

***Uncaria gambir* Roxb. mediated green synthesis of silver nanoparticles using diethanolamine as capping agent**

A Labanni¹, Zulhadjri¹, D Handayani² and S Arief¹

¹ Material Laboratory, Chemistry Department, Universitas Andalas, Limau Manis 25163, Padang, Indonesia.

² Laboratory of Sumatran Biota, Faculty of Pharmacy, Universitas Andalas, Limau Manis 25163, Padang, Indonesia.

Email: Syukriarief@gmail.com

Abstract. Studies of silver nanoparticles preparation has been developed increasingly due to the wide application in various areas and field, such as medicine, energy, catalysis, and electronic. An environmental-friendly method is needed to fabricate biocompatible silver nanoparticles without producing hazardous materials to the environment. In this study, we synthesized silver nanoparticles by green synthesis method, using leaf extract of gambir (*Uncaria gambir* Roxb.) as bioreducing agent and aqueous diethanolamine (DEA) solution as capping agents. The AgNO₃/DEA molar ratio was varied to investigate the effect of DEA concentration to the properties of silver nanoparticles. The formation of silver nanoparticles was indicated by colour changes to yellowish brown and confirmed by result of UV-Vis spectrophotometer analysis which shown absorption band at 400 to 410 nm. The absorbance was increased to the reaction time of 24 hours, and was decrease by the increasing of DEA concentration in reaction. TEM analysis showed that prepared silver nanoparticles were spherical in shape with diameter of 3,5 - 45,5 nm. The diameter of DEA capped silver nanoparticles was 13 nm, smaller than uncapped silver nanoparticles which was 26 nm It exhibited good stability to time reaction of one month which was potential to be developed in some fields.

1. Introduction

A nanoparticle is a microscopic particle with size of less than 100 nm [1]. In recent years, the study of nanoparticles has drawn the attention of scientists due to the excellent properties which bridge the gap between bulk and atomic or molecular structures. The unique properties include chemical stability, conductivity, and catalytic activity have made nanoparticles developed in some wide areas and application such as energy, environment, medical, sensor, catalysis [2]. Among all metal nanoparticles, silver has been increasingly developed due to the potential as antibacterial agents to be developed in medical utilizing. The preparation of silver nanoparticles have been carried out using various method, such as chemical reduction [3], electrochemical [4], sonochemical [5], microwave method [6], etc. Among all the methods, the chemical reduction method is the easiest and the most rapid way to conduct.

Unfortunately, the chemical reduction method employs chemical reagent to reduce Ag⁺ ions to Ag nanoparticles which can endanger the environment and threaten the living organisms. In addition, the



using of hazardous material will result in a low biocompatibility in the application. Therefore, an environmental method is developed by the scientists, called green synthesis using nonhazardous materials such as plants, bacteria, algae, etc. This method is considered as an environmentally friendly, economical, easy, low energy, and rapid method [7].

Uncaria gambir Roxb. is one of the commodities in West Sumatera used as materials for plywood. It is also used as traditional medicine for diarrhea, toothache, and fever due to the content of catechin which can act as a natural antioxidant [8]. In the synthesis of nanoparticles, catechin is expected to reduce Ag^+ ions to Ag^0 nanoparticles. Beside reducing agent, the reaction also involves capping agent to control the growth and size of the nanoparticles. In the previous study, we prepared silver nanoparticles using hydrothermal method with *Uncaria gambir* Roxb. as bioreducing agent, resulting colloidal silver nanoparticles with a diameter of 25 nm [9]. However, we need a simple route synthesis to investigate the optimum condition of capping agent and the effect on the characteristic of the prepared nanoparticles. In the present study, we synthesized silver nanoparticles using gambir leaf extract as bioreducing agent and diethanolamine in aqueous solution with various mole ratio of Ag^+/DEA in a facile and simple route.

2. Experimental

2.1. Materials

Fresh leaves of *Uncaria gambir* Roxb. were collected from Experimental Garden, Andalas University, Padang, Indonesia. Silver nitrate (AgNO_3) as precursor and diethanolamine (DEA) as capping agent were purchased from Merck, and distilled water.

