

International Journal of Scientific Research in Chemistry

Available online at : **www.ijsrch.com**





doi : https://doi.org/10.32628/IJSRCH

Eco-friendly Synthesis, Characterization, and Antimicrobial Study of Mono Metallic Silver Nanoparticles from Khaya senegalensis Aqueous Stem Bark Extract Mela Yoro¹, Rebeccah Bethuel¹, Mary Asugu Mbahi², Robinson Ayibanuah Favour³

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ARTICLEINFO

ABSTRACT

Article History:

Accepted: 05 March 2024 Published: 18 March 2024

Publication Issue

Volume 9, Issue 2 March-April-2024

Page Number

01-09

Healthy Khaya senegalensis stem bark were collected from the Billiri area and were carefully washed under running water, rinsed multiple times with distilled water, and then allowed to dry in the shade to eliminate any remaining moisture. Using a mortar and pestle, the dried materials were ground into a fine powder which was used in making the extract. 10ml of Khaya senegalensis stem bark extract was added to 100ml of 0.01 M aqueous AgNO3 solution. On the hot plate, the mixture was heated to 60°C while being stirred continuously and after some time, AgNPs were formed. The UV analysis showed a maximum absorbance of 2.926 AU at a matching wavelength (λ max) of 400 nm. An OH bond stretching may be the cause of a peak at 1845.08 cm⁻¹. The shoulder peak of aldehydes belongs to the C=O group and is found at 1651.32 cm-1. The peak at 1314.99 cm-1 displays the fingerprint region of functional groups such as carboxylic and alcohol groups. The absorption peak at 1089.75 cm-1 could be the result of the CH2-CH3 bend. The FTIR analysis reveals that the aldehyde (C=O), hydroxyl (OH), and alkane (C-H) are the main agents in the conversion of Ag⁺ to g⁰ nanoparticles. The XRD result revealed that, the nanoparticles under study has face-centered cubic (fcc) and spinel-like structures, with biosynthesized AgNPs having an average particle size of 55.4 nm. Silver nanoparticles were tested for their antibacterial properties against two fungi, Candida albicans and A. niger, as well as gram-positive bacteria, K. pneumonia, and B. subtilis. several concentrations of silver nanoparticles were examined (100, 200, 300, 400, and 500 µg/L) against each pathogen. The inhibitory zone grows as silver nanoparticle concentration increases. The zones of inhibition for B. subtilis, K. pneumonia, A. niger, and C. albicans were 14.5 mm, 25.3 mm,

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23.5 mm, and 19.5 mm, respectively, at a higher concentration of $500\mu g/L$. Compared to all other pathogens under research, K. pneumonia showed a higher zone of inhibition for each concentration examined. The study's findings therefore showed that silver nanoparticles made from Khaya senegalensis stem bark extract had strong antibacterial activity against the pathogenic microorganisms investigated.

Keywords: Eco-friendly Synthesis, Characterization, Antimicrobial, Monometallic, Nanoparticles, *Khaya senegalensis*

I. INTRODUCTION

Since nanoparticles can interact with animals, plants, and microbes by exhibiting unique qualities based on their size, shape, and morphology, nanotechnology is one of the most researched fields of modern materials science. Because of their unique properties resulting from their surface area and particle size, silver nanoparticles (Ag NPs) in particular are among the most promising materials that have garnered much attention and have been extensively studied in various fields including materials science and engineering, biomedical, antimicrobial, and catalytic applications [1-2]. Their unique physical and optical features such as high surface to volume ratio, Surface Plasmon Resonance (SPR), as well as Surface Enhance Raman scattering (SERS) have led in the metallic nanoparticles recent development [3]. Because of these unique qualities, metal nanoparticles are used more often in the fields of sensing and bioimaging, medicine, cosmetics, agriculture, water treatment, and textile waste management [4–11]. Scientists have been very interested in conducting research on advanced nanomaterials of noble metals, such as silver, due to their physiochemical properties, which include size, distribution, and morphology. These properties have also been studied for their magnetic, optical, electronic, and antimicrobial properties [12-16]. Because it offers numerous advantages over chemical

