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Biosynthesis of silver nanoparticles using the leaf and stem bark of *Diospyros discolor* Willd. (Ebenaceae)

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Abstract. The synthesis of nanoparticles using parts of plants is extremely attractive because it is environmentally friendly. This study examined the suitability of the leaf and stem bark extracts of the plant *Diospyros discolor* for the synthesis of silver nanoparticles. The reaction could be monitored by visual examination, because the formation of silver nanoparticles was accompanied by a color change of the solution. The characterization of silver nanoparticles was performed using UV-visible, particle size analyzer, and transmission electron microscopy analyses. We observed that the process of nucleation of the silver nanoparticles required several hours. The biosynthesis of silver nanoparticles using the stem bark extract was faster than that using the leaf extract. In addition, the volume ratio of the extracts and AgNO₃ solutions greatly affected the yield of silver nanoparticles. This effect was more significant in the case of the leaf extract. This biosynthesis successfully afforded spherical silver nanoparticles of about 30 nm in diameter, which were relatively stable on the basis of zeta potential measurements.

1. Introduction

In the last decade, silver nanoparticles have been extensively studied because of the availability of various synthetic methods for their preparation and the interesting physical and chemical properties inherent to nanoparticles [1–3], which render them applicable in a variety of fields (such as the packaging industry, pharmacy, antimicrobial applications, and sensors [4–7]). Although nanoparticles can be readily produced by chemical synthesis, biosynthesis is gradually becoming a more preferable alternative because it is environmentally friendly and reduces the production cost [8,9].

Recently, a plant of the genus *Diospyros* was used as material for the biosynthesis of silver nanoparticles [10]. Another plant of the same family, *Diospyros discolor*, which is commonly found in Austronesian peripheral regions, has been the target of several studies in order to evaluate its characteristics [11,12]. Motivated by this background, in this study, we compare the suitability of the leaf and stem bark extracts from *Diospyros discolor* for synthesizing silver nanoparticles.

2. Materials and methods

2.1. *Diospyros discolor* wild leaf and stem bark extraction

Diospyros discolor leaves and stem barks were obtained from the collection of trees in the garden of Universitas Indonesia. Both materials were washed and then placed in an oven preheated to 40 °C for



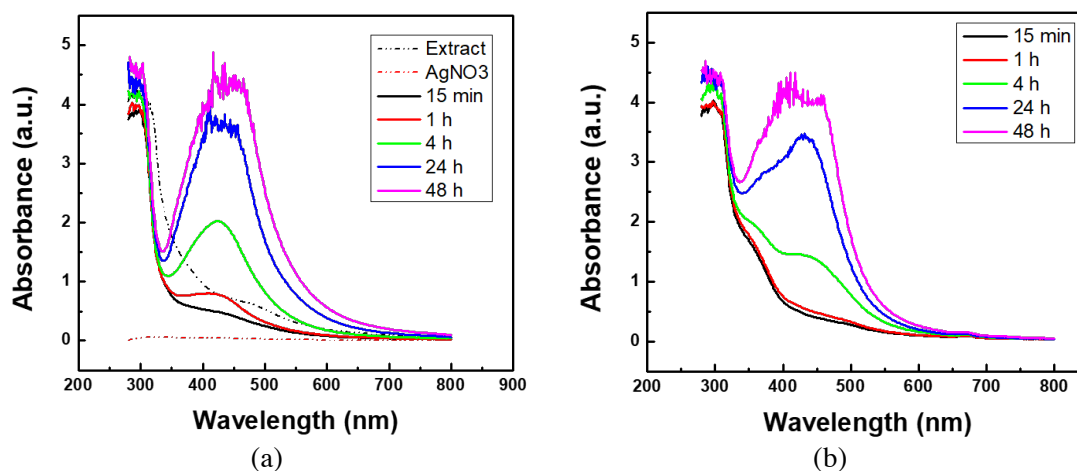


Figure 1. UV-vis spectra for *Diospyros discolor* (a) stem bark extract and (b) leaf extract corresponding to the 1:2 ratio as a function of silver nanoparticle nucleation time.

drying. During this process, the materials were weighed every hour until no mass change was detected, indicating complete evaporation of the water content. The dried materials were then ground to a fine powder, which was used for the preparation of aqueous extracts with 2 % powder content in 65 mL of aquabidest. Next, the resulting solutions were magnetically stirred to their boiling points in order to obtain the extract from both materials, and boiling was maintained for 15 min before cooling to room temperature. The solvent was then screened through Whatman grade 1 filter paper to separate undissolved plant excess.