2.2 Gambir leaf extract preparation.

Gambir leaf extract was prepared using modified method of Arief et al (2015) [9]. Firstly, the fresh and healthy leaves of *Uncaria gambir* Roxb. were washed thoroughly using fresh water and distilled water to remove contamination and unnecessary materials. The clean leaves then dried in shadow condition at room temperature for several days. The dried leaves then were ground to obtain gambir powder. The aqueous extract of gambir was made by dissolving the fine powder into deionized water with ratio of 1:10 g/ml. The mixture then was stirred for 2 hours at temperature of 65 °C. After cooling, the mixture then was filtered to get the extract and was stored at temperature of 4 °C for further use.

2.3 Silver nanoparticles preparation

Gambir leaf extract was added to a 100 ml Erlenmeyer flask containing diethanolamine while was stirring in a constant rate at room temperature. The silver nitrate then was added to the mixture with variation of AgNO_3 : MEA molar ratio was 1:10, 1:20, and 1:30 which were coded as AgNps-DEA1, AgNps-DEA2, and AgNps-DEA3. The reaction was also carried out without capping agent as a comparator, coded as AgNps0. The mixture solution was continuously monitored using UV-Vis spectrophotometer in wavelength range of 200-700 nm. The obtained colloidal silver nanoparticles then stored in an airtight bottle at room temperature for further measurements.

2.4 Characterization

The optical properties of colloidal silver nanoparticles were obtained using Thermo Scientific Evolution 201 UV-Vis spectrophotometer at wavelength ranges of 200-700 nm. The morphological properties were analysed using JEOL-4000 Transmission Electron Microscopy (TEM) operated at 120 kV.

3. Results and Discussion

3.1 UV-Vis spectroscopy study

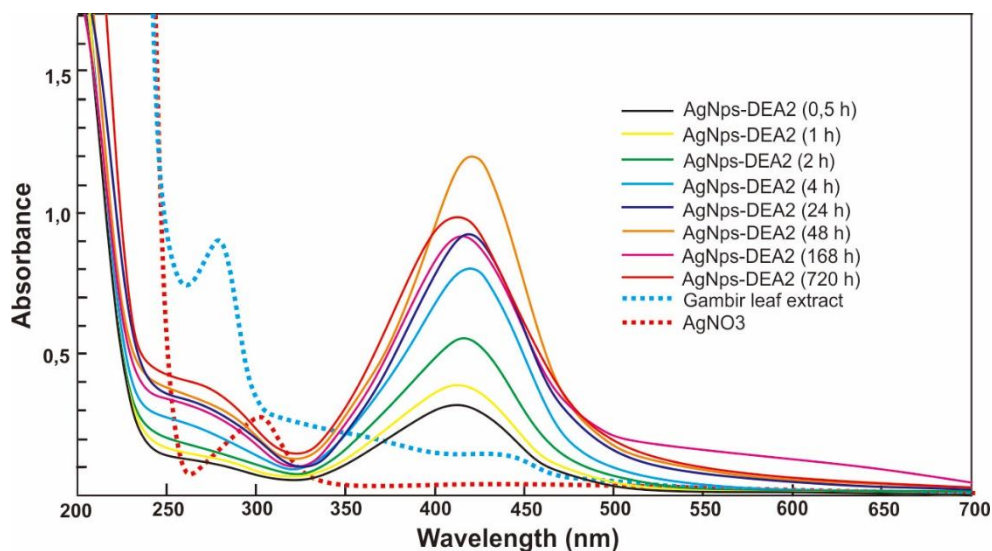


Figure 1. UV-Vis analysis of DEA stabilized gambir leaf extract mediated colloidal AgNps

The reaction was conducted in various molar ratio of AgNO₃/DEA of 1:10, 1:20, and 1:30 based on the preliminary study. The formation of silver nanoparticles was visually recognized by colour changing from pale yellow to yellowish brown with the increasing of intensity as the increasing of reaction time. This was supported by UV-Vis spectrophotometer analysis. Gambir leaf extract showed a peak at 278 nm which referred to the presence of catechin [10], while AgNO₃ exhibited a peak at 301 nm (figure 1). These two peaks were shifted to the wavelength at 398-438 nm (Table 1) as colour changing, which strongly indicated a formation of silver nanoparticles. It was due to the reduction of Ag⁺ to Ag⁰ by catechin in gambir leaf extract. It provided a specific band of nanoparticles material, called Surface Plasmon Resonance (SPR) phenomenon which is specifically shown by a peak at wavelength of 395-420 nm for silver nanoparticles [11]. Previous reports about green synthesis of silver nanoparticles using *Catharanthus roseus* Linn. G. Donn. [12] and *Cleistanthus collinus* [13] showed suitable results where SPR bands peak were observed at 420 nm. In addition, the green synthesis studies have been also carried out using *Diospyros paniculata* [14] and *Alternanthera sessilis* Linn. [15], providing SPR band peaks at 418-430 and 435 nm, respectively.

