>max</sub> is at 531 nm, but the shape of the absorption peak has started sharp. There is still noise being observed due to the influence of the CLM system.

MLE with a concentration of 0.005% has a wavelength with a peak of 533 nm with noisy absorption. This indicates that the AuNP formed is larger than 0.003%, and the distribution is heterogeneous in size. A higher concentration than the optimum concentration of the reduction process of Au<sup>3+</sup> to AuNP is formed more rapidly, resulting in uneven particle nuclei followed by agglomeration to produce a much larger size AuNP.

The concentration of 0.007% and 0.009% is the optimum concentration in the formation of AuNP by the CLM method with the smallest wavelength of 530 nm. At this concentration, the AuNP formed is small, with higher absorbance than at concentrations of 0.005%, 0.003%, and 0.001%. The sharp absorption peak also indicates that the size of the AuNP produced is homogeneous. The smaller the size of AuNP, the capping agent process using Mangkokan leaf extract is more easily formed in the hexane-water interphase system.



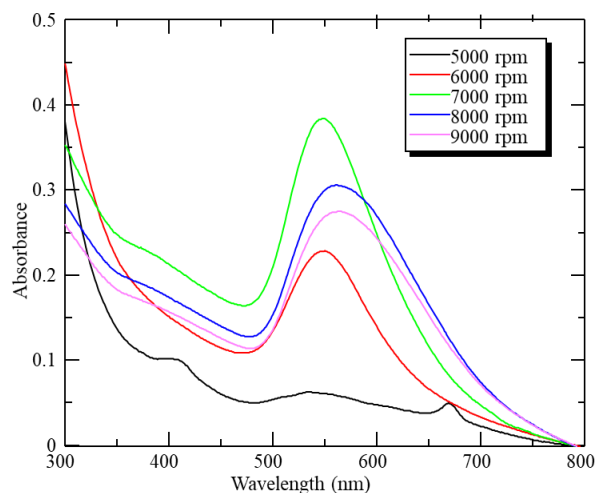
**Figure 2.** UV-Vis AuNP absorption spectra at various concentrations of (a) 0,001%, (b) 0,003%, (c) 0,005%, (d) 0,007%, (e) 0,009% in the CLM system

### 3.4. Rotation Speed Variations in Centrifugal Liquid Membranes

Rotation speed variations are carried out to determine the optimum rotation speed, which can provide the smoothest UV-Vis absorption spectra. From the results of UV-Vis detection combined with the CLM system, it can be seen as the CLM's rotation stability. The fineness of the UV-Vis absorption spectra depends on the stability of the CLM rotation position when operated. At a certain speed, CLM rotates stably, and the rotation is homogeneous and fast so that the resulting spectra are smooth, which is marked by the peak of absorbance that is visible.

Rotational speed affects the formation of thin membranes in the cell wall as the liquid inside the glass cell experiences centrifugal force so that the liquid rises upwards lining the glass cell wall. Centrifugal force is defined as a circular motion force that moves away from the center of the circle. The nature of this centrifugal force causes the fluid in the curved tube to rise to a certain height.

The rotational speed used was 5000 rpm; 6000 rpm; 7000 rpm; 8000 rpm; 9000 rpm; and 10,000 rpm. Results The UV-Vis absorption spectra of samples synthesized with different rotational speeds are presented in Figure 3.



**Figure 3.** UV-Vis spectra of samples with different rotational speeds using the CLM system in the hexane-water system

Figure 3 shows the results of the UV-Vis absorption spectra of AuNP prepared with different rotational speeds. The smoothest AuNP colloidal spectra prepared by the CLM system, obtained at a rotational speed of 7,000 rpm. Figure 3 shows that the finer order of the spectra produced was 7000 rpm > 8000 rpm > 7000 rpm > 6000 rpm > 5000 rpm. Figure 3 also shows that the spectrum at 5000, 8000, and 9000 rpm rotations look broad due to large-sized AuNP being formed or agglomeration of the particles. The fineness of the spectrum also reveals that the higher the rotational speed, the smaller the λ<sub>max</sub> produced. These results are presented in Table 2.



