

Bio-prospective of *Polyscias fruticosa* leaf extract as reductor and stabilizer of gold nanoparticles formation

Y Yulizar* and Q Ayun

Department of Chemistry, Faculty of Mathematics and Natural Sciences, University of Indonesia, Depok 16424, Indonesia Tel. +62-21-7270027, Fax +62-21-7863432

*Email : yokiy@ui.ac.id

Abstract. Metal nanoparticle is a great interest to researches due to its applications toward catalysis, sensors, and drug delivery. Biosynthesis of gold nanoparticles (AuNPs) using aqueous leaf extract of *Polyscias fruticosa* (PFE) is reported in this article. PFE plays a role as reductor and stabilizer of AuNPs. The formation of PFE-AuNPs under radiation of natrium lamp for 15 min was monitored by UV – Vis spectrophotometer. The growth process and stability of PFE-AuNPs was observed from the colour and absorbance change in the wavelength range of 529-533 nm. The optimum synthesis condition of PFE-AuNPs was obtained at 0.06% (w/v) of PFE concentration. Size and its distribution of PFE-AuNPs were identified by particle size analyzer (PSA) as 35.02 nm and stable up until 21 days. The stable PFE-AuNPs was further characterized by Fourier transform infrared (FT-IR) spectroscopy to identify the functional group in phenolic compound of PFE interact with AuNps.

1. Introduction

Currently, nanoscience and technology have many applications for human life. Their applications can be found in biomedicine [1] agriculture, food [2], environment [3], electronics [4], and energy [5]. Metal nanoparticles have been used to study, such as gold nanoparticles (AuNPs). AuNPs has attracted an attention of researchers due to has a high stability, catalytic activity and its application in many fields [6] as biomedical material [7], electronics [8], catalyst [9], optical applications [10], sensors and biomedicine [11].

Synthesis of AuNPs has been successfully carried out by the physical and chemical methods. The chemical method was obtained using reductor such as sodium borohydride [12]; N,N-dimethylformamide, trisodium citrate, hydrazine, Tollens reagents and polyethylene glycol. However, synthesis of AuNPs using chemical methods has disadvantages: toxic and hazardous material for the environment [13]. There are alternative more eco-friendly methods of nanoparticle synthesis using microorganism, enzym, and plants or plant extracts. Hence, green synthesis method using plant extract is proposed in this research. The green synthesis method is a simple, low-cost, stable, and eco-friendly method [14]. It can be applied for nanoparticles synthesis from many metals such as Pt, Au, Ag, Cu, and Zn [15-16]. Au metal is prepared by green synthesis due to possess a distinctive characteristic. Synthesis of AuNPs using the plant has been reported such as fruit extract [17], shoots [18], flowers [19] and rods [20]. Synthesis of AuNPs using leaf extracts have been conducted such as *Terminalia catappa* [21], *Euphorbia hirta* [22], and *Plumeria alba* [23].

Polyscias fruticosa (PF) plant is very abundant in Indonesia and used as herbal plant. Previous study reported the plants can be used as a reductor and stabilizer of Ag [24-25] and Au [26]. Plants are used as reductor and stabilizer in AuNP synthesis due to contain secondary metabolites such as



flavonoid. PF leaves have been studied contain flavonoid, steroid, triterpenoid, saponins, tannin, p-hydroxybenzoic acid, and vanillic acid [27]. Andarwulan N (2010) reported PF leaf contains a flavonoid (52.19 mg/100 g) higher than *Etlingera elatior* leaf (1.18 mg/100 g). PF leaf has potency as a reductor and stabilizer of AuNPs formation. Previous study has used a leaf extract through maceration in water, while in this research, maceration was conducted in methanol. Methanol extract was partitioned gradually using hexane, ethyl acetate and water. Water extract can be optimized to reduce and stabilize AuNPs. There is no report of PF leaf used to reduce and stabilize AuNPs. Therefore, the present study conduct the synthesis of AuNPs using water PF leaf extract as reductor and stabilizer of nanoparticle.

2. Experimental

2.1. Materials

Polyscias fruticosa (PF) leaves were collected from Sawangan, Depok West Java, Indonesia, and gold metal 99.9% purity obtained from PT Antam, Indonesia. Stock solution of HAuCl_4 was prepared from gold metal dissolved in aquaregia ($\text{HCl} : \text{HNO}_3 = 3:1$).