Table 1. Absorbance and wavelength of DEA stabilized silver nanoparticles

Reaction time	Wavelength (nm)				Absorbance			
	AgNps-DEA1	AgNps-DEA2	AgNps-DEA3	AgNps0	AgNps-DEA1	AgNps-DEA2	AgNps-DEA3	AgNps0
0 hr	-	-	-	429	-	-	-	0,011
0,5 hr	409,63	413,14	429,0	434	0,648	0,317	0,213	0,017
1 hr	406,76	415,86	430,0	437	0,850	0,380	0,250	0,022
2 hr	405,00	415,98	436,0	435	1,237	0,546	0,290	0,023
4 hr	404,07	419,72	438,0	441	1,319	0,799	0,364	0,020
24 hr	399,68	419,91	422,0	434	1,286	0,942	0,798	0,036
48 hr	403,99	420,16	422,0	443	1,214	1,190	0,827	0,065
1 week	402,91	414,30	421,0	437	1,213	0,929	0,749	0,036
1 month	398,00	412,00	420,7	441	0,767	0,980	0,741	0,022

The UV-Vis spectrophotometer analysis also showed that the absorbance were gradually increased as the increase of time reaction to 24 hours (Table 1). This was strongly revealed that silver nanoparticles were continuously formed during the reaction. After 24 hours, the absorbance was decreased which allegedly because the particles began to settle at the bottom. This absorbance value was decreased as the increasing of DEA concentration. In addition, it was observed that the absorbance value of uncapped silver nanoparticles (AgNps0) was greater than DEA-capped silver nanoparticles. It revealed that DEA in aqueous solution acted as capping agent as well as reducing agent. Jia et al (16) reported a study on silver nanoparticles preparation using triethanolamine, another kind of alkanolamine compound as reducing agent with PEG and PVP as capping agents, resulting nanoparticles with mean diameter of 40 nm.

Different with absorbance, the wavelength value was increased as the increasing of the DEA concentration. Based on Mie Theory, the size of spherical nanoparticles can be estimated from the wavelength values in UV-Vis spectroscopy analysis. The larger the wavelength value, the larger the particle size [17]. These results showed that the diameter of silver nanoparticles was increased as the blueshift silver nanoparticles with bigger DEA concentration.

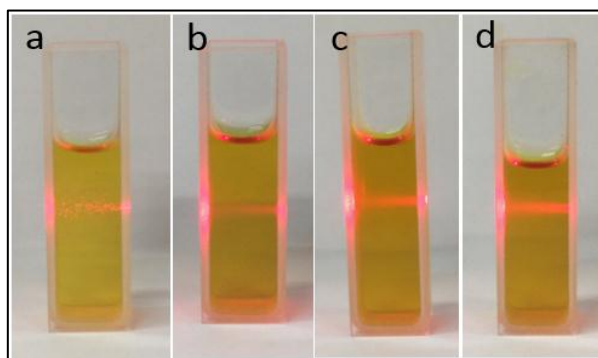


Figure 2. Laser beam radiation of silver nanoparticles in time reaction of a) 30 minute, b) 1 hour, c) 2 hours, and d) 24 hours

The laser beam radiation of colloidal silver nanoparticles was conducted to investigate the formation of silver nanoparticles colloid in the reaction based on the Tyndall Effect (figure 2). A homogenous solution is always transparent, light passes through with no scattering from solute particles which are molecule in size. While in colloid, a light scattering is occurs by the particles, resulting in a light line. Figure 2a showed a light line of silver nanoparticles in time reaction of 30 minutes which shown a light line indicating a formation of colloidal silver nanoparticles. It was observed that the intensity of the light was increased as the increase of reaction time to 24 hours (figure 2d), which confirmed an increase in the number of nanoparticles in the reaction. This sighting confirmed the result of UV-Vis spectroscopy analysis that silver nanoparticles were formed continuously during the reaction.

