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Morphological Characteristics Of Biosynthesized Silver Nanoparticles Derived From Ficussepticaleaf Ethanolic Extract

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Abstract

The purpose of this study was to determine the morphological characteristics of biosynthesized Silver Nanoparticles (AgNPs)derived fromFicussepticaleaf ethanolic extract. The extracts were subjected to UV-Vis spectrophotometer, Scanning Electron Microscopy and FTIR analysis for characterization and confirmation of the biosynthesized AgNPs.

The ethanolic extract of Ficusseptica was found to be effective in the biosynthesis of AgNPs in this research. Scanning Electron Microscopy was used to investigate the morphological dimensions of produced AgNPs from Ficusseptica ethanolic extract.The SEM revealed that the particles demonstrated an average size ranging from 77 nm to 103 nm, whereas shapes are spherical.

Keywords: Silver Nanoparticles. Ficusseptica, FTIR, UV-vis, Nanotechnology

Introduction

Nanoparticles technology is progressing rapidly and is being used in a wide variety of medical applications. The microbicidal nature of the chemically and biosynthesized nanoparticles become known as a novel alternative to deal with bacterial multidrug-resistance that is now universally encountered due to abuse of antibiotics.

According to Elliot et al. (2010) there has been no reported resistance of microorganisms to silver ions or silver-based compounds. Biosynthesis of silver nanoparticles from plant extracts is friendlier and less biohazardous to use because it does not need high temperature and toxic fume in generating large quantity of silver nanoparticles. The reducing agent and stabilizing agent used in the synthesis of silver nanoparticle can be derived from living organisms such as plants. Plant crude extract from leaves, stems, roots, and others contains novel secondary metabolites such as phenolic acid, flavonoids, alkaloids and terpenoids in which these compounds are primarily responsible for reducing the formation of ionic substances in metallic bulk nanoparticles.

FicussepticaBurm.f. commonly known as hauili or liwliw, is a shrub or tree of the family Moraceae, living at low altitudes from Northeast India to North Australia, and throughout Malaysian region including Philippines. As stated by Tsai in his study on 2006, the leaves were used as cure for skin disease, appendicitis, abscesses, poisonous snakebites and shortness of breath. As stated by Stuart in 2015, a Philippine tribe, the Ifugaos, utilized this plant for diarrhea, cough, malaria and stomach problems.

Some studies reported the phytochemical constituents of Ficussepticaand it revealed various isolated alkaloids. In 2005, Berg et al.include (-)-tilosrebin (hauptalkaloid), tiloforin, septisin, and antofin, and it also contains flavonoids A study of Kuoet al. in 2002 has reported the isolation of seven triterpene derivative, 13,27-cycloursan-3β-yl acetate, and two lignans from the non-alkaloidal fractions of the stem of Ficusseptica.

Ragasaetal. in 2016 also investigated the chemical constituents found in the dichloromethane extracts of Ficusseptica. The study led to the isolation of β-sitosteryl-3β-glucopyranoside-6'-O-fatty acid esters (1), α-amyrin fatty acid esters (2), and a mixture of β-sitosterol (3a) and stigmasterol (3b) in a 5:2 ratios from the twigs; and 3a, β-amyrin (4), and long chain saturated fatty alcohols (5) from the leaves.Secondary metabolites may form a protective layer over metal nanoparticles, preventing buildup and thereby stabilizing the medium.

Materials and Methods

Plant collection and Extract Preparation

Fresh Ficussepticaleaves were collected in Rizal, Cagayan. Leaves were thoroughly washed with distilled water for three times. The leaves were air dried under shade and was pulverized using a homogenizer. 150g of powdered Ficusseptica leaves was mixed in 1,350ml of ethanol in erlenmeyer flask, covered with aluminum foil and stored for five days.

The mixture was filtered using analytical filter paper and the resulting filtrate placed in rotary evaporator. After the procedure, the extract was collected and stored in the refrigerator.

Biosynthesis of silver nanoparticles

Ten ml (10ml) of the refrigerated filtrate was treated with 90 ml of 10^{-3}M $AgNO_3$ solution. The development of yellowish to brown color solution indicated the formation of AgNPs. The addition of $AgNO_3$ solution to the ethanolic extract lead to the formation of silver nanoparticle due to reduction of Ag^+ to Ag^0. The colored AgNPs solution was centrifuged at 6,000rpm for 15 minutes. The supernatant liquid was collected and was used for characterization of AgNPs.The method was adapted from Mohanta et al. (2017).

