Green Synthesis, Characterization and Antimicrobial Potency of Ag–Fe Bimetallic Nanoparticles from Papaya Leaf Extract

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DOI: 10.29322/IJSRP.12.02.2022.p12270
http://dx.doi.org/10.29322/IJSRP.12.02.2022.p12270

Paper Received Date: 30th January 2022
Paper Acceptance Date: 10th February 2022
Paper Publication Date: 21st February 2022

Abstract

Due to their new physical and chemical properties derived from synergistic effects between the two metals, Bimetallic nanomaterials have raised more and more significant concern from worldwide researchers in recent years. These hybrid species are highly desirable for specific technological applications, especially for antimicrobial study. In this article, Ag–Fe bimetallic nanoparticles were synthesized from papaya leaf extract using green method and the same were subjected to various characterization techniques including UV and FT-IR. The UV result of Ag–Fe bimetallic nanoparticles showed the characteristic UV–Vis spectrum with highest peak at 400nm. The FT-IR bands identified were 688.87 cm^{-1}, 1403.65 cm^{-1} 1628.68 cm^{-1}, 3144.50 cm^{-1}, 3697.30 cm^{-1} and 3788.00 cm^{-1}. The antimicrobial study of Ag-Fe Bimetallic Nanoparticles synthesized from papaya leaf extract demonstrated effective antibacterial activity on Pathogenic bacteria. For each concentration investigated, E. coli, demonstrated higher zone of inhibition as compared to other pathogens.

Keywords: Green Synthesis, Bimetallic nanoparticles, Antimicrobial activity, Papaya Leaf Extract

1. Introduction

Due to the wide range of properties and potential applications in the fields of chemistry, materials science, physics, catalysis and even biology, metallic nanoparticles have attracted special interest [1-3]. A bimetallic nanoparticle is a combination of two different metals that exhibit several new and improved properties, and as such, preferred over monometallic nanoparticles. These bimetallic nanoparticles like monometallic nanoparticles can be synthesized using different routes including chemical, physical and green method. As part of environmental remediation encompassing use of biomaterials because of their eco-friendliness, a paradigm shift in technology has recently led to the synthetic protocols involving application of green chemistry [4]. Studies have shown that bimetallic nanoparticles, more than their counterpart, exhibit new and improved properties both in antimicrobial and catalytic activities, hence, bimetallic nanoparticles such as Fe/Ru, Ag−Pd, Ag–Fe bimetallic nanoparticles among many others have been synthesized and put to applications [5-7]. For instance, research had shown that Au–Ag BMNPs show high-quality antibacterial activity against Gram-positive Bacteria *Staphylococcus aureus* and Gram-negative bacteria *Klebsiella pneumoniae* [8-10]. Furthermore, bimetallic nanoparticles such as Ag-Co Bimetallic nanoparticles can be used as potential control for larval population growth. Similarly, Cu-Co bimetallic nanoparticles synthesized using fruit extract palmyra palm can be used as a potential nanolarvicide for mosquito larvae management [11-12].

2. Materials and Methods

2.1 Materials

The materials employed during this work include $AgNO_3$, $Fe(NO_3)_3$, Papaya Leaf, deionized water, hot plate, Whatman no. 1 filter paper, crucible, beaker. All the reagents used during this work were of analytical grade.

2.2 Plant Sample Collection and Preparation

Fresh leaves of Papaya obtained from Banganje, Billiri Local Government Area of Gombe State were transported to Federal University of Kashere via road. The leaves were washed with running tap water and rinsed with de-ionized water. The samples were cut into small sizes and ground with the aid of a crucible. About 50 g of it was weighed and mixed with 500 mL of de-ionized water and boiled at 90^0C on a hot plate with frequent stirring for 20 minutes. The extract was cooled and filtered through Whatman no. 1 filter paper

2.3 Synthesis of Ag–Fe bimetallic nanoparticles.

A solution containing 200 ml each of 0.02 M $AgNO_3$ and $Fe(NO_3)_3$ were gradually mixed with 100 mL of the prepared Papaya Leaf extract on a hot plate at 90^0C while stirring for 30 minutes in a 1000 ml beaker. The formation of the nanoparticles was monitored and confirmed by a change of color of the mixture that changed from light brown to deep brown. The reaction mixture was cooled at room temperature, centrifuged at 14,000 rpm for 10 min, and the supernatant was discharged. The product was subsequently dispersed in sterile distilled water to get rid of any uncoordinated biological materials and finally left to dry at room temperature for 24 hours.