**Table 2.** Relationship between rotational speed and the resulting  $\lambda_{\max}$ 

rotational speed (rpm)	$\lambda_{\max}$ (nm)
5000	670 nm
6000	557 nm
7000	540 nm
8000	580 nm
9000	580 nm

AuNP synthesized at a rotational speed of 5000 rpm is in a stable state, as seen from the results of the absorption spectrum formed even though the resulting spectrum is slightly rough. The rotational speed of 5000 rpm provides enough centrifugal force to make the liquid rise perfectly over the wall to form a membrane, but the size of the formed nanoparticles is still large, as seen from the value of  $\lambda_{\max}$  of 670 nm. Compared with a rotation of 5,000 rpm, the absorption spectrum of a sample of 7,000 rpm is much finer and has a very sharp peak.

At a rotational speed of 6000 rpm, the CLM rotation is in a stable state, as seen from the smooth AuNP spectrum. This speed is enough to make the liquid rise perfectly lining the wall to form a membrane. However, the size of the nanoparticles formed is still quite large, as seen from  $\lambda_{\max}$  of 557 nm.

At a rotational speed of 7000 rpm, the formation of thin membranes in glass cells is fast, homogeneous, stable, and entirely coated walls. The AuNP absorption spectrum produced is excellent; the absorbance at  $\lambda_{\max}$  is observed (without the aid of Gaussian fitting analysis). AuNP growth was observed, and the resulting  $\lambda_{\max}$  was the smallest among the other rotational speeds, which was 540 nm. The speed of 7,000 rpm is summed up as the ideal speed because the results produce the smoothest absorption spectrum. In this rotation condition, it is expected to reduce H<sub>2</sub>AuCl<sub>4</sub> by MLE to AuNP faster.

At a rotational speed of 8000 rpm, AuNP synthesis is well-formed, in which the membrane lining of the glass cell wall is perfectly formed. However, when viewed from the absorption spectrum, it looks rough compared to the 5000-rpm rotation. The resulting  $\lambda_{\max}$  is in the region of 580 nm and is higher than the 7000-rpm sample, so the formed nanoparticles are larger.

At a rotational speed of 9000 rpm, AuNP is well-formed, but the peak experiences broadening because the membrane coating on the glass cell wall is not fully formed and undergoes aggregation so that the size of the AuNP becomes larger. The resulting  $\lambda_{\max}$  is 580 nm.

#### 4. Conclusion

AuNP synthesis using the centrifugal liquid membrane method with the hexane-water phase system using Mangkokan leaf extract (*Polyscias scutellaria*) as a capping agent was successfully carried out. Preliminary studies show that the results of the UV-Vis absorption spectrum for the hexane phase are at a wavelength of 220 nm. The interface region has a wavelength of 523

nm, and this is in accordance with the surface plasmons resonance (SPR) of AuNP. While the aqueous phase has a wavelength of 350 nm. The results of measurements using CLM-Spectrophotometry showed the formation of AuNPs-MLE. Variations in the concentration of Mangkokan leaf extract reached optimum concentrations of 0.007% and 0.009% in the hexane-water system with  $\lambda_{\max}$  of 530 nm. Meanwhile, the most optimum rotation speed in the hexane-air system is obtained at 7000 rpm. At this speed, AuNP growth is observed, the  $\lambda_{\max}$  produced is the smallest compared to other rotational speeds, which is at 540 nm. The spectrum obtained is smooth and sharp. The results of measurements using CLM-Spectrophotometry show that the formation of AuNPs-PS in the hexane-water system has an  $\lambda_{\max}$  of 534 nm.

#### Acknowledgment

This research was funded by the Ministry of Research and Technology Grant, National Research and Innovation Agency, with a contract number of 051/LL3/PG/2020.

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