2.2. PF leaves extract preparation

PF leaves were washed and dried for one week in room temperature. Methanol of 500 mL was added to powder dried PF leaves, and macerated for 5 days in room temperature. Filtrate from maceration was then concentrated using vacuum rotatory evaporator. Methanol extract of PF leaves was gradually partitioned using hexane, ethyl acetate, and water solvents. Final fraction of water was concentrated using vacuum rotatory evaporator to get stock solution of PF extract (PFE) for AuNPs synthesis.

2.3. Preliminary phytochemical screening

Each extract was phytochemically tested for the presence of flavonoids, tannins, phenols, saponins, steroids, terpenoids and alkaloids [28]. Test of flavonoid was done using alkaline reagent. Test of phenols and tannins were done with addition of FeCl_3 solution in PFE. Test of saponins with addition of distilled water in PFE, and was shaken vigorously. Test of steroids with addition of chloroform, concentrated H_2SO_4 and acetic acid in PFE. Test of terpenoids with addition of PFE in 2 mL of chloroform, and evaporated to dryness. 2 mL of concentrated H_2SO_4 was added and heated for 2 min. Test of alkaloids with mixing PFE and 2 mL of 1% HCl , and heated gently. Mayer's and Wagner's reagents were then added to the mixture. Turbidity of the resulted precipitate was observed.

2.4. Synthesis of AuNPs using PFE and its stability

9.0 mL of HAuCl_4 0,1 mM was added 1 mL PFE with various concentration 0,02 – 0,10% (v/w). The mixture was reacted for 15 min under Na-light exposure. The AuNPs stability was observed against reaction time using UV-Vis spectrophotometer and particle size analyzer (PSA).

2.5. Characterization of AuNPs

UV-Vis spectrophotometer (Shimadzu 2600) was used to get absorption spectra of surface Plasmon resonance (SPR) for AuNPs formation. Average particles diameter, particle size distribution were measured by particle size analyzer (PSA), and its nanoparticle colloidal stability observed from the potential Zeta charge (PZC) Malvern ZEN 1600. Identification of functional groups of biocompounds in PFE leaves extract before and after reaction was done with FTIR spectroscopy (Perkin-Elmer). Transmission electron microscopy JEM 1400 - selected area electron diffraction (TEM-SAED) was used to observe the shape, size, and crystalline of AuNPs.

3. Result and discussion

3.1. Identification of aqueous PFE

Identification of aqueous PFE was done by phytochemical test. It is conducted to qualitatively analyze the presence of secunder metabolites. Phytochemical test of methanol fraction PFE positively

contained flavonoids, steroids, alkaloids, and saponnin, while of water fraction PFE positively contained flavonoids.

Identification of aqueous PFE and PFE stabilized AuNPs (PFE-AuNPs) were also measured using FTIR to investigate the interaction of functional groups in PFE and PFE-AuNPs colloids. Figure 1 shows the wavenumber shift from 3342 cm^{-1} (PFE) to 3385 cm^{-1} (PFE-AuNPs) as $-\text{OH}$ stretch that identified from flavonoid. This wavenumber is suitable with the research of Yu J et al [29]. Figure 1 shows also the wavenumber shift of 1613 to 1629 cm^{-1} as a $-\text{C}=\text{O}$ stretch. The significant wavenumber shifts of $-\text{OH}$ group (alcohol) and $-\text{C}=\text{O}$ (carboxylic acid and its derivative) from flavonoid were observed as reductor and stabilizer of AuNP formation. Based on phytochemical test, the water fraction PFE positively contained flavonoids, and this result of secondary metabolite in accordance with the FTIR data.