and physical approaches for creating nanoparticles, plant-based nanomaterial synthesis is currently a prominent area of nanotechnology. This approach has many advantages, not all of which are listed here: it is inexpensive, environmentally friendly, simple, easily scaled up for mass-scale synthesis, reasonably reproducible, and most importantly, it doesn't require high pressure, energy, hazardous chemicals, or high temperatures. It also frequently yields more stable materials. In addition, the application of green chemistry concepts to nanotechnology has grown in importance and drawn a lot of attention recently [17-18]. These days, biological techniques are used to synthesize metal and metal oxide nanoparticles since the final particles have the right size and morphology and the properties of the particles are enhanced in a greener way. owing to the rich biodiversity of plants and their potential secondary metabolites, plant parts have been fully deployed in modern days' bio fabrication of a variety of nanoparticles. furthermore, since plant extracts can act as both reducing and stabilizing agents for the biosynthesis of nanoparticles, the utilization of chemical as reductants and stabilizers have been avoided. In comparison to other organisms, plants and their parts help to produce metal nanoparticles that are much stable and can minimize the metal ions faster than that of microbes. Equally they minimize the cost of isolation and culturing bacteria and fungi, thereby enhancing,

compared to their monometallic entities, their costcompetitive feasibility of nanoparticle production and improved antimicrobial properties [19–23].

II. MATERIAL AND METHODS

2.1 Materials

Khaya senegalensis stem, distilled water, silver nitrate (AgNO₃), nutrient agar, culture bottles, and incubators are a few of the supplies used in this study.

2.2. Methods

2.2.1 Sample Gathering and Plant Extract Preparation

The botanist recognized healthy plant samples that were gathered from the Billiri area. The samples were carefully cleaned under running water, rinsed multiple times with distilled water, and then allowed to dry in the shade to eliminate any remaining moisture. Using a mortar and pestle, the dried materials were ground into a fine powder. A 250 ml glass beaker containing approximately 10g of the samples was filled with 100 ml of sterile distilled water, then heated to 60°C for 20 minutes on a hot plate. This was left to cool. The solution was then filtered using Whatman No. 1 filter paper, and the resulting filtrate was employed right away to create silver nanoparticles.



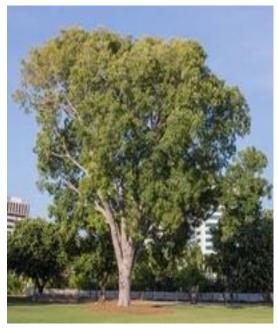


Figure 1: Khaya senegalensis Stem bark

2.2.3 Synthesis of Silver Nanoparticles

With a few minor adjustments, the following procedure which has been documented in the literature [24] was used to synthesize silver nanoparticles:

A standard reaction protocol involved adding 10 milliliters of Khaya senegalensis stem bark extract to 100 milliliters of room temperature 0.01 M aqueous AgNO₃ solution. On the hot plate, the mixture was heated to 60° C while being stirred continuously. It was noted that the hue of the final solution showed that AgNPs had formed.

2.2.4 Antimicrobial Analysis

Here, the antibacterial and antifungal activity of the green generated Silver nanoparticles made from Khaya senegalensis leaf extract was examined using the Agar well diffusion method against a few chosen gram positive bacteria and fungi.

2. 3. Characterization of the Sample Synthesized

2.3.1. UV-Visible Spectral Analysis

By measuring the wavelength of the reaction mixture in the UV-vis spectrum at a resolution of 1 nm (from 200 to 800 nm), the presence of silver nanoparticles was verified.

2.3.2. FT-IR Analysis:

FTIR analysis was used to study the characterization of the active functional groups on the surface of silver nanoparticles (AgNPs) generated from Khaya senegalensis leaves extract. The spectra were scanned at a resolution of 4 cm–1 in the range of 4000–400 cm–1.

2.3.3. X-ray Diffraction (XRD) Analysis

The green produced silver nanoparticles' particle size was measured with an X-ray diffractometer utilizing Cu K (α) and 45 kV of voltage and current.

III. RESULTS AND DISCUSSION

3.1. Identification of color shifts during the production of Ag NPs from stem extract

This time, the addition of crude extract caused a rapid color shift from yellow to light brown after 25 minutes, indicating the rapid reduction of Ag+ to Ag0 in AgNO3 solution. This phenomenon could be attributed to the surface Plasmonic excitement of AgNPs, a report that is consistent with that of the literatures [25].



