Green Synthesis of Silver Nanoparticles using Extract of *Pinus merkusii* Jungh & De Vriese Cone Flower

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**Abstract.** The paper studies recent application of cone flower waste from *Pinus merkusii* Jungh & De Vriese for an environmentally unclear method for synthesis silver nanoparticle. Phytochemical characterization using iron trichloride solution showed the extract of *Pinus merkusii* cone flower contains of phenolic group of secondary metabolite. This group acts as both reducing and stabilizing agents. For the synthesis of silver nanoparticle, solution of silver nitrate is added to the extract at 60 °C. The effect of extract concentration (5-20%) and time reaction (15-60 min) is investigated. The formation of silver nanoparticle is confirmed by the color change from yellowish to brown. Meanwhile, UV-Vis characterization of silver nanoparticle in extract 20% and 60 min reaction showed surface plasmon resonance (SPR) at 431 nm, and transmission electron microscope (TEM) revealed the particle size range in between 8 and 23 nm with a spherical in shape.

1. **Introduction**

Silver nanoparticles (AgNP) in recent years has aroused a growing interest due to their unique properties and potential application such as catalytic, antibacterial and anticancer drug delivery [1–3]. Although synthesis of silver nanoparticles (AgNP) have been applied by chemical methods, the reactants used in these reaction are potentially hazardous. Synthetic method based on environmentally benign is need for AgNP synthesis, do not only an alternative that means friendly of obtaining these nanoparticle, but also have a well-defined morphology and size.

Plant extract have been reported to bioreduce metal ion to metal nanoparticle currently. Some extract plants have achieved success in the synthesis of metal nanoparticle [4–6]. The previous reference exhibited plant wastes can be utilized for the synthesis of nanoparticle too. *Pinus merkusii* are one of Indonesian’s original pine forest, and the cone flower are waste. According to the reference, the *Pinus merkusii* contains of α-pinene, β-pinene [7], phenolic and flavonoid compound like pinosylvin, pinosylvin monomethyl ether, dimethyl ether pinosylvin, and pinocembrin [8]. The phenolic compound is capable of functioning in reducing metal ions to form nanoparticle (reducing agent). It has also been reported that metal nanoparticle was successfully synthesized from *Pinus merkusii* cone flower extract [9]. So, this study is proposed to study of silver nanoparticle formation using cone flower extract of *Pinus merkusii*, which is a simple and environmentally benign method.

2. **Experimental**

2.1. **Chemical and instrumentation**

Cone flower of *Pinus merkusii* were collected from Coban Rais, Batu city, East Java. AgNO₃ was purchased from Sigma Aldrich. UV-visible spectral analysis was recorded on a double beam spectrophotometer (SHIMADZU 1601). IR measurements were performed with a SHIMADZU 8400.
2.2. Procedure

The extract was produced by 50 g of cone flower powder boiled with 250 mL distilled water at 70°C for 2 hours. After that, the extract was filtered and stored at 4°C for further use, and the content of phenolic compound was tested phytochemically [9]. The synthesis of silver nanoparticles using Pinus merkusii cone flower extracts follows the procedure of Mo et al.,[5]. The reaction mixture was prepared by adding 25 mL of 0.1 M silver nitrate solution to 50 mL of extract (5%, 10%, 15%, 20%) at 60°C for 15, 30, 45, and 60 minutes. The color of the solution changed to brown which confirmed the reduction of silver nitrate to silver nanoparticles. The synthesized AgNP were separated by centrifugation at 10,000 rpm for 15 minute. Products were further analyzed by FTIR, UV-Vis and TEM.

3. Results and discussion

Cone flower extract of Pinus merkusii was composed of phenolic compound and confirmed by phytochemical test using FeCl₃. The extract color change from light brown to blackish-green that indicates the presence of phenolic compounds. The cone flower extract were used to reduce silver ion to silver nanoparticle. The reduction was confirmed by the color change to brown (Fig.1).

![Figure 1. Color change of AgNP](image)

Different extract concentration were used for the synthesis of silver nanoparticles and studied using UV-Vis spectroscopy (Fig. 2). The extract concentration varied from 5%, 10%，15% and 20% in 0.1 M silver nitrate solution. As a result, larger extract concentration lead to an increase in peak absorbance in UV-Vis spectrum. By increasing the extract concentrations, indicates nanoparticle production was also increased. The effect of reaction time on the synthesis of AgNP was showed at Figure 3. It can be seen that AgNP began to obtain within 30 min after the addition of silver nitrate to the Pinus merkusii cone flower extract solution. As the increased of reaction time, the intensity of peak absorbance increased gradually, indicating an increasing amount of AgNP formed. The maximum result of synthesis AgNP obtained in extract concentration 20% and reaction time 60 minutes, with absorbance maxima at 431 nm indicated the characteristic surface plasmon resonance (SPR) peak of AgNP.
Extract concentration effect

![Graph showing absorbance vs. wavelength for different concentrations of extract.](image)

**Figure 2.** UV-Vis absorption of AgNP with different concentration of extract

Reaction time effect

![Graph showing absorbance vs. wavelength for different reaction times.](image)

**Figure 3.** UV-Vis absorption spectra of AgNP as a function of the reaction time

The functional groups involved in the formation of AgNP and extract determined using FTIR spectroscopy were shown in Figure 4. The water extract of *Pinus merkusii* cone flower showed the peaks at 1640.14 cm\(^{-1}\) and 3451.18 cm\(^{-1}\). The broad peak around 3451.18 cm\(^{-1}\) in the spectra indicates the existence of O-H group of phenolics compound, and the band at 1640.14 cm\(^{-1}\) is characteristic of C=C aromatic. After nanoparticles synthesis, the band at 1640.14 cm\(^{-1}\) and 3451.18 cm\(^{-1}\) shifted to 1638.21 cm\(^{-1}\) and 3431.89 cm\(^{-1}\), and the band at 1383.63 cm\(^{-1}\) and 1032.61 cm\(^{-1}\) appear, which could be assumed that the phenolic hydroxyl groups are involved in the synthesis as reducing agent.
Figure 4. FTIR spectra of Extract (blue) and AgNP (orange)

The morphology of the synthesized AgNP using the water extract of Pinus merkusii cone flower were further confirmed by the TEM micrograph image. The images reveal that the AgNP are spherical in shape, with the particles size distribution between 9 to 23 nm (Fig. 5).

Figure 5. TEM image and size distribution of AgNP using cone flower extract of Pinus merkusii

4. Conclusion
Cone flower waste extract from Pinus merkusii, which are locally obtainable and environmentally benign were successfully used for the synthesis of silver nanoparticles. The green synthesis can obtain smaller particle size and avoided aggregation of AgNP. The characterization of AgNP was verified by FTIR, UV-Vis, and TEM. The AgNP were spherical in shape with particle size between 9 to 23 nm, respectively.
5. Acknowledgement
The authors acknowledge financial support provided by Direktorat Riset dan Pengabdian Masyarakat, Direktorat Jenderal Penguatan Riset dan Pengembangan Kementrian Riset, Teknologi dan Pendidikan Tinggi, according to the research contract number 063/SP2H/LT/DRPM/2017. The TEM image was recorded at Chemistry Department, Faculty of Science, UGM, Yogyakarta.

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