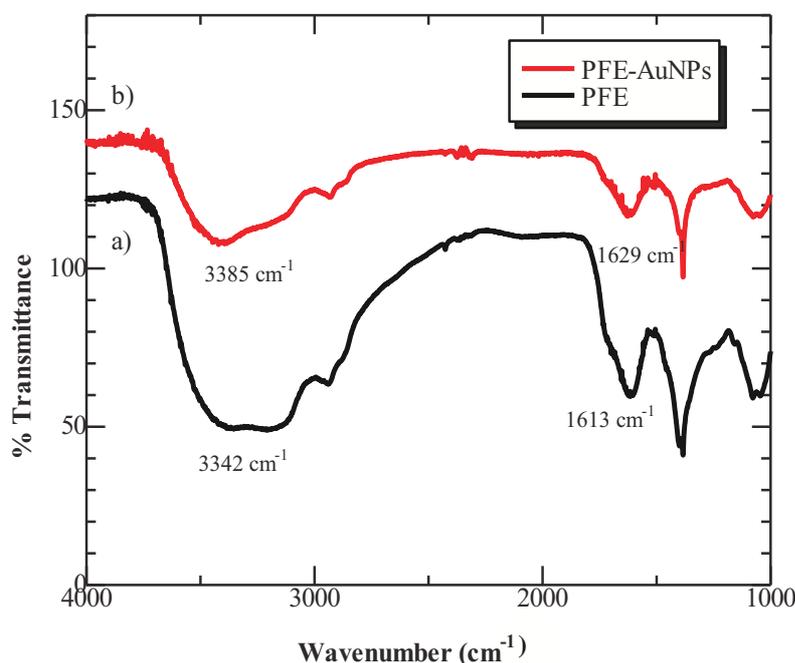


Figure 1. FTIR spectra of PFE and PFE-AuNPs.

3.2. Various PFE concentration of AuNPs synthesis

Synthesis of AuNPs was conducted by reducing the trivalent Au ions to uncharged Au atoms. In this research, synthesis of AuNPs was done with the reaction of aqueous PFE as reductor with HAuCl_4 solution under the exposure of Na-light. UV-Vis absorption spectra of PFE-AuNP were observed at various PFE concentration as shown in Figure 2 (a). AuNPs formation was confirmed by a colour change of aqueous PFE from pale yellow to bright red at $529\text{--}533\text{ nm}$ which was proved from SPR peak formation [30-31]. UV-Vis spectrophotometer was used to determine the optimum AuNPs formation through the change of wavelength and absorbance. Figure 2 (b) explains that optimum PFE concentration 0.06% was indicated by high absorbance, small maximum wavelength, λ_{max} value, and sharp peak. This is consistent with the research of Philip D (2010) which the optimum concentration for AuNPs formation was indicated by small λ_{max} and high absorbance [32]. The further experiment to confirm UV-Vis absorption spectra was conducted using TEM and PSA.