3.2 TEM morphology study

Figure 3 showed TEM analysis images of DEA stabilized silver nanoparticles with a molar ratio of AgNO_3/DEA was 1:10 and 1:30, and uncapped silver nanoparticles. All the particles were spherical in shape with diameter range of 4 - 52 nm for DEA capped and 9 - 39 nm for uncapped silver nanoparticles. This data support the result of UV-Vis spectrophotometer analysis and obviously showed a strong relation between wavelength and particle diameter based on TEM analysis result.

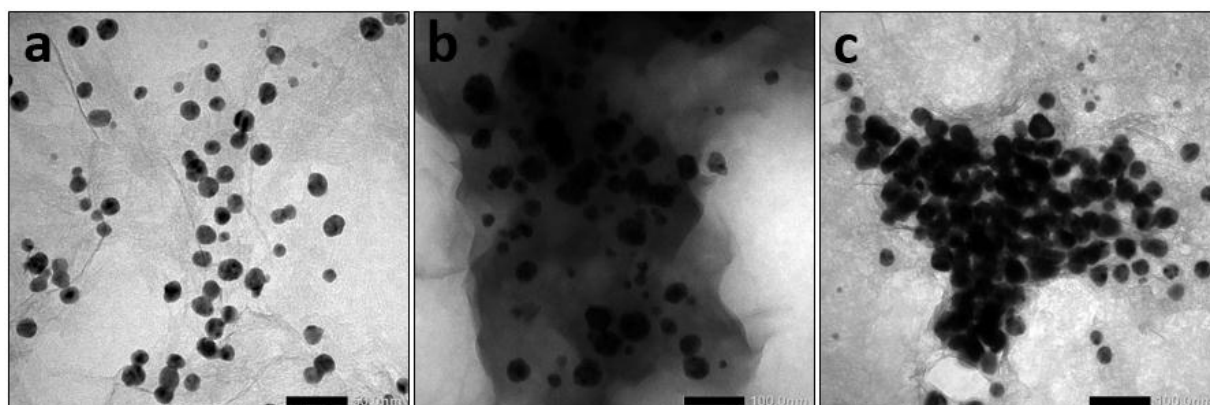


Figure 3. TEM images of a) AgNps-DEA1, b) AgNps-DEA3, and c) Uncapped AgNps

In addition, AgNps-DEA1 provided smaller mean diameter (13 nm) than AgNps-DEA3 (26 nm). The resulted silver nanoparticles were smaller than the silver nanoparticles prepared by Jia et al (16). Likewise, the size range of AgNps-DEA1 is less wide (4 - 20 nm) than AgNps-DEA3 (9 - 52 nm) (figure 4). This was due to the excessive using of capping agent. It was shown in figure 3b that there is a big clump between the particles which allegedly inhibit the interaction between silver ion with extract and decreasing the possible reduction reaction. Deficiency of nucleation reaction finally caused the growing reaction dominated and as a result, bigger particles were formed. In addition, AgNps0 exhibited bigger particle size than AgNps-DEA1, showed that the using of DEA as capping agent can reduce the particle size when used in an appropriate concentration. The most possible mechanism is that there was an interaction occurred between N and Ag surface, which then forms a protective monolayer on the surface of Ag nanoparticle. This layer can prevent an electrostatic interactions may occur among the particles, thereby control the growth and the particle size [18] These small-sized nanoparticles can provide better performance such as excellent conductivity, chemical stability, and catalytic activity, which can be developed in some areas of fields such as electronic, energy, catalysis, sensor, etc.

