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Green Synthesis and Characterization Of Silver Nanoparticles using Dadap Serep (*Erythrina lithosperma*) Leaves Extract as a Bioreductor

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Abstract: Silver nanoparticles (AgNPs) had been widely applied in various fields such as health, food, environment, catalysts, optics, and electronics. The synthesis of silver nanoparticles can be carried using a bottom-up method using by bioreductants derived from natural extracts (green synthesis). This study aims to describe the green synthesis of silver nanoparticles using a bioreductants from dadap serep *(E. lithosperma*) leaves extract and to determine its characteristics. The synthesis was carried out by varying the volume ratio of dadap serep leaves extract to silver nitrate solution at ratios of 1: 1, 1: 2, 1: 3, and 1: 4, followed by optimization at pH 9, 10, and 11. Characterization of the synthesized AgNPs was carried out using Ultra Violet-Visible (UV-Vis) spectrophotometer, Fourier Transform Infrared (FTIR) spectrophotometer, Particle Size Analyzer (PSA), Scanning Electron Microscopy (SEM), and X-Ray Diffraction (XRD). The results showed that the optimum synthesis reaction conditions occurred at a volume ratio of 1: 4 and pH 10. The synthesized silver nanoparticles had an average particle size of 92 nm (DI = 0.3124). The FTIR spectrum showed the presence of hydroxyl groups (3242,88 cm^{-1}), aromatic C=C (1568.35 cm^{-1}), C-O (1033.25 cm^{-1}), Ag-O. (519.23 cm^{-1}). The results of XRD analysis showed typical peaks of silver atoms at 2 θ: 37.71° (111), 43.84° (200), 64.07° (220), 77.04° (311). SEM analysis showed that the silver nanoparticles had an irregular surface morphology. Thus, dadap serep (*E. lithosperma*) leaves extract has the potential to be used as a bioreductor for the synthesis of silver nanoparticles.

Keywords: Dadap serep (*E. lithosperma*) leaves extract, silver nanoparticles, green synthesis

Abstrak: *Nanopartikel perak (AgNPs) telah banyak diaplikasikan dalam beragam bidang seperti kesehatan, pangan, lingkungan, katalis, optik, dan elektronik. Sintesis nanopartikel perak dapat dilakukan secara bottom up menggunakan bioreduktor yang berasal dari ekstrak bahan alami (green synthesis). Penelitian ini bertujuan untuk mendeskripsikan sintesis hijau nanopartikel perak menggunakan bioreduktor ekstrak daun dadap serep (Erythrina lithosperma) serta menentukan karakteristiknya. Sintesis dilakukan dengan melakukan variasi perbandingan volume ekstrak daun dadap serep dan larutan perak nitrat dengan perbandingan 1 : 1, 1: 2, 1 : 3, dan 1 : 4, lalu melakukan optimasi pada pH 9, 10, dan 11. Karakterisasai AgNPs hasil sintesis dilakukan menggunakan spektrofotometer Ultra Violet-Visible (UV-Vis), spektrofotometer Fourier Transform Infrared (FTIR), Particle Size Analyzer (PSA), Scanning Electron Microscopy (SEM), dan X-Ray Diffraction (XRD). Hasil penelitian menunjukkan bahwa kondisi reaksi sintesis yang optimum terjadai pada perbandingan volume 1 : 4 dan pH 10. Nanopartikel perak hasil sintesis memiliki rata-rata ukuran partikel 92 nm (DI = 0,3124). Spektrum FTIR menunjukkan adanya gugus hidroksil (3242,88 cm^{-1}), C=C aromatik (1568.35 cm^{-1}), C-O bond (1033.25 cm^{-1}), Ag-O. (519.23 cm^{-1}). Hasil analisis XRD menunjukkan puncak khas atom perak pada 2 θ: 37,71° (111), 43,84° (200), 64,07° (220), 77,04° (311). Nanopartikel perak memiliki morfologi permukaan yang kurang teratur berdasarkan hasil analisis menggunakan SEM. Dengan demikian ekstrak daun dadap serep (E. lithosperma) berpotensi digunakan sebagai bioreduktor untuk sintesis nanopartikel perak.*