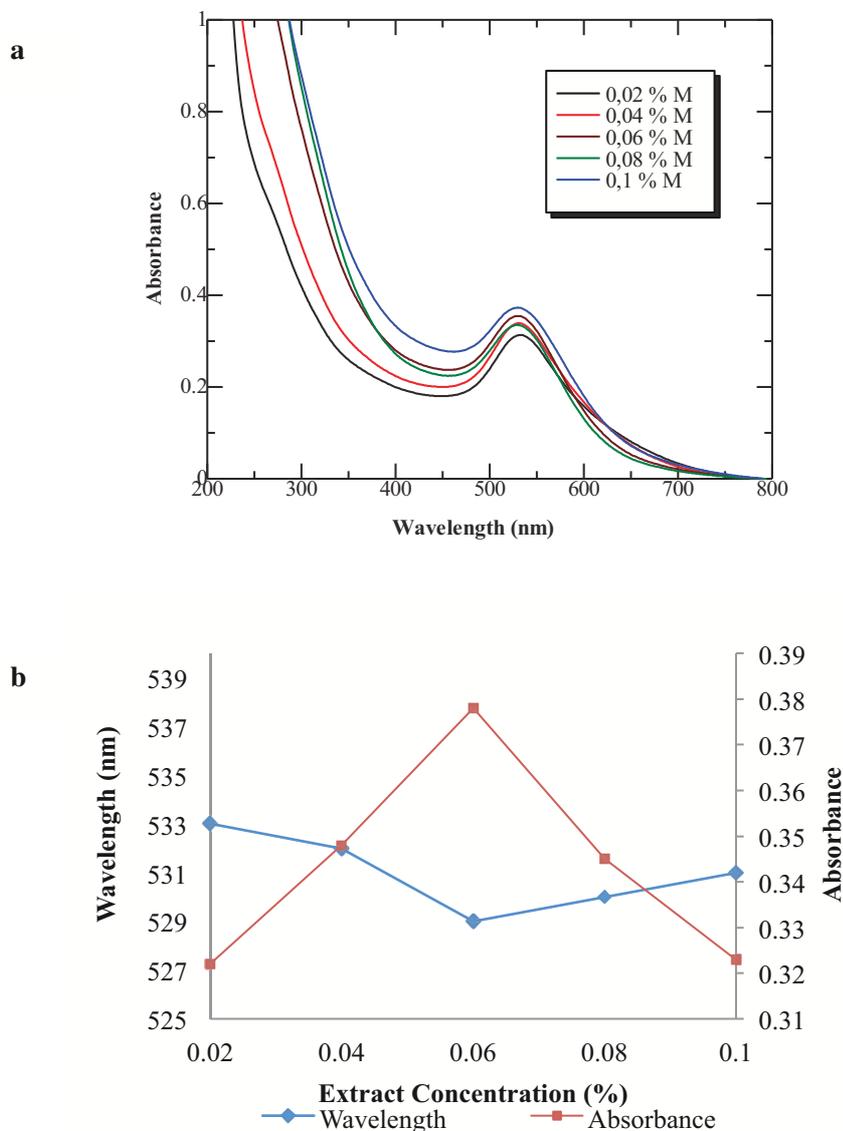


Figure 2. (a) UV-Vis absorption spectra of PFE-AuNPs and (b) relational curve between λ_{max} and absorbance against PFE concentration of PFE-AuNPs.

3.3. Stability of AuNPs

Synthesis of AuNPs will agglomerate due to existing of interparticle force which cause a strong interaction. Figure 3 shows the relation between absorbance value and λ_{max} against reaction time of PFE-AuNPs formation for 30 days. λ_{max} was observed to increase from 21 until 25 days, and shows that AuNPs was stable for 21 days. AuNP stability is affected by the active compounds in PFE which act as a stabilizer of AuNP. However, there is a trend of decrease after 21 days, which indicates AuNPs agglomeration.

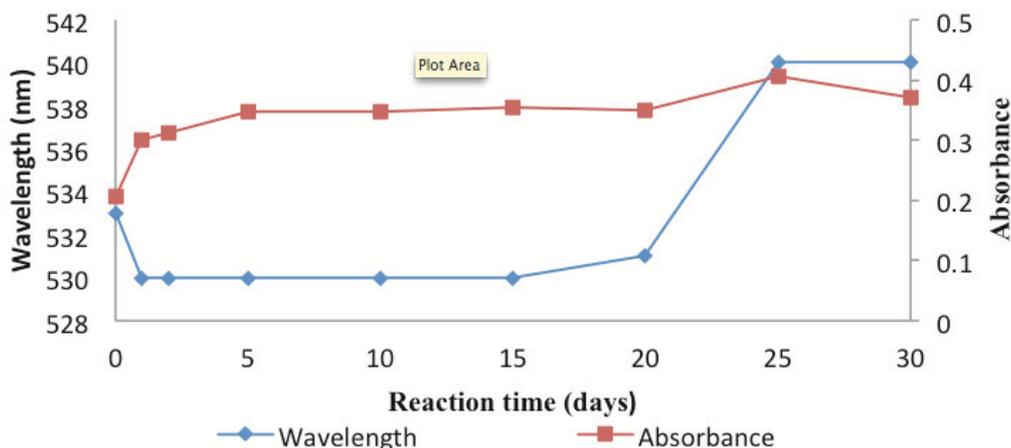


Figure 3. Curve of λ_{\max} and absorbance value against stability time of PFE-AuNPs.

Figure 4 shows potential zeta charge (PZC) of PFE-AuNPs. PZC measurement was conducted to observe the stability of PFE-AuNPs and to measure the total charge between PFE interacted on AuNPs surface. PZC value of PFE-AuNPs is -12.3 mV, which means the particles are stable due to the negative charge prevent aggregation between particles as an effect of repelling force between the charges. In this study, PFE-AuNPs was observed stable for 21 days, after that it starts to agglomerate each nanoparticle.

