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Biosynthesis and Characterization of Silver Nanoparticles Using Brimstone Leaf (*Morinda Lucida*)

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Abstract
Numerous branches of science have undergone a significant revolution. The creation and use of nanoparticles, which have numerous applications, is a function of nanoscience and nanotechnology. In addition to other nanoparticles, silver nanoparticles (AgNPs) have drawn a lot of attention because of their distinct characteristics. This research aimed to utilise a simple, economical, quick, and green strategy for the production of Morinda lucida silver nanoparticles (ML-AgNPs). The synthesis of ML-AgNP was assessed using colour change, UV spectrophotometry, and electron microscopy (scanning). The active (functional) components in the leaf extracts of the plant were evaluated using Fourier transform infrared (FT-IR) spectroscopy. According to FT-IR, the extract contained many active biochemical components that were accountable for the reduction of silver salt to nanoparticles, and SEM analysis also revealed that the produced nanoparticles shape is spherical and particle size was less than 50 nm, which was also confirmed by energy dispersive X-ray (EDX) spectroscopy to be highly made up of Ag, with excellent visual peaks around 3 keV. The findings demonstrate that these ML-AgNPs can be used effectively in a variety of application, including therapeutics and farming.

Keywords: Biosynthesis; *Morinda lucida*; Silver nanoparticles; Phytochemicals screening.

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Introduction

Nanoscience is regarded as a rapidly expanding field in contemporary science and mechanisation, with scientists around the world attempting to generate extremely efficient materials known as nanoparticles in cost-effective ways (Saini et al., 2020). Nanoparticles are either synthetic or natural small particles with diameters varying from 1 to 100 nm (Vanlalveni *et al*., 2021; Chinni *et al*., 2021). These particles have perfectly unique and increased properties such as size, distribution, and shape when compared to the bigger particles of the mass material from which they were formed. Among other nanoscale materials, silver nanoparticles (AgNPs) have drawn the attention of innumerable scholars because of their distinctive peculiarities (Chinni *et al*., 2021). AgNPs are important engineering materials that have been widely explored due to their electrical, biological optical, and thermal properties (Sooraj *et al*., 2021). This has led to the use of these nanomaterials in a variety of fields, such as nanomedicine, therapeutics, the development of nanodevices, and biomedical applications (Mehmood *et al*., 2021). Synthesis of AgNPs can be achieved through top-down and bottom-up techniques. The top-down approach entails using methods like ball milling, laser ablation, and sol-gel to fragment a bulk material into nano-sized pieces (Chinni *et al*., 2021). On the other hand, the bottom-up approach refers to the production of nanoparticles utilising tiny elements, such as chemical, physical, and biological techniques (Gebreslassie & Gebretnsae 2021). The increasing utility of chemically modified nanoparticles in industrial activities necessitates the development of a method that will not endanger the ecosystem or living organisms as a consequence of nanoparticle exposure, hence the biological approach. (Bawazeer *et al*., 2021; Periasamy *et al*., 2021). Biological procedures, known as green synthesis, are preferable to chemical and physical approaches because they are less expensive, more ecosystem friendly, and more widely available (Philip *et al*., 2018; Salmen & Alharbi, 2020). The utilisation of AgNps in recent technologies is based on their distinctive traits such as size, surface Plasmon features, and shapes, among others (Bawazeer *et al*., 2021). It has been reported that the use of leaf extracts from medicinal plants, such as Acalypha indica (Padole & Avari, 2021), Garcinia mangostana (Riaz et al., 2021) and Aloe vera (Salem and Fouda 2020) in the production of silver nanoparticles is effective as a result of the presence of hydroxyl groups (OH) and many other groups in plant phytochemicals, which have little or no toxic effect on the synthesised nanoparticles (Padole & Avari, 2021). The purpose of this investigation is to purify and characterise the biosynthesized silver nanoparticles from the Morinda lucida aqueous extract.