2.4 *Ultraviolet-Visible Spectroscopic Investigation*

The investigation of the optical properties of the green synthesized Ag–Fe bimetallic nanoparticles was carried out by UV-Visible Spectrophotometer Agilent technology model 6705 by determining the absorbance at wavelength of between 200 to 800 nm by placing 1mL sample of the bimetallic NPs synthesized in 1 x 1 cm cuvettes operated at a resolution of 1 nm and de-ionized water as the reference solvent. The results were recorded accordingly.

2.5 *Fourier Transform Infrared Spectrophotometry Analysis*

The green synthesized Ag–Fe bimetallic nanoparticles were characterized using Fourier Transform Infrared Spectroscopy (PerkinElmer Spectrum Version 10.03.09) to account for the functional groups involved in the bio-reduction and capping process from 450 to $4000cm^{-1}$.

2.6 Antibacterial Activity Assay

The antimicrobial potency was investigated by using the agar-well diffusion method as described by other researchers [13-14]. The bacterial isolates; gram-positive bacteria (*Staphylococcus aureus*, *Bacillus anthracis*) and gram-negative bacteria (*Escherichia coli*, and *Salmonella typhi*) first, were cultured in a nutrient broth for 12–18 h before it was used and was standardized to 0.5 McFarland standards (106 cfu ml−1). On a Mueller-Hinton agar (Hi Media), One hundred microliter of the standardized cell suspensions were spread and the agar medium was punched with a 6 mm diameter wells and filled with various concentrations (100, 200, 300, 400 and 500 µg/L) of Ag–Fe bimetallic nanoparticles solutions in equal amounts. After 24 h incubation at 37^0C, the plates were observed for zone of inhibition.

3.0 Results and Discussion

3.1 UV Result

The UV result of Ag–Fe bimetallic nanoparticles showed the characteristic UV–Vis spectrum, with highest peak at 400nm. Although an absorption spectrum resembling that of iron nanoparticles was observed, this suggests the formation of silver-iron core–shell bimetallic nanoparticles. Similar findings have been reported in the synthesis of diverse bimetallic nanoparticles [7, 14-16]. The nano size of the synthesized Ag-Fe BMNPs was confirmed by the characteristic surface plasmon resonance (SPR) band at around 400 nm as seen in UV spectrum (figure 1) and the result is in concordance with that of other researchers [17].

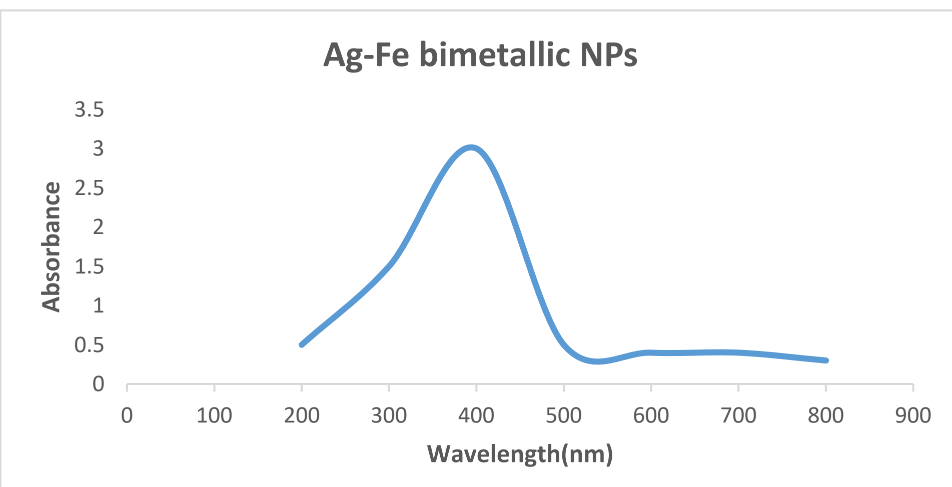

Figure 1. UV–visible absorbance spectra of Ag–Fe bimetallic nanoparticles synthesized using an aqueous extract from Papaya Leaf