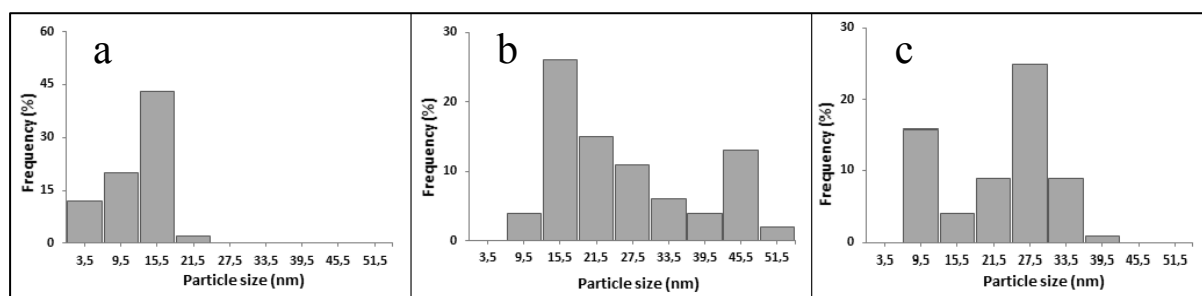


Figure 4. Size distribution of a) AgNps-DEA1, b) AgNps-DEA3, and c) Uncapped AgNps

4. Conclusions

Green synthesis of *Uncaria gambir* Roxb. leaf extract mediated silver nanoparticles had been successfully carried out using diethanolamine as capping agent. The formation of silver nanoparticles was confirmed by UV-Vis spectrophotometer analysis by a peak showed at wavelength range of 400 to 410 nm. The TEM analysis showed a formation of spherical silver nanoparticles with diameter of 3,5 - 45,5 nm. DEA capped silver nanoparticles provided smaller size than uncapped silver nanoparticles. This result showed an ability of DEA to control the growth and particle size, hence vindicate the stability of the silver nanoparticles. The increasing of DEA concentration caused the

increasing of diameter. The prepared silver nanoparticles showed a good stability in aqueous solution more than of 1 month.

Acknowledment

The authors would like to thank the Directorate General of Higher Education, Ministry of National Education of Indonesia for funding this research.

References

- [1] Iravani S 2011 *Green Chem.* **13** 2638
- [2] Rao C, Muller A and Cheetam A 2004 *Further Titles of Interest Nanoparticles Molecular Clusters of the Main Group Elements Electrochemistry of Nanomaterials* (Weinheim: Wiley-VCH Verlag GmbH & Co. KGaA p 32
- [3] Dipankar C and Murugan S 2012 *Coll. Surf. B Biointerfaces* **98** 112
- [4] Nasretdinova G, Fazleeva R, Mukhitova R, Nizameev I, Kadirov M and Ziganshina A 2015 *Electrochem. commun.* **50** 69
- [5] Kumar B, Smita K, Cumbal L, Debut A and Pathak R 2014 *Bioinorg. Chem. Appl.* **2014**
- [6] Kahrilas G, Wally L, Fredrick S, Hiskey M, Prieto A and Owens J 2014 *ACS Sustain. Chem. Eng.* 2013 2 376
- [7] Srikar S, Giri D, Pal D, Mishra P and Upadhyay SN. 2016 *Green Sustain. Chem.* **6** 34
- [8] Rauf A and Zuliayanti A 2015 *Procedia. Chem.* **14** 3
- [9] Arief S, Gustia V, Vanda D and Ban T 2015 *J. Chem. Pharm. Res.* **7** 189
- [10] Uchida K, Ogawa K and Yanase E 2016 *Molecules* **21** 1
- [11] Creighton J and Eadont D 1991 *J. Chem. Soc. Faraday Trans.* **87** 3881
- [12] Subbaiah V, Rao Y, Aparna S, Prasad T, Reddy A and Gopal D 2013 *J. Coll. Surf. B Biointer.* **105** 194
- [13] Kanipandian N, Kannan Sand Ramesh R 2014 *Mater. Res. Bull.* **49** 494
- [14] Rao N, Lakshmidhevi N, Pammi, Kollu P, Ganapaty S and Lakshmi P. 2016 *Mater. Sci. Eng. C* **62** 553
- [15] Niraimathi K, Sudha V, Lavanya R and Brindha P. 2013 *Colloids and Surfaces B* **102** 288
- [16] Jia Z, Sun H and Gu Q. 2012 *Colloids and Surfaces A* **419** 174
- [17] Baset S, Akbari H, Zeynali H and Shafie M. 2011 *Dig. J. Nanomater. Biostructures.* **6** 709
- [18] Yamamoto M, Kashiwagi Y and Nakamoto M 2006 *Langmuir* **22** 8581