Experimental

Materials

Distilled water (DH_2O) was acquired from the Food Technology Laboratory of Federal Polytechnic Ilaro, Ogun State, Nigeria, while silver nitrate ($AgNO_3$) salt, purity 99.9%, was acquired from Sigma Aldrich, St. Louis, USA.

Collection of samples and extraction of the bioactive component from the Brimstone plant (morinda lucida)

The brimstone (Mornda lucida) plant was harvested with a knife from the Federal Polytechnic Ilaro forest reserve in Ogun State, Nigeria, and delivered to the Science Laboratory department's laboratory. The plant leaves were scrubbed with tap water to eliminate dust particles that had adhered to the leaves of the plant before being rinsed with distilled water. The cleaned brimestone plant leaf was air dried at room temperature for fifteen (15) days before being blended into powdery form and preserved at room temperature for further use.

Synthesis of ML-AgNPs

A 1 mM aqueous solution of AgNO3 was prepared at room temperature using a pure AgNO3 salt. The Morinda lucida leaf extracts (5 ml) were then introduced to a 250 mL Erlenmeyer conical flask containing 95 mL of aqueous solution of AgNO3 on a magnetic stirrer at 50 °C for 30 minutes, until complete synthesis of AgNPs was indicated by colour change.

Characterization of ML-AgNPs

UV-Vis spectrophotometer

At room temperature, a JASCO 670-UV-Vis spectrophotometer was used to perform spectral analysis for the development of nanoparticles from 300 to 700 nm with a resolution of 1 nm. AgNPs produced a distinct peak in the visible region of the electromagnetic spectrum.

SEM-EDS Analysis of ML-AgNPs

The morphology of the produced nanoparticles was determined using a Scanning Electron Microscope (SEM). A drop of metal nanoparticle solution was placed on an aluminium grid sample holder and dried at room temperature. The sample's elemental composition was examined using energy dispersive X-ray spectroscopy (EDS) in conjunction with a SEM.

FTIR Analysis of ML-AgNPs.

The sample's Fourier transformed infrared (FTIR) spectrum was measured using a Fourier transform infrared spectrophotometer. By adding a KBr pellet with AgNPs, the FTIR spectrum ranged from 4000 to 450 cm-1 with a resolution of 4 cm1.

Results and Discussion

Phytochemical Analysis of Morinda lucida

Table 1 gives the summary of the bioactive components of the plant extract of Morinda lucida. According to the analytical approach, the content of biochemicals in the extract is qualitatively described by considering the colour of the mixture. In the examined morinda lucida, phenols, saponins, alkaloids, tannins, and flavonoids were confirmed while steroids, anthocyanides, and anthraquinones were absent. These bioactive components present have a sizeable position in the stabilisation and discount of Ag ions in the course of the bioformulation of Ag nanoparticles and may also have widespread medical application. Furthermore, phenols and flavonoids were responsible for the stabilisation and capping of Ag nanoparticles (Brungesh *et al*., 2017). Similarly, active biochemicals such as tannins, phenols, and alkaloids have polar groups like OH-hydroxyl group and CO-carbonyl group that may attract Ag ions. The two groups would possibly be essential in governing the nano-size of metallic Ag (Fatimah & Herianto 2018).

Table 1: Summary of the bioactive components of the plant extract of Morinda lucida

Phytochemicals	Inference
Flavonoids.	+
Saponins.	+
Anthocyanosides.	-
Alkaloids.	+
Phenols.	+
Anthraquinones.	-
Steroids.	+

Notation: + indicate presence of biochemicals while - indicate absence of biochemicals