Characterization of silver nanoparticles

The color change in the reaction mixture was noted through visual observation. The reduction of Ag^+ ions was monitored by measuring the UV-Vis spectrum of the reaction medium. UV-Vis spectral analysis was done using UV-Vis spectrophotometer Lambda 25 (PerkinElmer) at the wavelength of 200-800nm. Fourier-Transform Infrared Spectroscopy was performed to obtain an infrared spectrum of absorption of the samples using Shimadzu IR Prestige 21. Scanning Electron Microscope (SEM) was used to view the morphology and measure size of biosynthesized silver nanoparticles in JSM-5310 instrument operated at an accelerating voltage of 10Kv-15kV.

Results and Discussions

Characterization of the biosynthesized silver nanoparticles derived from Ficusseptica

The biosynthesis of AgNPswas established through visual indications brought by color change of the reaction substrate as shown in Figure 1. Jena et.al (2013) mentioned that color transition to brown imply the biotransformation of Ag^+ ion to Ag^0 indicating synthesis of silver nanoparticles.The change in the color of the solution is also due to the surface plasmon resonance exhibited by the biosynthesized silver nanoparticles (Erdoğan et al., 2016).

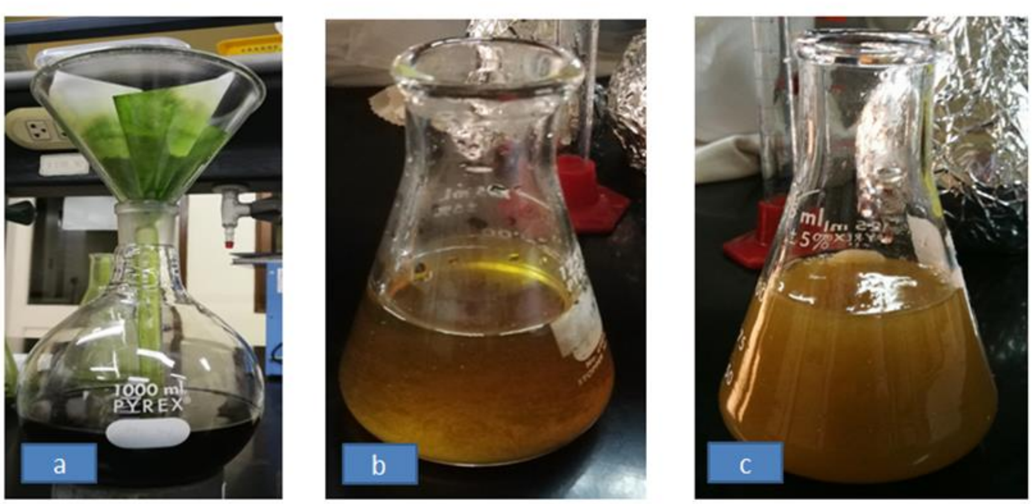


Figure 1: Biosynthesis of Silver Nanoparticles. a) Ethanolic leaf extract of Ficus septica b)Synthesized silver nanoparticles 30 minutes after the addition of Silver Nitrate c) Synthesized silver nanoparticles 3 hours after the addition of Silver Nitrate.

Using UV-Vis Spectroscopy, Figure 2 illustrated the formation of AgNPs at 389.55 nm surface plasmon peak (SPR) at the addition of $AgNO_3$ to Ficusseptica leaf ethanolic extract. Accordingly, there is no absorption band observed with Ficussepticaethanolic extract solution.

In a similar study by Kanjinkar&Londonkar (2017) on the characterization of Ficuskrishnae, they recorded an absorbance peak at 425 nm. This and from other literatures related to characterizing AgNPs reflected a slightly higher absorbance peak than what is revealed in the present research. However though, Govindaraju et al. and Jena et al. argued that for AgNPsbiofabrication, a broad SPR peak width of 320–580 nm as characteristic wavelength (λ_{max}) serves as an agreeable detector of the nanometal size and its polydispersity.