Kata kunci: *Ekstrak daun dadap serep (E. lithosperma), nanopartikel perak, sintesis hijau*

INTRODUCTION

Nanoparticles, especially metal nanoparticles, are one of the nanotechnology products that have attracted the attention of many researchers, because they can be widely applied in the fields of optics, electronics, biology, catalysts, medicine, food, and the environment (Yuliasari *et al*. 2014). One of the metals whose nanoparticles have been widely studied is silver (Ag). Silver nanoparticles (AgNPs) are one of the nanomaterials with high commercial value.

Silver has received significant attention due to its unique physical and chemical properties, including conductivity (2.3 x 105 S.m-1), thermal stability (973.22°C), and catalytic activity, which contribute to various new products and scientific applications (Chen *et al* 2017; Sampaio & Viana 2018). The unique properties and larger surface area of AgNPs have enable their widely used as antibacterial agents in industry, health, food storage, textile coatings, and a number of environmental and biomedical applications. In addition, AgNPs have also been applied as antifungal and antibacterial in various products such as socks, wet wipes, food storage containers, textiles, and others (Khaydarov *et al*. 2009; Handoko *et al*. 2022).

Nanoparticle synthesis can be carried out using the green synthesis method using plant extract bioreductants (Ahmed *et al*. 2016). This approach is more popular because it has several advantages, including being more economical, easy, a wide variety of reductants that can be used, and being environmentally friendly (Siddiqi & Husen 2016; Sugiyarti *et al*. 2021). Plant extracts that can be used as bioreductors are plants that contain phenolic secondary metabolites that can act as reducing agents for Ag^{+} ions in solution to Ag^{0} (Amin & Sutoyo 2022). The use of plant extracts for silver nanoparticle synthesis is advantageous because the materials are easily obtained and non-toxic, the chemicals used are relatively few, and a variety of secondary metabolites can be used as bioreductants for silver nanoparticles. Phenolic secondary plant metabolites that can function as reducing agents include flavonoids, tannins, and alkaloids (Siddiqi & Husen 2016).

Silver nanoparticles have been widely synthesized from plant extracts, including bay leaves (Taba *et al*. 2019), green tea leaves (Rahim *et al*. 2020), sappanwood (Amin & Sutoyo 2022), meniran leaves (Rahman & Sutoyo 2022), and banyan leaves (Rati e*t al*. 2024). However, the synthesis of AgNPs using bioreductant dadap serep leaves (*E. lithosperma*) has never been reported. This is the reason why researchers conducted research using dadap serep leaves extract as a bioreductant in AgNPs synthesis. The dadap serep plant (*E. lithosperma*) is a plant in the Fabaceae family that has been widely known by the public as a traditional medicine to treat stomach aches, skin inflammation, reduce fever, stimulate breast milk production, prevent miscarriage, and inhibit internal bleeding (Awatiszahro *et al*. 2023; Susilawai *et al*. 2023). Dadap serep leaves are known to contain phenolic secondary metabolites, namely flavonoids, alkaloids, and tannins (Utami 2019; Phukhatmuen *et al*. 2021). These phenolic compounds have the potential to act as bioreductants for AgNPs synthesis using the green synthesis method. This potential is also supported by research showing that the ethanol extract of dadap serep leaves has very strong antioxidant activity with an IC_{50} of 3.45 ppm (Chotimah *et al*. 2019).

Therefore, in this study, silver nanoparticles will be synthesized using dadap serep (*E. lithosperma*) leaves extract as a bioreductant. The synthesized silver nanoparticles will be characterized using UV-Vis spectroscopy, FTIR spectroscopy, PSA, XRD, and SEM.