Figure 2 : (a) Set up for Synthesis of AgNPs (b) stem extract, AgNO₃ & AgNPs from the stem

3.2 UV- Visible Spectrophotometric Analysis

At 200–800 nm, the decrease of Ag+ to Ag0 was measured on a regular basis, with distilled water serving as the blank. Plotting an Ag NP spectrum showed absorbance on the y-axis and wavelength on the x-axis. The creation of Ag NPs was seen through the excitation of the surface Plasmon vibration in the Ag NPs, as indicated by the highest absorption peak observed at absorbance of 2.926 AU with matching wavelengths (λ max) at 400nm. The UV finding concurs with those published in the literature [26–27]. Figure 3 displays the produced Ag NPs' UV–Vis absorption spectra.

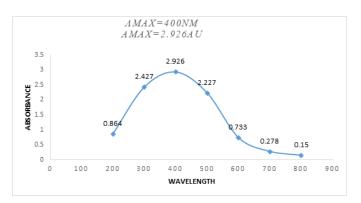


Figure 3: UV-Vis of AgNPs Synthesized from *Khaya* senegalensis stem extract

4.2.2 AgNps from Khaya senegalensis stem analyzed using FTIR.

FTIR analysis was done to look into the functional groups of the Khaya senegalensis stem, and the results

are displayed in figure 5. The stem of Khaya senegalensis exhibits several absorption peaks, which is indicative of its intricate character. A peak at 1845.08 cm-1 might be the result of an OH bond stretching. It is possible to attribute the absorption peak at 1830.98 cm-1 to the C-H stretching vibration of the C-H functional group. The C=O group of aldehydes is allocated the shoulder peak at 1651.32cm-1. The fingerprint region of alcohol and carboxylic groups, which exist as functional groups, is shown by the peak at 1314.99 cm-1. The presence of the CH₂-CH₃ bend may be responsible for the absorption peak at 1089.75 cm-1. The FTIR analysis shows that the primary players in the reduction of Ag+ to Ago nanoparticles are the aldehyde (C=O), hydroxyl (OH), and alkane (C-H) groups of the stem of Khaya senegalensis. Figure 4 below displayed the AgNps FTIR Spectra from the stem bark extract of Khaya senegalensis [26-27]

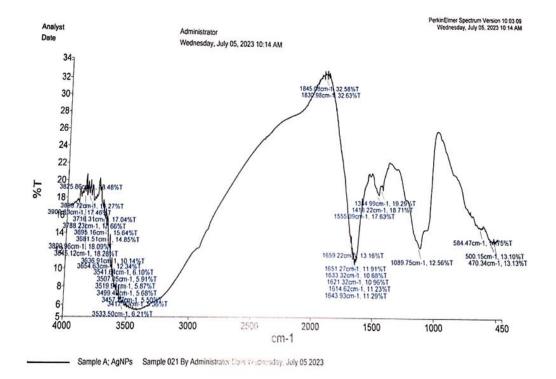


Figure 4: FTIR Spectra of AgNps from Khaya senegalensis stem extract

Frequency (cm ⁻¹)	Bonds/F.group	Vibration Mode	Intensity	
1845.08	ОН	stretch	Strong	
1830.98	C≡C	stretch	Weak	
1651.32	C=O	Bend	Strong	
1314.99	С-Н	Stretch	Weak	
1089.75	С-Н	Bend	Medium	

Table 1: results summary of FTIR of AgNps from Khaya senegalensis stem extract

3.4. X-ray Diffraction

Using Scherer's formula, the XRD pattern shows that the synthetic AgNPs have face-centered cubic and spinel-like structures, with an average particle size of $55.4nm: D=K\lambda/\beta cos\theta$

where D stands for the diameter of silver nanoparticles, λ is the X-ray source wavelength, β is the full width half maximum, and θ is the appropriate diffraction angle to the lattice plane. In addition to adhering to the accurate description of nanoparticles, which are minuscule materials with a size range of 1 to 100 nm, this discovery is consistent with previous research published in several journals [28]. Furthermore, the spectrum showed three strong peaks in the range of 100 to 800. The 2 θ values of 32.49 indicated the prominence of the Bragg reflections.

