Eksakta : Berkala Ilmiah Bidang MIPA
VOLUME 23 NO 04 2022, pp 255-265
ISSN : Print 1411-3724 — Online 2549-7464
DOI : https://doi.org/10.24036/eksakta/vol23-iss04/337

Eksakta
Berkala Ilmiah Bidang MIPA
http://www.eksakta.ppj.unp.ac.id/index.php/eksakta

Article

Green Synthesis and Antibacterial Activity of Silver Nanoparticles using Turi (*Sesbania grandiflora* Lour*)* Leaf Extract

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Article Info

Article history :

Received August 31, 2022
Revised September 15, 2022
Accepted September 25, 2022
Published December 30, 2022

Keywords :

Ag Nanoparticles, green synthesis, turi leaves

Abstract. Research has been carried out on the ability of antibacterial activity with using Ag nanoparticles with a variation of the $AgNO_3$ mole ratio of 0.5 mM and 1.5 mM as much as 90 ml and the amount of turi leaf extract added 1 mL. The material was characterized by UV-vis spectroscopy. Determination of antibacterial activity was carried out through Escherichia coli bacteria after interacting with nanoparticles Ag. Green synthesis of silver nanoparticles can be carried out using an aqueous extract of turi leaves, with the optimum concentration of 0.5 mM $AgNO_3$ and 1.5 mM $AgNO_3$ being synthesized for one day at room temperature. Resulting in silver nanoparticles with energy band gap values of 3.9 eV and 3.88 Ev having antibacterial activity of Escherichia coli with inhibitory power of 5.52 mm and 6.65 mm, respectively.

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1. Introduction

Nanotechnology has attracted the attention of researchers in recent years because of its wide application and can be applied in various fields. Nanoparticles are foundation in nanotechnology and has wide applications compared to other materials [1-3]. Based on the results of the study showed that carbon-based nanomaterials such as graphene oxide nanoparticles [4-5], carbon nanotubes [6-7] and fullerenes[8-10], metal nanoparticles [11-12], magnetite nanoparticles [13-14], etc. have strong antimicrobial properties. The more advantageous use of silver nanoparticles is due to their relatively low cost, availability and has excellent antifungal, antibacterial, anticancer properties [15-18].

Nanotechnology is one of the most dynamic materials science subjects, and the production of nanoparticles is increasing rapidly worldwide. There are two types of nanoparticles that can be synthesized, namely inorganic and organic nanoparticles. Inorganic nanoparticles including metals nanoparticles (such as Au, Ag, Cu, Al), types of nanoparticles (such as Co, Fe, Ni), and semiconductor nanoparticles (such as ZnO, ZnS, CdS), whereas organic nanoparticles include carbon nanoparticles (such as quantum dots, carbon nanotubes) [19-22].

Nanoparticles are particles that are 1–100 nm in size [23-24]. there are two methods synthesis of nanoparticles, namely physical methods and chemical methods [1][25]. Synthesis of nanoparticles using chemical methods has a detrimental impact because some chemicals can damage the environment. However, both of these methods use materials Excessive chemicals that can cause environmental pollution, and need the cost is not cheap for the process [24-25]. The green synthesis nanoparticle method is synthesis method that forms nanoparticles metal with the help of naturally derived materials of organisms (plants and microorganisms) both land and sea [26-27]. Biosynthesis of nanoparticles such as synthesis by using microbes [28-29], enzymes [30-31], algae [32-33] and plants [34-35] are more profitable because they do not involve chemicals so they are environmentally friendly.

Plants contain bioactive compounds such as secondary metabolites, such as groups of terpenoids and flavonoids can play a role in the reduction process metal ions [36-37]. Sesbania grandiflora L. (also known as turi leaf. syn. *Aeschynomene grandiflora*) belonging to the family *Fabaceae*. turi leaf is famous green vegetables and also traditional medicinal plants In Indonesia, Grandiflora has antibacterial, antifungal, antidiabetic, antioxidant and antitumorigenic activities[38-39]. There are various types of plant groups that contain secondary metabolites, one of which is turi leaves. Turi leaves contain alkaloids, saponins, quinones, phenolics, triterpenoids, steroids and flavonoids [40-42].