MATERIALS AND METHOD

Materials

Plant material that used in this research are the dried powder of dadap serep (*E. lithosperma) leaves,* $AgNO_3$ (Merck), whatman filter paper (No. 42), and demineralized water.

Preparation of Plant Materials

Samples of the dadap serep (*E. lithosperma)* (5 kg) were collected from Plosokandang Village, Kedungwaru District, Tulungagung Regency, East Java, Indonesia. Before further investigation, the sample was identified at LIPI Purwodadi Botanical Garden, East Java. Furthermore, the sample is cleaned of attached dirt, then dried at room temperature for seven days. The dried sample was grinded into a fine powder (1.5 kg) that was ready for extraction (Sutoyo *et al*. 2021).

Preparation of AgNO3 0.01 M

Silver nitrate powder was weighed as much as 0.17 g, put into a 100 mL measuring flask, then added with distilled water up to the mark. The mixture was stirred with a magnetic stirrer to ensure the solution could dissolve homogeneously (Khoirunnisa & Sutoyo 2024).

Extraction of the *E. lithosperma*'s Leaves

The dried powder of dadap serep (*E. lithosperma*) leaves (10 g) was extracted by infution method using 100 mL of demineralized water as solvent. The mixture was heated at 50°C for 30 minutes while stirring continuously. The mixture was then cooled and filtered using Whatmann No. 42 filter paper, thus obtaining *E. lithosperma* leaves extract which will be used as a bioreductor (Taba *et al*. 2019).

Synthesis of Silver Nanoparticles (AgNPs)

The synthesis was carried out by mixing dadap serep (*E. lithosperma)* leaves extract with $AgNO_3$ 0.01 M with variations in the volume ratio of the extract to the $AgNO_3$ solution of 1:1 (30 mL: 30 mL), 1:2 (20 mL: 40 mL), 1:3 (15 mL: 45 mL) and 1:4 (12 mL: 48 mL). The mixture was stirred using a magnetic stirrer at a speed of 1500 rpm for 30 minutes at a temperature of 50°C. Each mixture was optimized at pH 9, 10, and 11, by adding NaOH 0.1 M dropwise. the UV-Vis spectrum was measured to determine the maximum absorption wavelength and absorbance, so that a mixture with the optimum composition and pH was obtained. The mixture with the optimum composition and pH was centrifuged at a speed of 5000 rpm for 30 minutes. The silver nanoparticle precipitate was separated by decantation and then dried using a freeze dryer (Martin Christ Alpha 1-2) for 8 hours. The synthesized silver nanoparticles were characterized using UV-Vis spectroscopy (Shimadzu UV-1800), FTIR spectroscopy (Perkin Elmer), PSA (Biobase BK-802N), XRD (Shimadzu XRD-6000), and SEM (Hitachi Flex SEM 100) (Khoirunnisa & Sutoyo 2024).

Characterization of Silver Nanoparticles (AgNPs)
UV-Vis Spectrophotometer

A UV-Vis spectrophotometer (Shimadzu UV-1800) was used to determine the maximum absorption wavelength of the synthesized silver nanoparticles. The formation of silver nanoparticles is indicated by the appearance of an absorption peak at a wavelength between 400-500 nm . Measurements were carried out by taking a 1 mL sample of the synthesized silver nanoparticles and diluting it with 9 mL of demineralized water, followed by scanning the wavelength range from 300 and 700 nm (Manosalva *et al*. 2019; Widatalla *et al*. 2022).

FTIR Spectrophotometer

The presence of functional groups in the synthesized silver nanoparticles was analyzed using an FTIR spectrophotometer (Perkin Elmer). Measurements were performed by placing a 1.0 mg solid sample of the synthesized silver nanoparticles onto the sensor. After the sensor was covered, the infrared spectrum was recorded in the range of 400 and 4000 cm^{-1} (Chand 2019).