2.2. Green synthesis of silver nanoparticles

Silver nitrate was obtained from Merck (Germany). Silver nanoparticles were synthesized by mixing the extracts with a 0.01 M solution of silver nitrate in 1:2, 1:5, 1:10, and 1:20 extract:silver nitrate ratios. Here, *Diospyros discolor* acts as a silver ion reductor for the formation of silver nanoparticles. The nucleation process was visually monitored by observing the solution color change after 0.25, 1, 4, and 24 h of reaction. The solution was also subjected to UV-visible (UV-vis) spectrometry (Thermo UV-vis 10 S Genesys) as a function of reaction time. The size distribution of the silver nanoparticles was determined using a particle size analyzer (PSA) Melvern Zetasizer Nano Series, and their structures were characterized through transmission electron microscopy (TEM) using the FEI Technai G2 Spirit 120 KV microscope.

3. Results and discussion

3.1. UV-vis absorption spectra characterization

Figure 1a and figure 1b shows the absorption spectra of the silver nanoparticles biosynthesized from the leaf and stem bark extracts at the 1:2 extract:AgNO₃ ratio as a function of reaction time, along with the spectra for the AgNO₃ solution and the plant extract for comparison (figure 1a). The presence of absorption peaks at approximately 420 nm of wavelength in figure 1 confirms the formation of silver nanoparticles [13,14]. This peak is not present in the spectra of AgNO₃ and plant extract.

The observed absorption values were found to be strongly influenced by the reaction time, which indicates that the silver nanoparticle nucleation takes several hours to complete. Figure 1a shows that the silver nanoparticles from the stem bark extract required shorter nucleation time than those from the leaf extract. Thus, in the biosynthesis with stem bark extract, the silver nanoparticle absorption peak can be observed from 1 h reaction time and increases at 4 h, whereas that with leaf extract displays the corresponding absorption peak after 4 h of reaction.

Figure 2 shows the UV-vis spectra for both *Diospyros discolor* extracts at different extract:AgNO₃ ratios and the corresponding solution colors. Because the absorption peaks were not intense within the first hours of reaction (figure 1), the nucleation time of 24 h was selected for the visual examination included in figure 2. A distinctive difference in the absorbance as a function of solution concentration

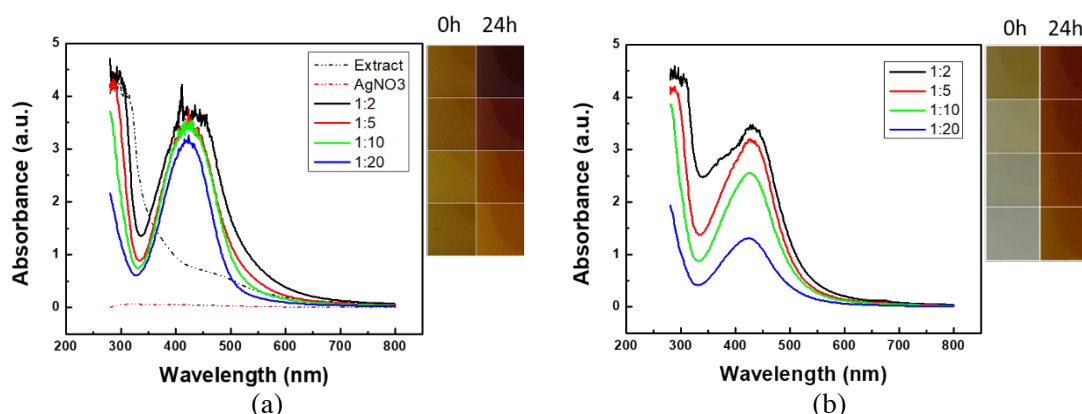


Figure 2. UV-vis spectra at different extract:AgNO₃ ratios and solution colors at 24 h nucleation time for *Diospyros discolor* (a) stem bark extract and (b) leaf extract.