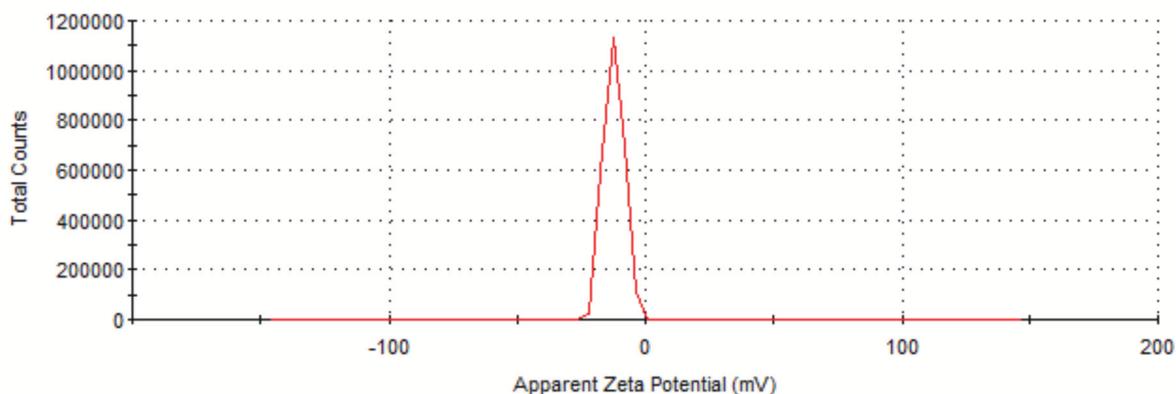


Figure 4. Zeta potential charge of optimum PFE-AuNPs.

3.4. Particle size of AuNPs

Particle size of PFE-AuNPs was measured using PSA and TEM. Figure 5 explains PSA spectrum for intensity (percent) against particle size (nm) of PFE-AuNPs at optimum PFE concentration (0,06%). Intensity at Figure 5 indicates the amount of formed nanoparticles that scatter the light. Particle size

and its distribution of AuNPs were polidispersed with size majority at 35.02 nm.

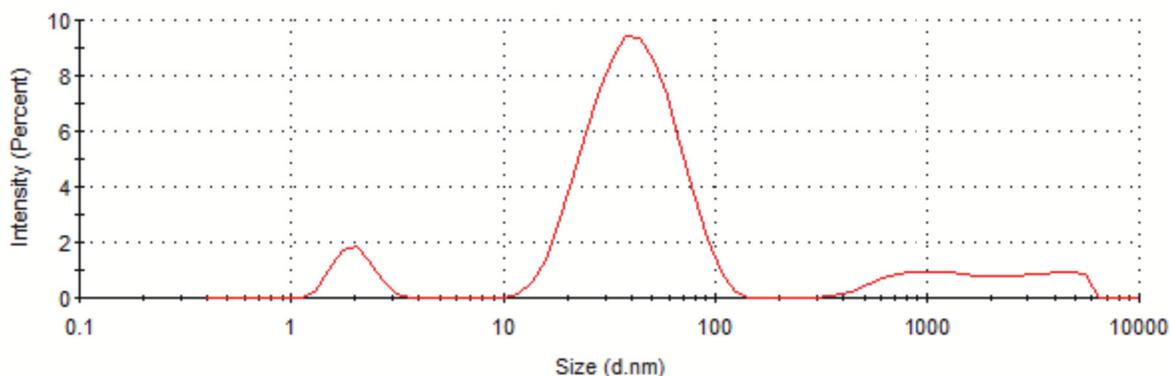


Figure 5. Particle size and distribution of PFE-AuNPs.

TEM characterization was conducted to identify morphology, shape, and size of nanoparticles. TEM measurement of PFE-AuNPs was conducted at optimum PFE concentration (0.06%). TEM image showed a homogenously dispersed PFE-AuNPs and has an average diameter of 20 nm with relatively spherical shapes as shown in Figure 6. The homogenously dispersed and controlled nanoparticle size due to the aqueous PFE played a role as a good stabilizer through capping on AuNPs surface.

The resulted data shows that AuNPs size from PSA was greater than TEM. The nanoparticle size using PSA measured PFE capped AuNPs through its scattering total, while TEM data only show image of AuNPs due to it has a high density.