Particle Size Analyzer (PSA)

The diameter or size of the synthesized silver nanoparticles was analyzed using a PSA instrument (Biobase BK-802N). The analysis was carried out by dissolving 0.1 g of the synthesized silver nanoparticles in 5 mL of distilled water, followed by sonication for 15 minutes. The solution was then placed in a portable cuvette and inserted into the PSA instrument (Putri & Sutoyo 2025).

X-Ray Diffraction (XRD)

An XRD instrument (Shimadzu XRD-6000) was used to obtain information on the degree of crystallinity (determination of the crystal-amorphous structure) and the crystal orientation (hkl) of the synthesized silver nanoparticles. The instrument was equipped with a Cu radiation source and a Ni filter and was set at 30 kV/30 mA. The analysis was carried out by taking 0.1-1.0 g of the synthesized silver nanoparticles solid and grinding them into a fine powder (10-50 µm). XRD data were collected under the same experimental conditions in the angular range of $20° \leq 2\theta \leq 80°$ (Amer & Awwad 2021; Rahmawati *et al*. 2024).

Scanning Electron Microscopy (SEM)

An SEM instrument (Hitachi Flex SEM 100) was used to obtain information on the surface morphology of the synthesized silver nanoparticles. The analysis was carried out by attaching a solid sample of the synthesized silver nanoparticles to a specimen holder using double-sided carbon tape. The sample was coated with gold for 1 minute to improve its conductivity. After being placed in a vacuum chamber, the sample was scanned to observe the surface morphology of the synthesized silver nanoparticles (Julinawati *et al*. 2015).

RESULT AND DISSCUSION
Optimation composition and pH in synthesis silver nanoparticles and the UV-Vis Analysis

In this research, silver nanoparticles were synthesized using variations in the volume composition of *E. lithosperma* extract and 0.01 M $AgNO_3$ solution of 1: 1, 1: 2, 1: 3, and 1: 4 as well as at pH variations of 9, 10, and 11. UV-Vis spectroscopy was used to determine the optimum composition and pH value. Based on the UV-Vis measurements of the four composition variations and three pH variations, the data obtained are presented in Table 1 and the UV-Vis spectrum at the optimum composition and pH are presented in Figure 1.

Based on the Table 1, in the mixture composition of 1:4, silver nanoparticles were successfully produced because the absorbance values of the mixtures were within the range of 400-500 nm, namely 421 nm (pH 9), 424 nm (pH 10) and 420 nm (pH 11) (Widatalla *et al.* 2022). If the silver ions in the mixture have not been reduced to silver nanoparticles, an absorption wavelength of the silver nitrate solution will appear at 305 nm (Bagur *et al.* 2020). The optimum composition and pH in the synthesis of silver nanoparticles using *E. lithosperma* leaves extract bioreductant are a composition of 1:4 and pH 10. This composition produced the most silver nanoparticles because it had the highest absorbance value (1.880) (Sulistiorini *et al.* 2024). Dadap serep leaves extract contains phenolic compounds, namely flavonoids, alkaloids and tannins which have a high reducing ability under alkaline conditions. In these conditions, the phenolic hydroxyl group will react with sodium hydroxide solution to produce phenolate ions whose reducing ability to convert silver ions (Ag^+) into silver atoms (Ag^0) is higher than that of the phenolic hydroxyl group. Consequently, a greater amount of silver nanoparticles is produced as indicated by the higher absorbance values (Utami 2019; Phukhatmuen *et al.* 2021).