The most important thing in the synthesis of nanoparticles is through an environmentally friendly and green approach in the synthesis of nanoparticles [43-44]. There is an article that reports synthesis of silver nanoparticles using water extract of *Sesbania grandiflora* leaves. The formation of Ag can be observed visually through the formation of a yellow color as a marker for the formation of silver nanoparticles using the UV-Vis spectrum. Silver nanoparticles exhibit antimicrobial activity of tested Ag tested against the selected pathogen and a positive test result is achieved. As a result, the synthesized Ag exhibited effective antimicrobial activity for innovative applications in the medical field [45-46].

The aim of this research is to synthesize nanoparticles, using a method that actually uses inexpensive and renewable materials. In In this study, we report one synthesizing silver nanoparticles using a bioreductant of turi leaf extract. Here, silver nitrate is used as a silver precursor, and turi leaf extract as a stabilizer as well as a reducing agent. Silver nanoparticles were tested with a UV-Vis spectrum and tested for its antibacterial activity against *Escherichia Coli* Bacteria.

2. Experimental Section

2.1. Materials

The materials used in this study were turi leaves, silver nitrate ($AgNO_3$), aquades, ethanol, Whatman paper, disc paper, nutrient broth (NB), nutrient agar (NA), bacterial suspension *Eschericia coli*.

2.2. Procedure

2.2.1 Sample Preparation

Turi leaves are washed with running water and then with distilled water, cut into small pieces and then dried in the oven. After drying, the turi leaves are blended until they become powder and then sieved with a 50 mesh sieve.

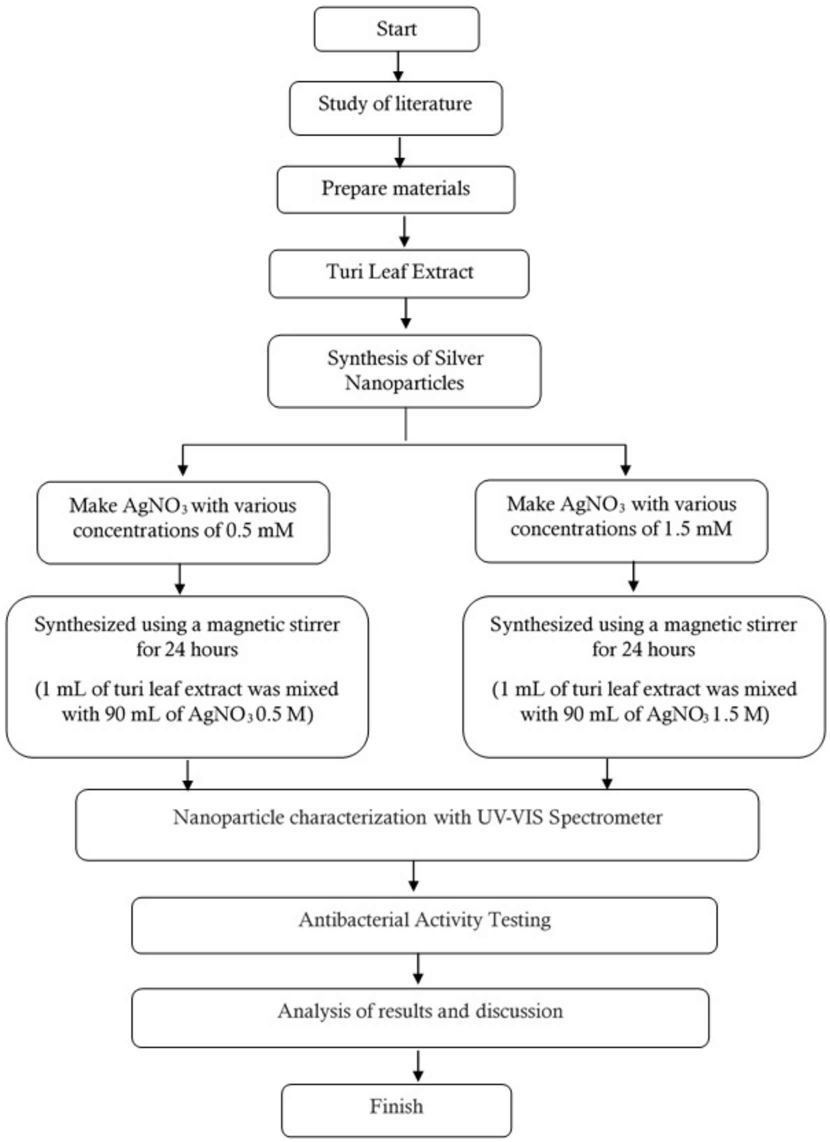

Figure 1. Research flow chart

2.2.2 Sample Extraction

10 grams of turi powder were mixed with 100 ml of distilled water and boiled for 2 hours at 80 C. During the process, a light brown solution was formed. Then the prepared extract was allowed to cool at room temperature and finally, it was filtered using Whatman filter paper [36-37].