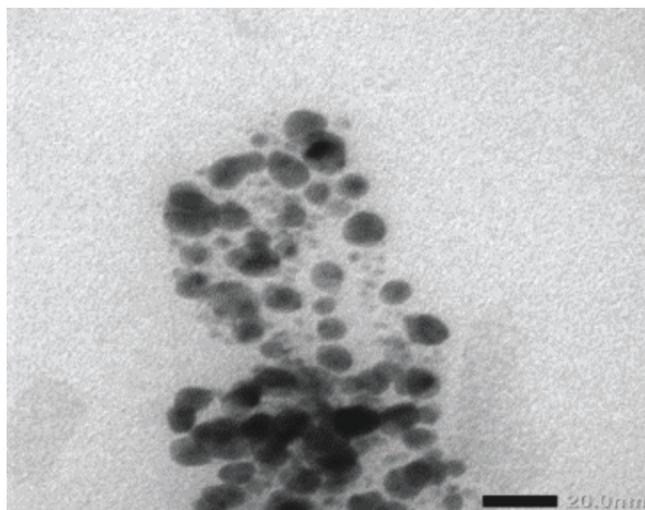


Figure 6. TEM image of AuNPs.

3.5. Analysis of AuNPs using TEM-SAED

TEM image showed spherical shapes with size of 20 nm. SAED pattern in Figure 7 shows that AuNPs formed a crsytalline structure of face center cubic (fcc) with Miller index (111), (200), (220), and

(311). SAED analysis of AuNPs indicates the existence of some ring that adjusted with JCPDS of Au on (111), (200), (220), and (311).

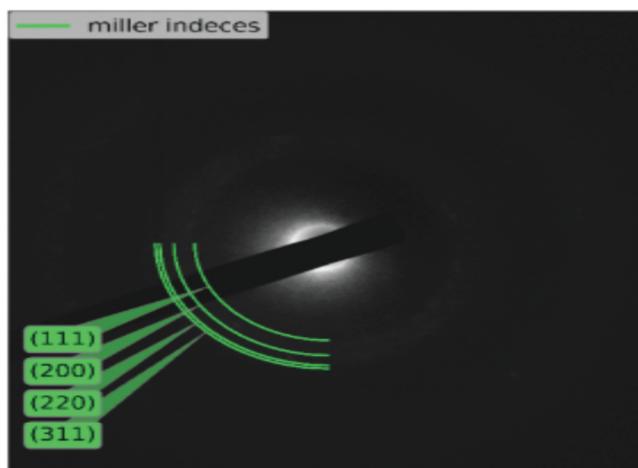


Figure 7. SAED pattern of AuNPs.

4. Conclusion

AuNPs were successfully synthesized using *Polycias fruticosa* leaf extract (PFE) in water fraction by green synthesis method. PFE played a role as reductor and stabilizer of PFE-AuNPs. Synthesis of AuNPs was done at optimum PFE concentration of 0.06%. Various PFE concentration has affected the maximum wavelength (λ_{\max}), absorbance value, and spectrum peak shape of AuNPs. AuNPs were stable for 21 days, and has optimum λ_{\max} 531 nm. Particle size of PFE-AuNPs based on PSA measurement is 35.02 nm, and the PZC value is -12.3 mV. Based on FTIR characterization, PFE contained a flavonoid that acted as reductor and stabilizer of AuNPs formation. The result of TEM image showed that AuNP has an average diameter of 20 nm with spherical shape. Analysis of SAED pattern of AuNPs indicated the presence of ring that adjusted with JCPDS of Au on (111), (200), (220), and (311). This explains that the crystalline characteristic of AuNPs is fcc (*face center cubic*).

Acknowledgment

This work was funded by hibah PITTA 2016, from the Universitas Indonesia through the directorate of research and community services, Universitas Indonesia (No.2040/UN2.R12/HKP.05.00/2016).