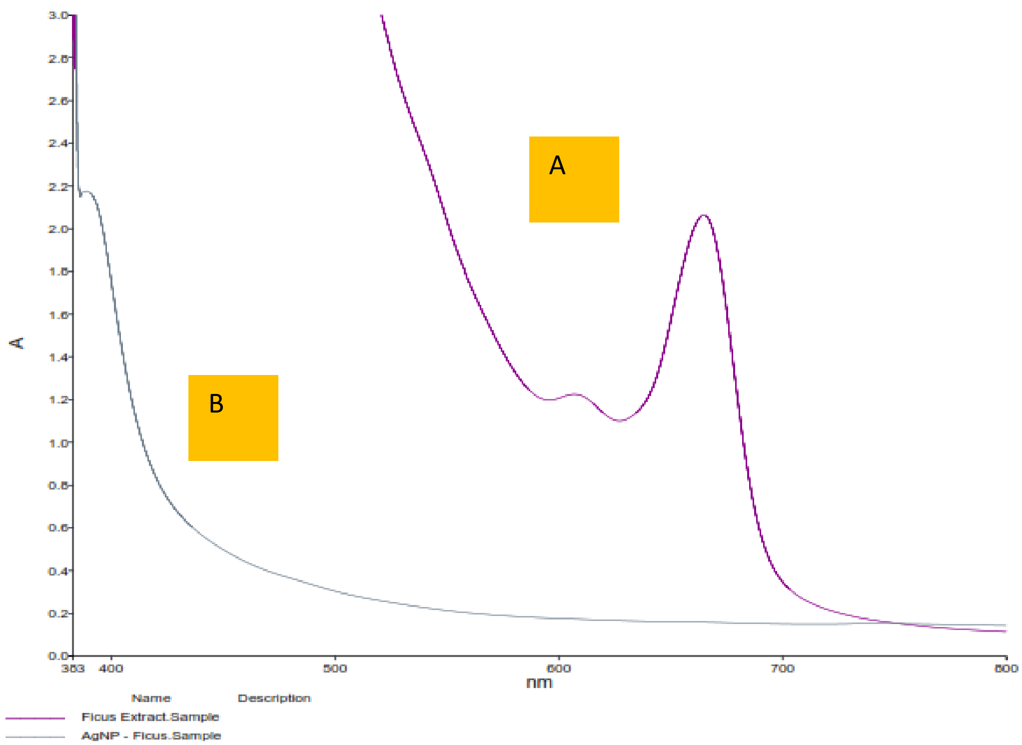
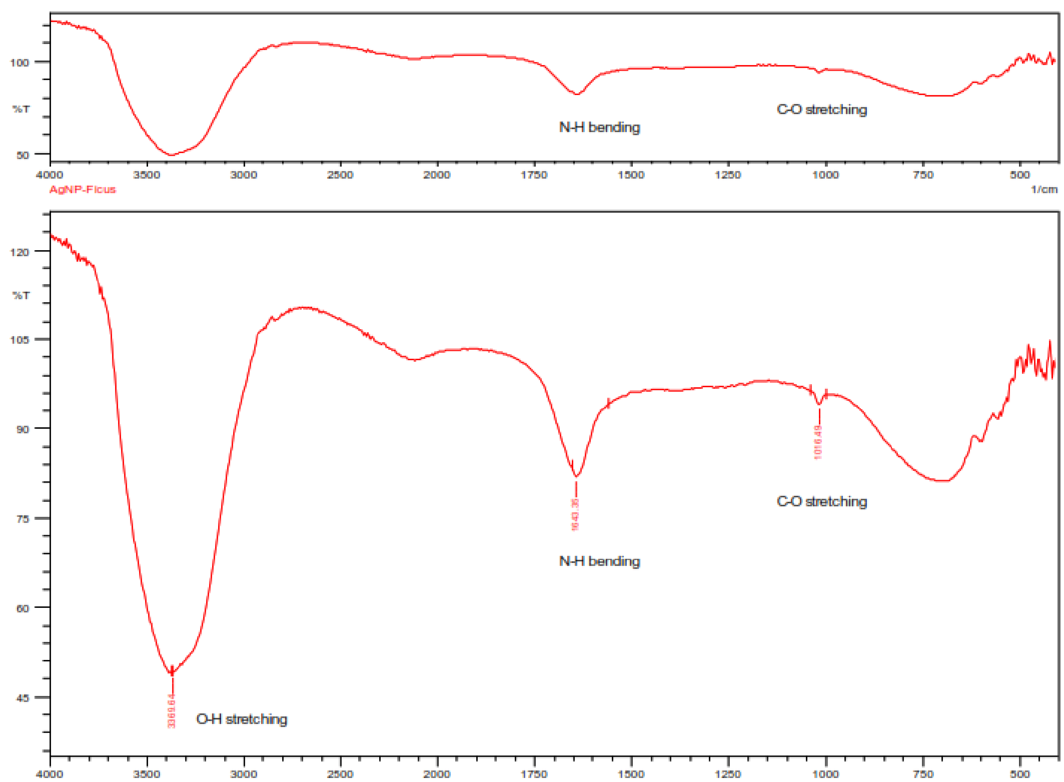


Figure 2. UV-visible absorption spectrum of (A) Ficussepticaethanolic extract and (B) biosynthesized silver nanoparticles ofFicusseptica

To reveal the possible potential biomolecules that participated in the bioreduction of silver and stabilization of AgNPs, Fourier Transform Infrared Spectroscopy (FT-IR) analysis was conducted. The FT-IR profile shown in Figure 3 illustrated 3 peak positions at 1016.49, 1643.35, and 3369.64 cm^{-1} for the AgNPs reaction mixture. Nandiyanto et.al (2019) classifies this sample as a simple spectrum because it has less than 5 absorption bands. Hence, the compound analyzed has small molecular weight.