2.2.3 Synthesis of Silver Nanoparticles

The synthesis of silver nanoparticles refers to the procedure developed. Silver ions were reduced to nano size using plant extract bioreductors. 1 mL of turi leaf extract was mixed with 90 mL of $AgNO_3$ with various concentrations of 0.5 mM and 1.5 mM synthesized using a magnetic stirrer for 1 day until a color change occurred [47-48]. The formation of silver nanoparticles is indicated by a change in the color of the solution to brown yellow. Furthermore, the absorbance value and max are known by UV-Vis spectroscopy.

2.2.4 Measurement of UV-Vis Spectrophotometer

The formed silver nanoparticles were characterized by UV-Vis spectrophotometer aimed at determining the formation of silver nanoparticles and their stability. The UV-Vis spectrophotometer characterization refers to the research of with a range of 300 nm-600 nm using a quartz cuvette with distilled water blank. The reduction of silver ions is known from UV-Vis spectra [49-50].

2.2.5 Antibacterial Activity Testing

The antibacterial activity test was carried out qualitatively referring to the research of [51-52].The qualitative test was carried out by observing the inhibition zone by immersing the paper disc into colloidal silver nanoparticles and then pasting it on the surface of the Nutrient Agar that had grown the test bacteria. Nutrient Agar which has been attached to disc paper is incubated for 24 hours at 37^{o}C.

3. Results and Discussion

3.1 Synthesis of Silver Nanoparticles using Turi Leaf Extract

$AgNO_3$ solution of 1.5 mM and 0.5 mM was pipetted as much as 90 mL and each solution was put into a 250 mL Erlenmeyer, then 1 mL of turi leaf extract was added. The mixture was stirred with a magnetic stirrer until a color change occurred, then analyzed using a UV-Vis spectrophotometer. Figure 3.1 shows the results of the synthesis of Ag nanoparticles with $AgNO_3$ concentrations of 0.5 mM and 1.5 mM.

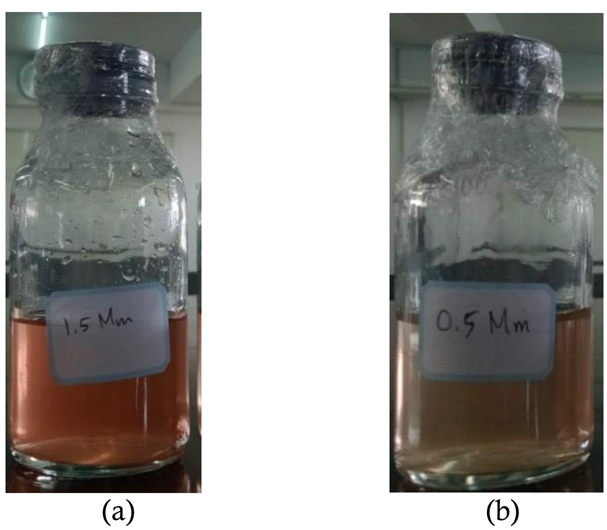

Figure 2. The results of the synthesis of Ag nanoparticles with $AgNO_3$ concentrations of (a) 0.5 mM and (b) 1.5 mM

Based on the UV-Vis spectrum testing of the colloidal solution, it was confirmed that silver nitrate was reduced to silver nanoparticles by turi leaf extract. Turi leaf extract was added to the stirred silver nitrate solution at room temperature for 24 hours. The color of the solution changed from transparent to red-brown indicating that silver nanoparticles were formed. There are many different phytoconstituents of leaf extracts that cause discoloration.

3.2 Analysis of the Optical Properties of Silver Nanoparticles with Turi Leaf Extract

UV-Vis spectrum characterization was carried out to analyze the formation of silver nanoparticles through the absorbance spectrum obtained from each sample. The maximum absorbance value of silver nanoparticles is obtained at a wavelength of 200-290 nm.