3.2 FTI-IR Result

An FT-IR analysis was conducted (figure 2) so as to identify the biomolecules contained in the Papaya Leaf extract, as these are responsible for the reduction of Ag^+ and Fe^+ ions in the synthesis of nanoparticles. 688.87 cm^{-1}, 1403.65 cm^{-1}, 1628.68 cm^{-1}, 3144.50 cm^{-1}, 3697.30 cm^{-1} and 3788.00 cm^{-1} were identified as the FTIR bands. The band at the 3500–3000 cm^{-1} region corresponds to hydroxyl group stretching (OH). The band at 3144.50 cm^{-1}, may be attributed to the stretching of aliphatic hydrocarbon (C–H). The bands at the 1403.65 and 1628.68 cm^{-1} region can be assigned to C=C and C=O stretching, respectively. The band at 688.87 cm−1 may be assigned to the stretching of C-O and finally the band at 3788.00 cm−1 may be attributed to N-H stretching vibration. The result of the FT-IR analysis investigated in this research, corresponds to that of other researchers (17-18). More so, the FT-IR results agrees with the UV result

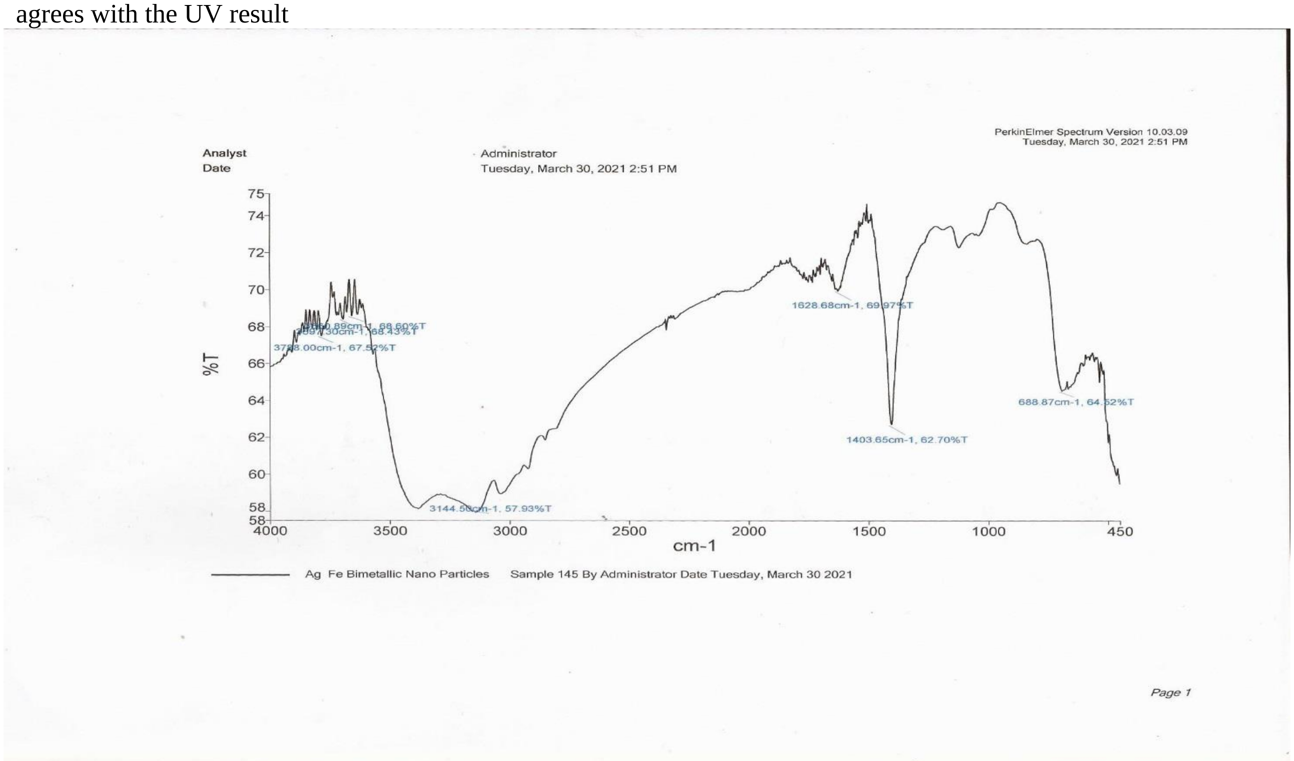

Figure 2. FT-IR Spectra of Ag–Fe bimetallic nanoparticles

Table 1: FT-IR studies of Ag-Fe Bimetallic nanoparticles

Peak(cm^{-1})	Bond	Mode	Transmittance (%)	Intensity
1403.65	C=C	Stretch	62.70	Strong
1628.68	C=O	Stretch	69.97	Strong
688.87	C-O		64.52	Strong
3144.50	C-H	Stretch	57.93	Weak
3697.30	O-H	Stretch	68.43	Strong
3788.00	N-H	Stretch	67.52	Strong