Uv- vis spectrophotometry analysis

An essential method for confirming the emergence of nanoparticles in solutions is UV-VIS analysis (Chinni et al., 2021; Periasamy *et al*., 2022). While introducing the aqueous extract of morinda lucida leaves into the Ag ion solution. The colour of the mixture transformed from colourless to golden brown. The colour transformation made it visually obvious that silver ions were being bioreduced to silver nanoparticles. Changes in colour of the mixture may be attached to bioactive components of the plant extract (Bawazeer *et al*., 2021; Periasamy *et al*., 2021) and surface plasmon vibrations in ML-AgNPs (Sooraj, Nair and Vineetha 2021; Afreen *et al*., 2020). As seen in Figure 2, the UV-Vis spectrum of Morinda lucida leaf extract demonstrated the generation of an absorption band at 380 nm. As stated in a previous study, the broad bands seen at 360 and 400 nm in the ML-AgNPs UV spectrum may be associated with the bioactive components of the Morinda lucida leaf extract as well as surface plasmon resonance (SPR) of green synthesis ML-AgNPs (Padole & Avari, 2021).

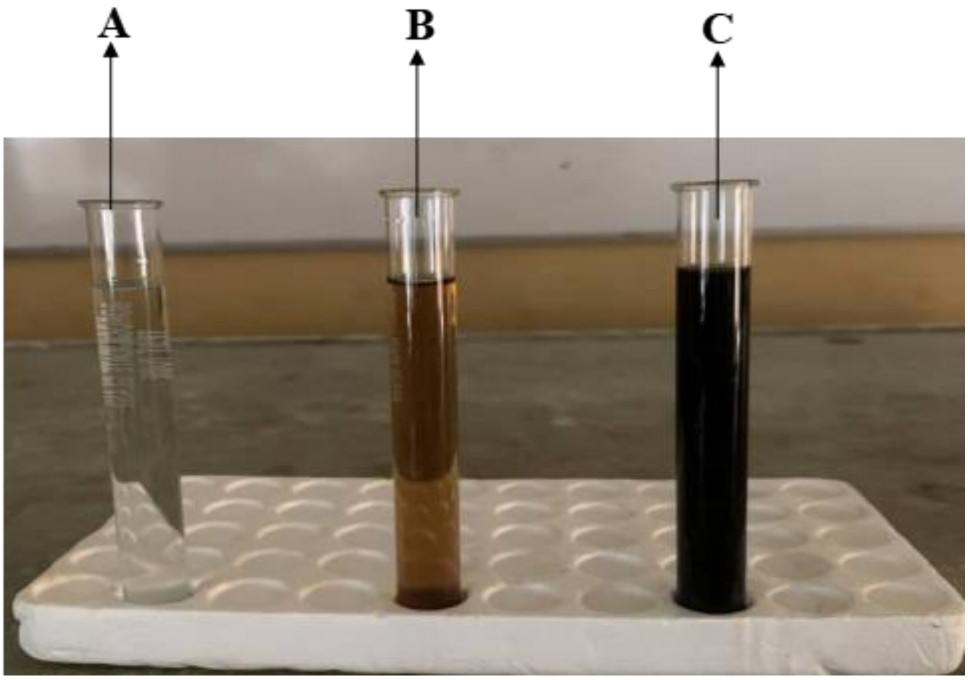

Fig.1: Colour transformation from colorless to brown indicate the formation of morinda lucida sliver nanoparticle (a) colorless solution of $AgNO_3$ (b) morinda lucida sliver nanoparticle (c) morinda lucida leaves extract.