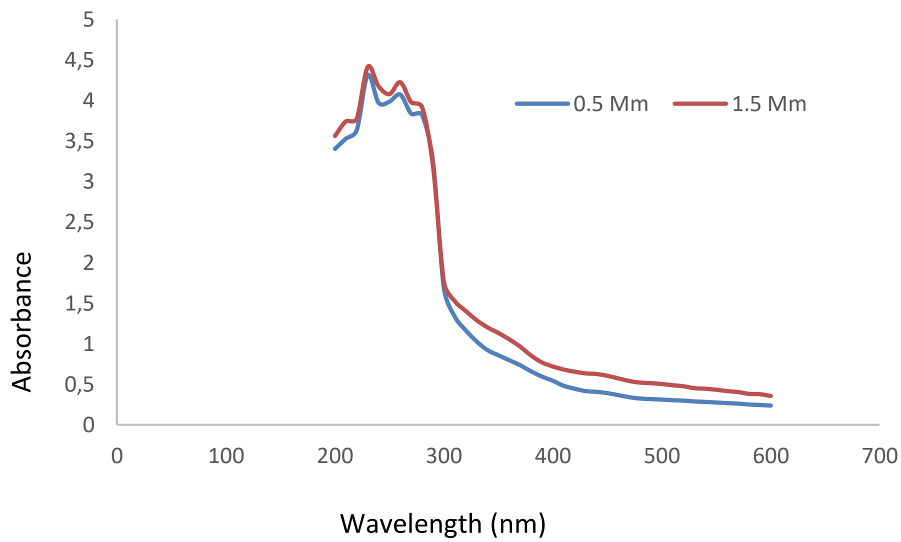

Figure 3. Absorbance spectrum of silver nanoparticles turi leaf extract with $AgNO_3$ concentrations of 0.5 mM and 1.5 Mm

Based on the absorbance spectrum of silver nanoparticles of turi leaf extract with $AgNO_3$ concentration of 0.5 mM and 1.5 Mm, it shows that the highest absorbance peak is at a wavelength of 230 nm with a maximum absorbance value of 4,315 for silver nanoparticles of turi leaf extract with $AgNO_3$ concentration of 0.5 mM. and 4,415 with silver nanoparticles of turi leaf extract with $AgNO_3$ concentration of 1.5 mM. Peak absorbance of Ag nanoparticles shift to long the larger wave is at about the length wave 200-290 nm [36-37]. Based on visual observation, the more $AgNO_3$ concentration increases, the color of the solution becomes more brown. The results of previous studies show that the color change informs the formation of silver nanoparticles [53-54]

Based on UV-Vis spectrophotometer measurements, the energy band gap of Ag nanoparticles can be measured. The energy band gap of Ag nanoparticles was obtained by plotting absorption data using the direct transition equation. Plot $(\alpha h\upsilon)^2$ vs $h\upsilon$ is shown in figure 3.3, with extrapolating the linear part of the curve to the line zero absorption gives the energy band gap value for direct transition [38-39].

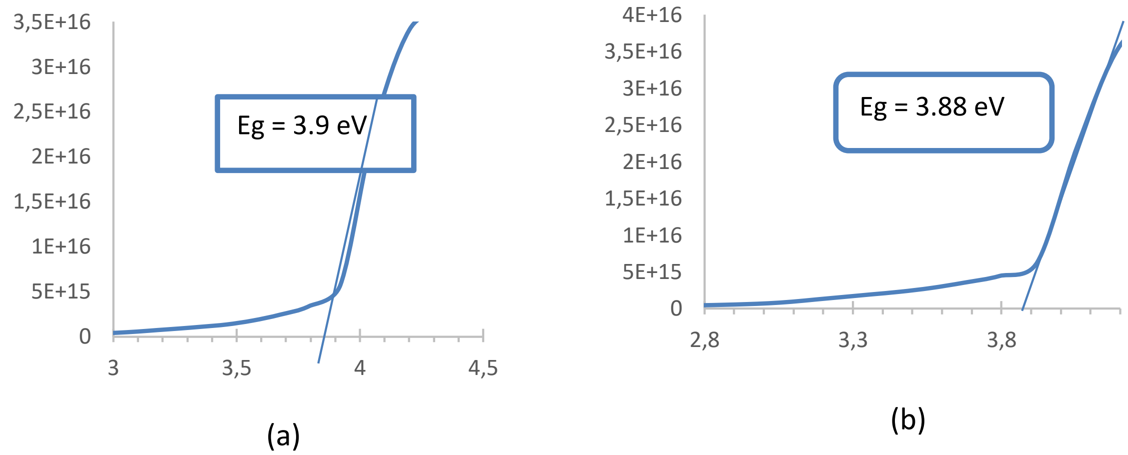

Figure 4. (a) Energy band gap of Ag nanoparticles with $AgNO_3$ concentrations of 0.5 mM and (b) Energy band gap of Ag nanoparticles with $AgNO_3$ concentrations of 1.5 Mm