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Table 1. The UV-Vis absorbance of synthesized silver nanoparticles

Composition	Absorbance value at pH		
	9	10	11
1 : 1	1.037	1.805	0.908
1 : 2	1.010	1.010	0.999
1 : 3	1.390	1.495	1.301
1 : 4	1.825	1.880	1.653

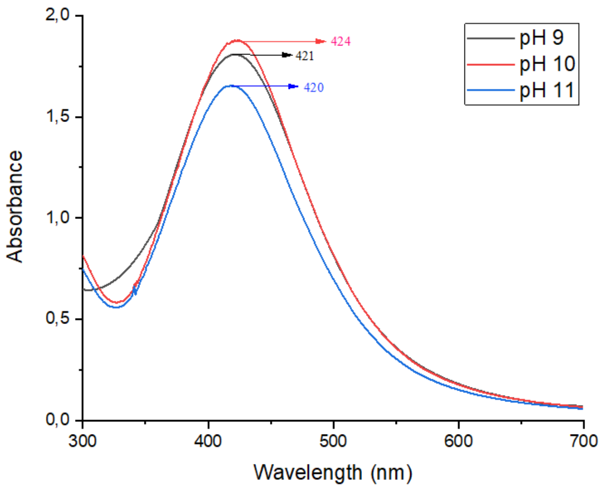

Figure 1. The UV-Vis spectrum of synthesized AgNPs at optimum composition (1 : 4) with various of pH

Particle Size Analyzer (PSA) of Silver Nanoparticles

The particle size of the synthesized silver nanoparticles under optimum conditions was determined using a Particle Size Analyzer (PSA).

Based on the measurement results, the synthesized silver nanoparticles had an average particle size of 92 nm, with a Dispersion Index (DI) = 0.3124. The PSA results indicated that the synthesized silver nanoparticles were in the nanoscale range, as the particle size was between 1-100 nm (Sutoyo *et al*. 2021; Dua *et al*. 2023). A Dispersion Index value lower than 0.5 indicated that the synthesized silver nanoparticles have a homogeneous particle size distribution. The PSA measurement results are presented in Figure 2.

Fourier Tranform Infrared Spectroscopy (FTIR) Analysis of Silver Nanoparticles

FTIR spectroscopy was used to determine functional groups and the changes occuring in those functional groups during the synthesis of silver nanoparticle synthesis using dadap serep leaves extract as a bioreductant. The FTIR spectrum of the synthesized silver nanoparticles showed stretching vibrations of hydroxyl groups (3242,88 cm^{-1}), aromatic C=C bonds (11568.35 cm^{-1}), C-O bond (1033.25 cm^{-1}), and Ag-O bonds (519.23 cm^{-1}). The appearance of the Ag-O vibration peak confirmed that silver nanoparticles had been successfully synthesized using dadap serep serep (*E. lithosperma)* leaves extract as a bioreductor (Dua *et al*. 2023). The FTIR spectrum of dadap serep (*E. lithosperma)* leaves extract showed absorption bands of stretching vibrations of hydroxyl groups (3235.32 cm^{-1}), C=O bond (1705.72 cm^{-1}), aromatic C=C bonds (1590.17 cm^{-1}), and C-O bonds (1035.97 cm^{-1}). The presence of hydroxyl groups, C=O bond and aromatic C=C bonds in the FTIR spectrum of dadap serep (*E. lithosperma)* leaves extract indicated the content of phenolic compounds in the extract which act as bioreductors. The decrease in the absorption intensity of the hydroxyl group from the extract to silver nanoparticles indicated that the phenolic hydroxyl group were involved in the reduction of silver ions (Ag^+) to silver atoms (Ag^o) (Sulistiorini *et al*. 2024; Rati *et al*. 2024). The FTIR spectrum of silver nanoparticles and dadap serep (*E. lithosperma)* leaves extract are presented in Figure 3.

X-Ray Diffraction (XRD) Analysis of Silver Nanoparticles

The silver nanoparticles synthesized using dadap serep leaves extract as a bioreductant were characterized using XRD to determine the presence of silver atoms in the nanoparticles. This measurement was carried out using Cu Kα radiation with a wavelength of 1.5406 Å at 40 kV and 30 mA with a scan rate of 0.02 cm^{-1}. The diffractogram of the measurement results with the XRD instrument is presented in Figure 4.