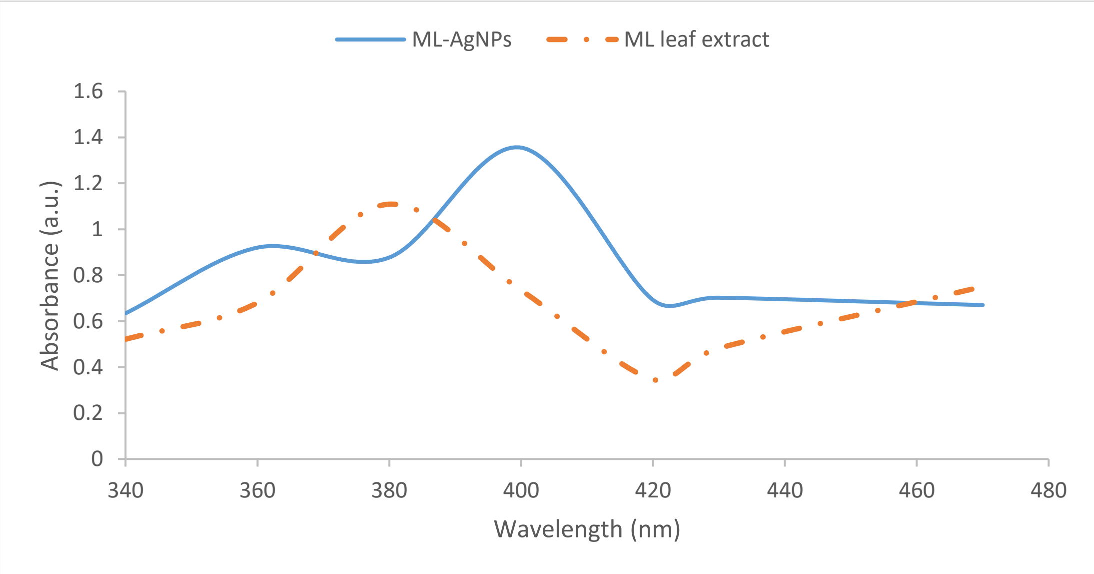

Figure 2. Uv-vis result of both Morinda lucida leaves extract and ML-AgNPs

FTIR Analysis

The FTIR spectrum of Morinda lucida and its synthesised AgNPs is shown in Fig. 3. The spectrum shows peaks for various active biochemical groups, including carboxylic acids, alcohols, phenols, esters, ethers, aldehydes, alkanes, and others, which are involved in the reduction and stabilisation of Ag nanoparticles. The broad band detected at 3300 cm^{-1} in the spectrum could be associated with the overlapping stretching mode of the O-H hydoxyl group and the N-H group of amines, amine salts, C=O of carboxylic acid, and C-H of alkanes and aliens (Chamizo-Ampudia, et al., 2021). The prominent sharp band at 1630cm-1 may be allocated to N-H of amides and amine salts, including C=C oscillation of alkenes, corresponding to alkane (Varadavenkatesan *et al*., 2021). The stretching vibrations of the C-O groups of anhydrides, esters, ethers, alcohols, and phenols; of the C-O-H of alcohols and phenols; and of the C-N of amines are responsible for the peaks centering at 1320cm-1, 1300cm-1, and 1210cm-1. The indicated C=O stretching of carbonyl groups, which suggests the presence of a group of carboxylic acid, the C-H stretching of methyl, methylene, or methoxy groups, was observed at 2910 cm1(Varadavenkatesan et al., 2021). Also, the peaks at 1440 and 1530 cm-1 indicate O-H bending in carboxylic acids and N-O stretching of nitro compounds (Padole and Avari, 2021). Thus, FTIR analysis shows that the main participants in the reduction of Ag+ ions to Ag0 nanoparticles in the Morinda lucida extract extract were the-C=O (carboxyl), -OH (hydroxyl), and N-H (amine) groups. According to the FTIR spectroscopic study, the Morinda lucida leaf extract's carboxylic acid and ascorbic acid may also act as reducing and stabilising tools for silver nanoparticles. The different band evaluations are in line with those reported in the literature for related studies ((Mehmood, Murtaza, Bhatti, and Kausar, 2021). These organic substances were recognised as capping and stabilising groups that helped reduce silver ions to metallic silver, particularly the O-H hydroxyl and carbonyl groups. (Gangwar *et al*., 2021).