Based on the measurement of the energy band gap, it shows that the greater the concentration of $AgNO_3$ produces a narrower energy band gap. Based on the measurement results of the UV-Vis spectrum of the silver nanoparticle solution of turi leaf extract with $AgNO_3$ concentrations of 0.5 mM and 1.5 Mm, the energy band gap value was obtained. Silver nanoparticles of turi leaf extract with $AgNO_3$ concentration of 0.5 mM have an energy band gap of 3.9 Ev and silver nanoparticles of turi leaf extract with $AgNO_3$ concentration of 1.5 mM have an energy band gap of 3.88 Ev.

3.3 Silver Nanoparticle Antibacterial Activity Test

Results of antibacterial activity testing with paper disc method testing of antibacterial activity was carried out by diffusion method using paper disc. The bacteria used is *Escherichia coli*. The silver nanoparticles tested were turi leaf extract silver nanoparticles with 0.5 mM $AgNO_3$ concentration which were synthesized for 1 day and turi leaf extract silver nanoparticles with 1.5 mM $AgNO_3$ concentration which were synthesized for 1 day. In this antibacterial activity test, 90 ml of $AgNO_3$ solution with concentrations of 0.5 mM and 1.5 mM and 1 ml of turi leaf extract were also tested for antibacterial activity. Both of these samples are materials or initial compositions that form silver nanoparticles, so it is necessary to analyze their antibacterial activity. These two samples were compared to see if there was an increase in antibacterial inhibition after these two components formed silver nanoparticles.

The incubation process was carried out at 37°C for 48 hours. After 48 hours of incubation, the diameter of the inhibition zone was measured using a caliper. Table 4 shows the results of the measurement of the inhibition zone on the results of the antibacterial activity test. Table 4.3 shows the results of the measurement of the inhibition zone on the results of the antibacterial activity test.

Table 1. Antibacterial activity test results

No	Sample Nanoparticles $AgNO_3$ (mM)	Replication	Sample Inhibitory			
			D1 (mm)	D2 (mm)	Amount	Average (mm)
1	0.5 mM	1	5.50	5.00	10.50	5.25
		2	6.00	5.30	11.30	5.65
		3	6.10	5.20	11.30	5.65
2	1.5 mM	1	6.10	7.50	13.60	6.80
		2	7.10	7.00	14.10	7.05
		3	6.10	6.10	12.20	6.10

In the test, it was found that the silver nanoparticle solution of turi leaf extract showed an inhibition zone on Escherichia coli bacteria. The inhibition zone produced by Silver Nanoparticles of Turi Leaf Extract with $AgNO_3$ Concentration of 0.5 mM was 5.52 mm, while the inhibition zone of Silver Nanoparticles of Turi Leaf Extract with 0.5 mM $AgNO_3$ Concentration was 6.65 mm. this shows that the greater the concentration of $AgNO_3$ the greater the inhibition zone of Escherichia coli bacteria.

The mechanism of reduction by the content of compounds in plants to metal salts can be explained in several stages. The first phase is the phase in which the reduction of metal ions and nucleation of the reduced metal atoms, followed by a spontaneous coalition of a number of adjacent nanoparticles to form particles with a larger size, along with the increase in the thermodynamic stability of the nanoparticles, this process refers to the Ostwal ripening principle. The final phase is where the final shape of the nanoparticles is formed [38-39].The results of previous studies explain that Silver nanoparticles also attack DNA mechanisms in cells. based on microscopic analysis of *Escherichia coli (E. coli)* and *S. aureus* revealed that bacterial cell DNA had shrunk and viscous after treatment with silver nanoparticles [55-56]. The results show that nanoparticles can inhibit the growth of microorganisms, including human and animal pathogens such as E. *coli*, S. *typhi* or S. *aureus* [57-58].

4. Conclusion

Green synthesis of silver nanoparticles can be carried out using an aqueous extract of turi leaves, with the optimum concentration of 0.5 mM $AgNO_3$ and 1.5 mM $AgNO_3$ being synthesized for one day at room temperature. resulting in silver nanoparticles with energy band gap values of 3.9 eV and 3.88 Ev having antibacterial activity of Escherichia coli with inhibitory power of 5.52 mm and 6.65 mm, respectively.

5. Acknowledgement

The researchers thank the Harapan Bersama Polytechnic for the research or sponsors who provide financial support

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