The peaks appearing on the diffractogram were compared with the ICDD data center No. 03-065-2871 (2θ Ag : 38.1°, 44.3°, 64.4°, and 77.4°). The 2θ

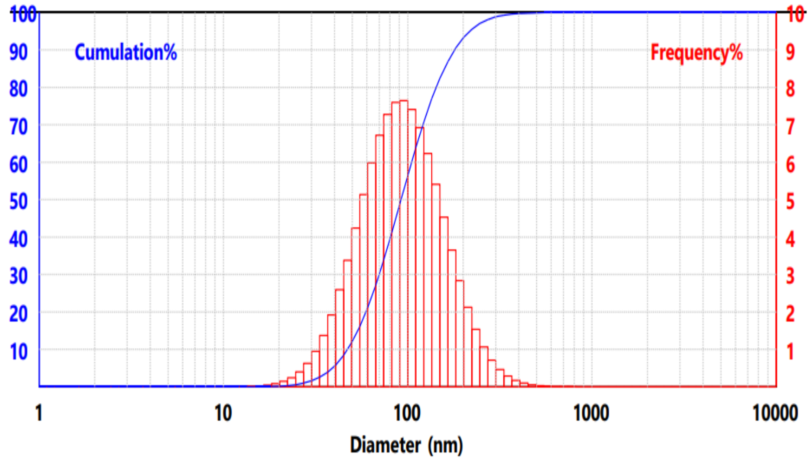

Figure 2. Characterization results of synthesized AgNPs by PSA

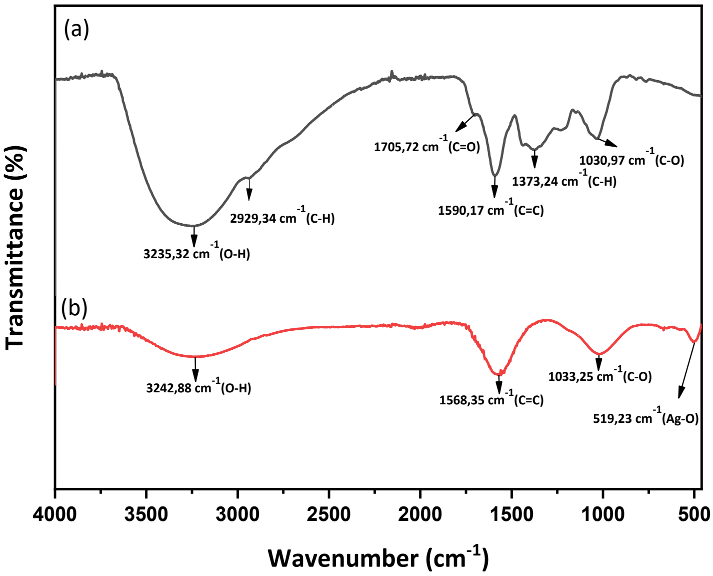

Figure 3. (a) FTIR spectra of *E. lithosperma* leaves extract; (b) synthesized AgNPs.