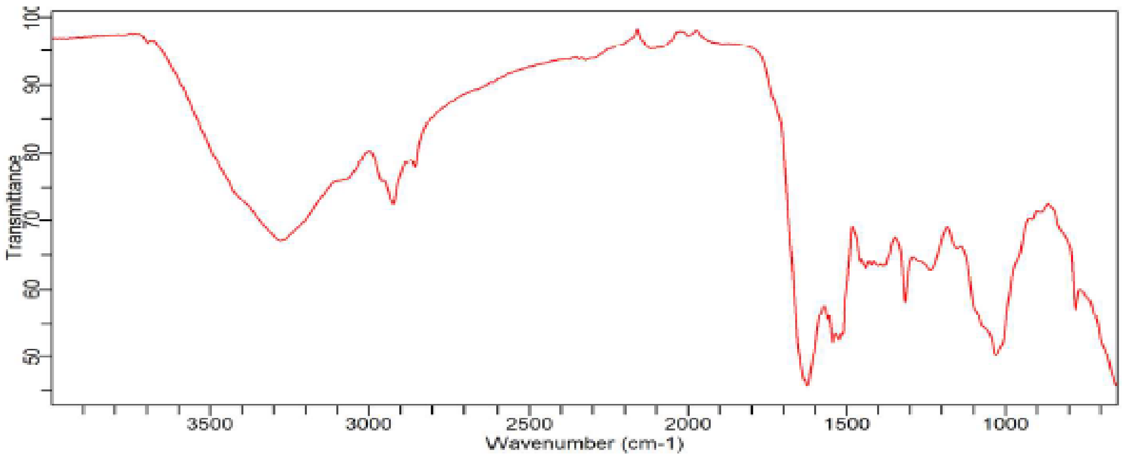

Figure 3. FTIR spectrum of silver nanoparticle using the leaves extract of Morinda lucida

SEM Analysis

The densely packed Ag-NPs produced by the Morinda lucida leaves in the SEM image (Figure 4) further supported the emergence of silver nanostructures. The nanoparticle surface morphology of the filtrate revealed that the Ag-NPs are spherical in shape, evenly distributed, and free from clumping in solution. Through EDS analysis, elements that may be involved in the formation of nanoparticles can be evaluated quantitatively and qualitatively. Silver nanoparticle formation is confirmed by Figure 5, which displays the elemental profile of nanoparticles created using Morinda lucida leaf extracts. Silver nanoparticles in Figure 4 also result in higher counts at 3 keV. Surface plasmon resonance typically causes metallic silver nanocrystals to exhibit an optical absorption peak at about 3 keV. The silver content of the silver nanoparticles depicted in the figure had the highest elemental content, followed by oxygen. The biomolecules attached to the silver nanoparticles' surface are what cause the O peaks to appear (Padole and Avari, 2021).