3.3 SEM RESULT

The surface morphology of the Ag-Fe bimetallic nanoparticles synthesized by green method from the extract of papaya leaves was observed with the aid of scanning electron microscope and it reveals that the papaya leaves have a great potency to synthesize Ag-Fe bimetallic nanoparticles which were hexagonal in shape. The result agrees with some researchers' submissions in the earlier literatures [18-19]. Below is the SEM spectrum of Ag-Fe bimetallic nanoparticles from papaya leaf extract.

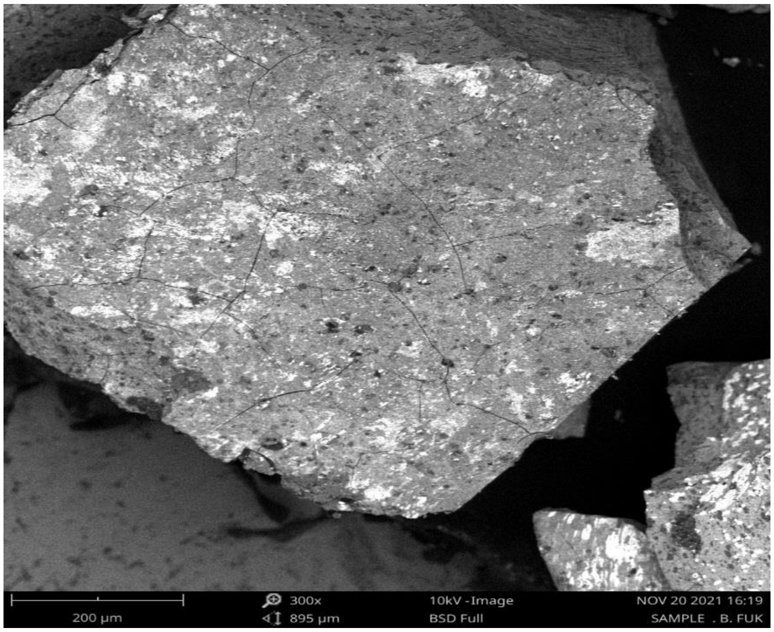

Figure 3. SEM Spectrum of Ag–Fe bimetallic nanoparticles

3.4 Antibacterial Result

Presented below (table 2) is the result of antibacterial investigation of Ag-Fe Bimetallic Nanoparticles against *Staphylococcus aureus*, *Bacillus anthracis*, *Escherichia coli*, and *Salmonella typhi.* Throughout the studies, Augmentin was used as control at concentration of 300µg/L. Different concentrations of 100, 200, 300, 400 and 500µg/L of Ag-Fe Bimetallic Nanoparticles was tested against each pathogen. With increase in concentrations of Ag-Fe Bimetallic Nanoparticles of all the bacteria, the inhibition zone generally increases. This report is in agreement with the one observed by other researchers [7, 19]. At higher concentration of 500µg/L, the zones of inhibition were in the following order; 22.5mm 20.5mm, 15mm and 14mm for *B. anthracis*, *E.* coli, *S. aureus* and *S. typhi* respectively. . For each concentration investigated, *E.* coli, demonstrated higher zone of inhibition as compared to other pathogens. The results of this research therefore indicated that Ag-Fe Bimetallic Nanoparticles synthesized from Papaya Leaf extract demonstrated effective antibacterial activity on Pathogenic bacteria.

Table 2: Antibacterial activity of Ag-Fe bimetallic nanoparticles

Ag-Fe BNPs	TEST of organim	Concentration (mm)					
		100µg/L	200µg/L	300µg/L	400µg/L	500µg/L	Control (Augmentin)
	S. aureus	10.5mm	11.5mm	13mm	14mm	15mm	19mm

B. anthracis	12.5mm	13mm	15mm	17mm	22.5mm	25.5mm
E. coli	13mm	15mm	17.5mm	19.5mm	20.5mm	22mm
S. typhi.	8mm	10mm	12.5mm	13.5mm	14mm	23.5mm

4. **Conclusions:** The green synthesized Ag-Fe Bimetallic Nanoparticles from papaya leaf extract demonstrated effective antibacterial activity on Pathogenic bacteria with E. coli having higher zone of inhibition compared to other pathogens.

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