References

- [1] Bansal A and Zhang Y 2014 *Acc. Chem. Res.* **47** (10) 3052-60.
- [2] Sekhon B S 2014 *Nanotechnol.* **7** 31-53.
- [3] Tang S C N and Lo I R M 2013 *Water Res.* **47** (8) 2613-32.
- [4] Alshehri A H, Jakubowska, Lozniak M M, Horaczek A, Rudka M, Free D, and Carey J D 2012 *ACS Appl Mater Interfaces.* **4** (12) 7007-10.
- [5] Ahn S J, Kim K, Cho A, Gwak J, Yun J H, Shin K, Ahn S K, and Yoon K 2012 *ACS Appl Mater Interfaces.* **4** (3) 1530-6.
- [6] Sadeghi B, Rostami A, Momeni S S 2014. *Facile green synthesis of silver nanoparticles using seed aqueous extract of Pistacia atlantica and its antibacterial activity*. Department of Chemistry, Islamic Azad University, Iran.
- [7] Cao X L, Cheng C, Ma Y L, Zhao C S 2010 *J Mater Sci Mater Med.* **21** 2861-8.
- [8] Mohan Y M, Lee K J, Premkumar T, Geckeler K E 2007 *Polymer.* **48** 158-64.

- [9] Feng X, Qi X, Li J, Yang L W, Qiu M C, Yin J J, Lu F, Zhong J X 2011 *Appl Surf Sci.* **257** 5571- 5.
- [10] Hayward R C, Saville D A, Aksay I A 2000 *Nature.* **404** 56–9.
- [11] Sugihara K K, Sujitha M V, and Kannan S 2013 *Spec Act Part A.* **102** 15–23.
- [12] Yulizar Y, Kadja G T M and Safaat M 2016 *Reac Kinet Mech Cat.* **117** 353-63.
- [13] Joseph S, Mathew B 2014. *Microwave assisted facile green synthesis of silver and gold nanocatalysts using the leaf extract of Aerva lanata.* Department of Chemistry, Kerala, India.
- [14] Ahmed S, Ahmad M, Swami B L and Ikram S 2016 *J Adv Res.* **7** 17-28.
- [15] Mohanpuria P, Rana N K and Yadav S K 2008 *Nanopart Res.* **10** 507–17.
- [16] Raveendran P, Fu J and Wallen S L 2003 *J Am Chem.* **125** 13940-1.
- [17] Elumalai K, Velmurugan S, Ravi S, Kathiravan V and Adaikala Raj G 2015 *J Adv Pow Tec.* **26** 1639-51.
- [18] Armendariz V, Herrera I, Peralta V J R, Jose Y M, Troiani H, Santiago P, and Gardea T J L 2004 *J Nanopart Res.* **6(4)** 377–82.
- [19] Anand K, Gengan R M, Phulukdaree A, Chuturgoon A 2014 *J Ind Eng Chem.* **21** 1105-11.
- [20] Suman T Y, Rajasree R S R, Ramkumar R, Rajthilak C and Perumal P 2013 *Spec Act Part A.* **118C** 11–6.
- [21] Ankamwar B 2010 *E-J Chem.* **7** 1334-9.
- [22] Annamalai A, Christina V L P, Sudha D, Kalpana M, and Lakshmi P T V 2015 *J Colloid interf sci.* **108** 60– 5.
- [23] Mata R, Bhaskaran A and Sadras S R 2016 *Particuology.* **24** 78–86.
- [24] Zuas O, Hamim N and Sampora Y 2014 *Mater Lett.* **123** 156-9.
- [25] Kasthuri J, Veerapandian S and Rajendiran N 2009 *Colloid Surface. B* **68** 55-60.
- [26] Kumar N R B, Kumar K H A and Mandal B K 2015 *J Mol Liq.* **211** 868-75.
- [27] Andarwulan N, Batari R, Agustini D S, Bolling B and Wijaya H 2010 *Food Chem.* **121** 1231-35.
- [28] Yadav R N S and Agarwala M 2011 *Phytology.* **3** (12) 10-4.
- [29] Yu J, Xu D, Guan H D and Wang C 2016 *Mater Lett.* **166** 110-2.
- [30] Yallapa S, Manjanna J, Dhananjaya B L 2015 *Spechtrochim Acta Part A.* **137** 236-43.
- [31] Devi P R, Kumar C S, Selvamani P, Subramanian N, and Ruckmani K 2015 *Mater Lett.* **139** 241-4.
- [32] Philip D 2010 *Spechtrochim Acta Part A.* **75** 1078-81.