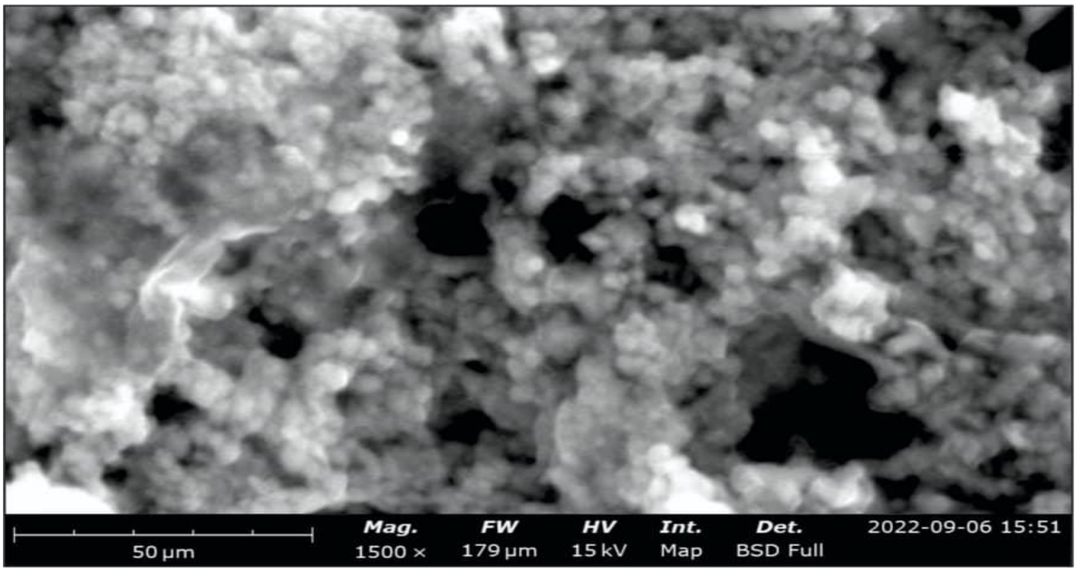

Figure 4. SEM image of silver nanoparticle using the leaves extract of Morinda lucida

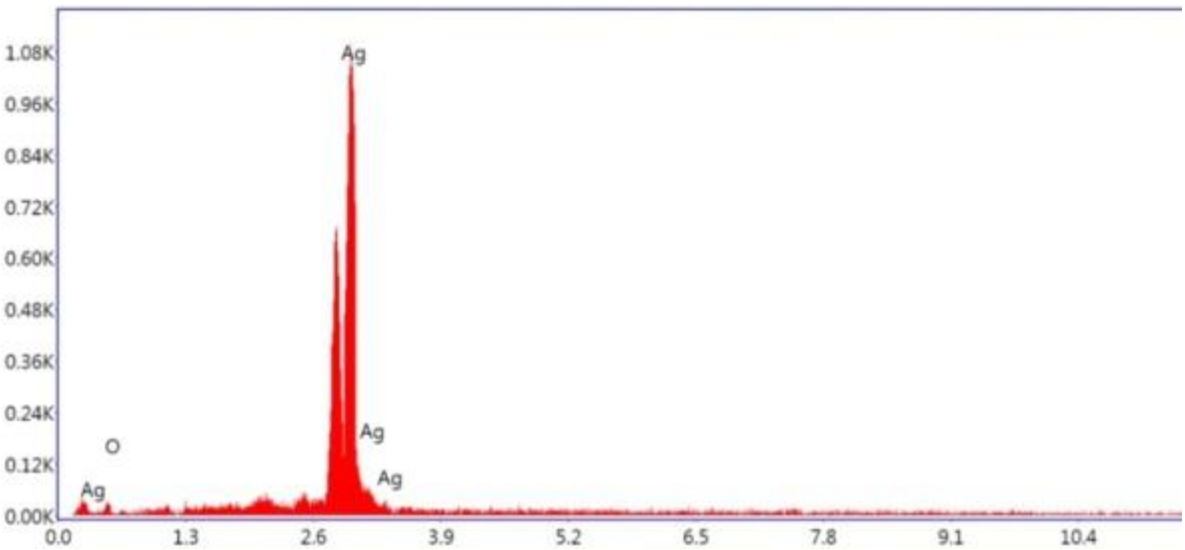

Figure 5. EDS result of silver nanoparticle using the leaves extract of Morinda lucida

Conclusion

There are many advantages to adopting green nanotechnology for the quick, simple, inexpensive, and ecologically responsible production of metallic nanoparticles such as AgNPs using plant extracts. This study used Morinda lucida leaf extract to efficiently bioreduce aqueous AgNO3 and create ML-AgNPs. The creation of stable AgNPs in a variety of sizes and forms was confirmed by the various characterization techniques. According to the SEM results, the synthesised nanoparticles were spherical in shape, less than 50 nm in size, and had an absorbance of 360-400 nm. This was made possible by the bioactive compounds found in these plants. Due to their remarkable eco-safety and financial viability, these green synthesised AgNPs must be used extensively in the medicinal and agricultural fields.

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