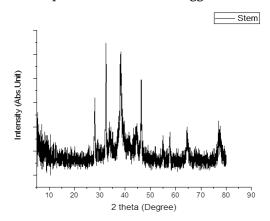


Figure 5: XRD Diffractogram of Green Synthesized AgNPs

The results of the antimicrobial study of Silver Nanoparticles generated using Khaya senegalensis stem bark extract against two fungi, Aspergillus flavus and Candida albicans, as well as Bacillus subtilis and Klebsiella pneumonia (gram +ve bacteria), are shown below (Table 2). Throughout the study, a 300µg/L concentration of augmentin was employed as the control. Various quantities of silver nanoparticles (100, 200, 300, 400, and 500 µg/L) were evaluated against each pathogen. An overall rise in the inhibitory zone was seen with an increase in the concentrations of Silver Nanoparticles of all the pathogens. At a dose of 500µg/L, Bacillus subtilis, Klebsiella pneumonia, Aspergillus flavus, and Candida albicans showed the following zones of inhibition: 14.5 mm, 25.3 mm, 23.5 mm, and 19.5 mm. Compared to all other pathogens under research, K. pneumonia showed a higher zone of inhibition for each concentration examined. According to the study's findings, the pathogenic microorganisms that were chosen for the silver nanoparticles' effective antibacterial activity were those of Khaya senegalensis stem bark extract. It's interesting to note that this result supports the earlier researcher's results [29].

Test of organisms	Concentrations(mm) 100µg/L 200µg/L 300µg/L			Control (Augmentin) 300µg/L 400µg/L 500µg/L		
B. subtilis	7mm	8.5mm	11.5mm	13.5mm	14.5mm	25.5mm
K. pneumonia	13mm	16mm	21.5mm	24mm	25.3mm	29.5mm
Asp. Flavus	11.5mm	13.5mm	15.5mm	21mm	23.5mm	30.5mm
C. albicans	6mm	12mm	14.5mm	15.5mm	19.5mm	21.5mm

Table 2: Antimicrobial activity of AgNPs synthesized from Khaya senegalensis stem bark extract

IV.CONCLUSION

Khaya senegalensis extract was used to bio fabricate silver nanoparticles. Because Khaya senegalensis extract is used in the synthesis at room temperature without the need for a chemical reductant, the process is regarded as environmentally benign because it produces no pollution in the surrounding area. A range of characterization methods, such as FT-IR, XRD, and UV-Visible, were used to ascertain the crystalline size, functional group, and absorption peaks of the relevant nanoparticles. Characterization results from FT-IR, SEM, XRD, and UV spectroscopy revealed that the produced particles are crystalline and in the nanoscale range. The concentration of AgNO₃ and the heat used to prepare the extract are responsible for the particles' stability and small size. The size and capping agents employed have an impact on the AgNPs' antibacterial activity. As evidenced by characterization studies, the particles fall within the nanoscale range. То further establish their antimicrobial agent potency, an antimicrobial assay was conducted against four distinct pathogens: Bacillus subtilis, Klebsiella pneumonia (a grampositive bacterium), Aspergillus Flavus, and Candida albicans. The results of the investigation demonstrated effectiveness of the synthesized the silver nanoparticles against the chosen microbes.

Authors' Contributions: All authors worked together to complete this work. Author MY was responsible for the study's conception and design, statistical analysis, protocol writing, and initial paper draft. The literature searches and study analyses were overseen by authors RB, MAM AND RAF. The final manuscript was read and approved by all writers.

Funding: There was no outside support for this study.

Acknowledgments: The authors express their gratitude to Federal University of Kashere for providing the workspace.

Conflict of Interest: The authors state that there were no financial or commercial ties that might raise questions about a possible conflict of interest while the research was done.

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Cite this article as:

Mela Yoro, Rebeccah Bethuel, Mary Asugu Mbahi, Robinson Ayibanuah Favour, "Eco-friendly Synthesis, Characterization, and Antimicrobial Study of Mono Metallic Silver Nanoparticles from Khaya senegalensis Aqueous Stem Bark Extract", International Journal of Scientific Research in Chemistry (IJSRCH), ISSN : 2456-8457, Volume 9, Issue 2, pp.01-09, March-April.2024