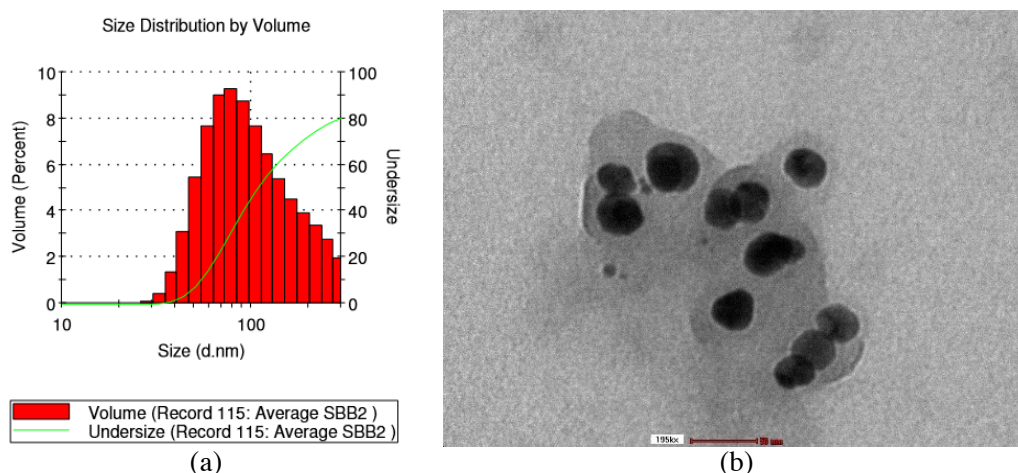


Figure 3. (a) Size distribution by volume from the PSA result and (b) TEM image for the silver nanoparticles biosynthesized using *Diospyros discolor* stem bark extract.

between the stem bark and leaf extracts was observed. In the case of the stem bark extract, the absorbance from the produced silver nanoparticles was not significantly altered by the change of ratio, although a slight increase in the absorbance value with the AgNO₃ concentration was detected. In the case of the leaf extract, the absorbance was highly sensitive to the leaf extract:AgNO₃ ratio, with the highest AgNO₃ concentration affording the lowest absorbance. This result indicates that the optimal leaf extract:AgNO₃ ratio to reduce Ag⁺ to Ag⁰ was 1:2.

3.2. Size distribution and TEM imaging

Figure 3 and figure 4 show the result of the PSA and TEM analyses for the silver nanoparticles obtained from the stem bark and leaf extracts, respectively. The average size of nanoparticles produced from the stem bark extract was determined to be approximately 117 nm (figure 3a), which was twice the average size of nanoparticles produced from the leaf extract (63 nm, figure 4a). Zeta potential measurements for both the stem bark and leaf silver nanoparticles afforded similar values of 15.7 and 16.2 mV, respectively, which correspond to relatively stable nanoparticles, albeit still vulnerable to agglomeration forming larger nanoparticles [15]. This result is in line with that obtained from PSA and UV-vis. Thus, the PSA spectra display wide nanoparticle size distributions, in accordance with the UV-vis spectra, which exhibit full widths at half maximum characteristic of equally wide absorbance peaks. The TEM images show spherical silver nanoparticles with sizes ranging from 30 to 100 nm (figure 3b and figure 4b). This further supports the tendency of nanoparticles to experience agglomeration.

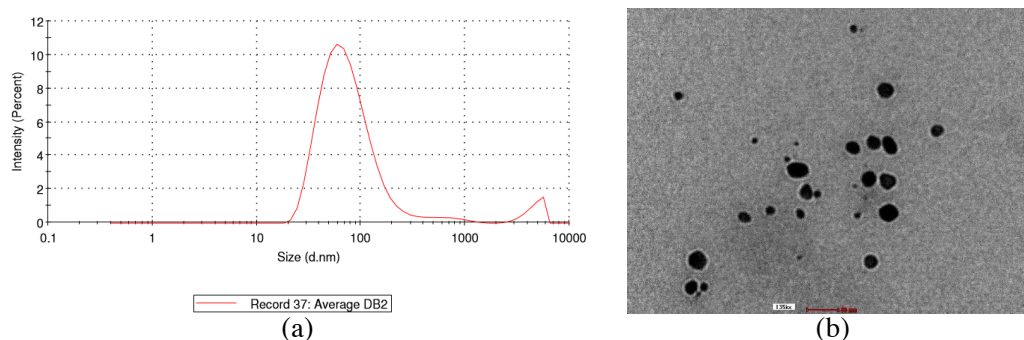


Figure 4. (a) PSA result and (b) TEM image for the silver nanoparticles biosynthesized using *Diospyros discolor* leaf extract.

4. Conclusions

Silver nanoparticles were successfully produced through biosynthesis using the leaf and stem bark extracts from *Diospyros discolor*. The process of nanoparticle nucleation during the biosynthesis was strongly influenced by the reaction time, requiring several hours to complete, with the biosynthesis from leaf extract being slower than that from stem bark extract. In addition, the ratio of plant extract and AgNO_3 solution affected the yield of silver nanoparticles. This feature was more significant in the case of the leaf extract. The as-prepared silver nanoparticles exhibited wide particle size distribution, ranging from 30 to 100 nm, which is indicative of their tendency to agglomerate forming larger nanoparticles.

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