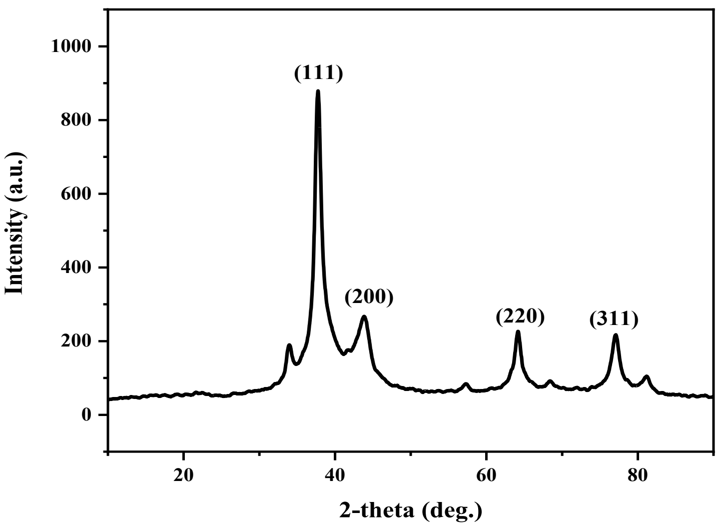

Figure 4. XRD diffractogram of synthesized AgNPs

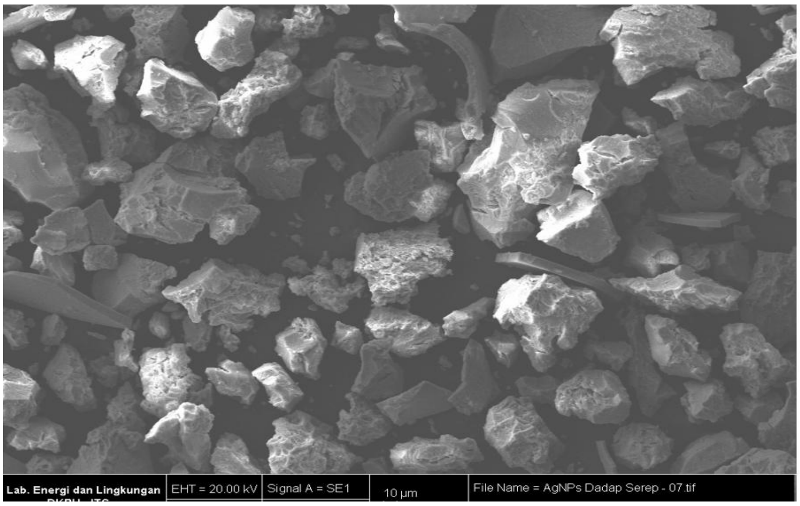

Figure 5. Scanning electron microscopy (SEM) images of synthesized AgNPs

Ag peaks obtained from the XRD analysis of the synthesized AgNPs were observed at 37.71^{o} (111), 43.84^{o} (200), 64.07^{o} (220), and 77.04^{o} (311), indicating a cubic crystal system. These results were supported by the study of Azarbani *et al*. (2020) who reported 2θ Ag peaks at 38.45^{o} (111), 44.54^{o} (200), 64.64^{o} (220), 77.65^{o} (311), and by Sampaio & Viana (2018) who obtained 2θ Ag peaks at 38.24^{o} (111), 44.32^{o} (200), 64.56^{o} (220), 77.52^{o} (311). Therefore, it can be confirmed that the synthesized nanoparticles contain silver atoms or AgNPs.

Scanning Electron Microscopy (SEM) Analysis of Silver Nanoparticles

Characterization using Scanning Electron Microscopy (SEM) was conducted to determine the surface morphology of the nanoparticles. Based on the SEM analysis of silver nanoparticles synthesized using dadap serep leaves extract as a bioreductant, the result obtained is presented in Figure 5.

The SEM analysis showed that the synthesized silver nanoparticles had an irregular shape. The irregular morphology of silver nanoparticles was also reported by other researchers, including Taba *et al* . (2019); Rahim *et al.* (2020); Kinjal *et al*. (2024) who synthesized silver nanoparticles using bioreductants from leaves extract of *Syzygium polyanthum*, green tea (*Camellia sinensis*), and *Azadirachta indica*, recpectively.

CONCLUSION

Dadap serep leaves extract can be used as a bioreductor in the synthesis of silver nanoparticles (AgNPs). The optimum synthesis reaction conditions occured at a volume ratio of 1: 4 and pH 10. The synthesized silver nanoparticles had an average particle size of 92 nm, hydroxyl groups, aromatic C=C bond, C-O bond, and Ag-O bond. The synthesized AgNPs showed typical peaks of silver atoms at 2 θ: 37.71^{o} (111), 43.84^{o} (200), 64.07^{o} (220), 77.04^{o} (311). Silver nanoparticles had less regular surface morphology, based on the results of SEM analysis.

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