Moreover, a broad absorption band in the range of between 3650 and 3250 cm-1, indicates a hydrogen bond. Specifically, the FT-IR analysis indicated an O-H stretching followed by spectra at frequencies at 1600 cm^{-1} and below. This suggests that the compound contains a hydroxyl group. Figure 3 also shows distinctive broad spectral bands at around 1643.35 cm^{-1}, a characteristic to the N-H bend vibration. This proves the presence of a primary amine in the compound (Coates, 2000). Lastly, the third absorbance peak evidently suggests of a hydroxyl compound. Coates (2000) listed the C-O stretch at ~1050 cm^{-1} as primary alcohol. Overall, the FT-IR suggests that the hydroxyl group containing alcohols from the Ficusseptica extract could have possibly formed a layer covering the metal nanoparticles to prevent accumulation and thereby stabilizing the medium (Kyomuhimbo, 2019).



No.	Peak	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area
1	1016.49	94.129	1.984	1039.63	997.20	0.884	0.159
2	1643.35	82.054	2.940	1654.92	1560.41	5.311	0.439
3	3369.64	49.063	0.213	3375.43	3365.78	2.974	0.010

No.	Peak	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area
1	1016.49	77.593	18.011	1062.78	958.62	4.931	2.936
2	1651.07	88.507	0.254	1799.59	1649.14	1.858	-1.239
3	2837.29	95.813	5.013	2858.51	2806.43	0.337	0.545
4	3373.50	58.499	0.132	3375.43	3369.64	1.345	0.003

Figure 3. FTIR spectrum of biosynthesized silver nanoparticle of Ficusseptica and Ficusseptica ethanolic extract (second table)

Table 1 summarizes the results of the characterization in terms of shape and size. The tests reveal that the synthesized AgNPs are spherical in shape and demonstrated an average size ranging from 77nm to 103nm. The morphology dimensions are shown in Figure 4 of the synthesized AgNPs from Ficusseptica ethanolic extract.

Table 1.Results of the Scanning Electron Microscopy (SEM) in characterizing the silver nanoparticles (AgNPs) derived from F.septica ethanolic leaf extract.

Characteristics	SEM Results
Shape	Spherical
Size	77 nm to 103 nm

There are no published studies on characterization of Ficusseptica to compare the results of this study. However, of similar genus, Kanjikar et.al (2017) showed in their study on Ficuskrishnae that its particle size was ranging from 15 to 28 nm while the shape is also spherical. Bocarando-Chacon et al. (2014) recorded a diameter to be about 15 nm for Ficusindica.

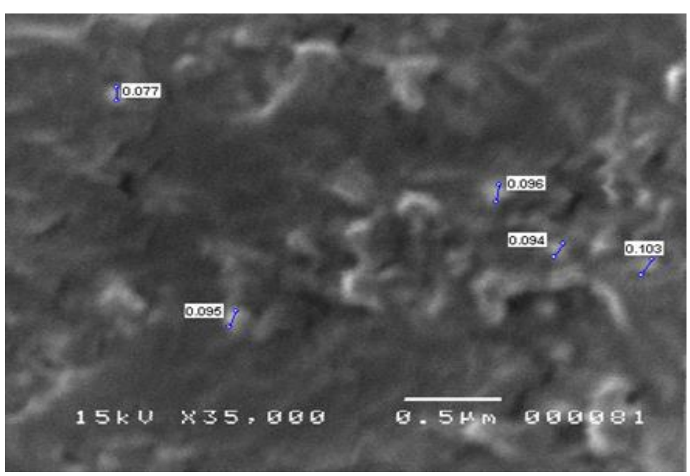


Figure 5.SEM image of biosynthesized AgNPs

Conclusion

Results of this study revealedthat the ethanolic extract of Ficusseptica can be used efficiently for the biosynthesis of AgNPs. The SEM revealed that the particles demonstrated an average size ranging from 77 nm to 103 nm, whereas shapes are spherical.

Acknowledgments

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