

Article

Green Synthesis, Characterization and Antimicrobial Potency of Silver Nanoparticles from *Psidium guajava* Leaf Extract

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Abstract: In this Research work, Silver Nanoparticles were green synthesized from *Psidium guajava* leaves and different Characterization techniques including UV-Visible, FT-IR, SEM and XRD were all employed to ascertain the absorption peaks, functional group, surface morphology and crystalline size of the nanoparticles respectively. These nanoparticles green synthesized were applied against four different pathogens namely, *S. aureus* (gram- positive bacteria), *E. coli* (gram- negative bacteria), *C. albicans* (gram- positive fungus) and *Aspergillus niger* and the investigation showed that the Silver nanoparticles synthesized were potent against the selected pathogens. From the UV-Vis spectral analysis, it was observed that highest absorption peaks appeared at 400nm and 500nm reflecting the surface plasmon resonance of silver Nanoparticles from guava leaves which is characteristic of Silver Nanoparticles. From the FT-IR spectrum of the sample under studied, the peaks 3416.85 cm^{-1} , 2923.51 cm^{-1} , 1618.95 cm^{-1} , 1384.49 cm^{-1} and 1033.63 cm^{-1} were observed where the absorption band at 3416.85 cm^{-1} corresponds to the stretching due to N-H, while the band at 2923.51 cm^{-1} is associated with C-H stretch of alkane and O-H stretching. The peak at 1618.95 cm^{-1} shows C=C stretching, 1384.49 cm^{-1} reveals the existence of C-H bending and 1033.63 cm^{-1} depicted C-O stretching. The SEM analysis revealed the shape of the nanoparticles as being spherical while XRD result admits that the average size of the green synthesized Ag NPs was 45.5 nm using the Scherrer's formula. Augmentin was used as control at concentration of 300 $\mu\text{g/L}$ throughout antimicrobial studies. Different concentrations of 100, 200, 300, 400 and 500 $\mu\text{g/L}$ of Silver Nanoparticles were tested against each pathogen. It was discovered that with increase in concentrations of Silver Nanoparticles of all the pathogens, there generally appeared to be increase in inhibition zone. At higher concentration of 500 $\mu\text{g/L}$, the zones of inhibition were in the following order; 22.50 mm, 17.00mm, 15.44mm, and 13.23mm for *E. Coli*, *S. aureus*, *C. albicans* and *Aspergillus niger* respectively. For each concentration investigated, *E. coli*, demonstrated higher zone of inhibition as opposed to all other pathogens investigated in this research.

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1. Introduction

The term “Nanoparticles” simply refer to tiny materials with size ranging from 1-100nm. Due to their high surface area and nanoscale size, nanoparticles possess unique physical and chemical properties [1] Such materials and systems can be rationally designed to exhibit novel and significantly improved chemical, biological and physical properties [2]. Though nanoparticles can be synthesized via physical and chemical routes, the

green method is widely used due to its Eco friendliness and cost effectiveness. These nano materials have a range of applications including drug delivery, X- ray imaging, agriculture and photo thermal therapy among others [3]. Furthermore, nanoparticles are known to have antibacterial, fungicidal, and antiviral properties. Because of their characteristics, Ag NPs are useful countermeasures against infectious diseases, which constitute a major issue in the medical field [4,5]. Silver and any other metal like cobalt can be hybridized using green route to form bimetallic nanoparticles. This hybrid green synthesized have improved properties which gives it the potency to check *Culex quinquefasciatus*-Borne Diseases [6]. Furthermore, Bimetallic nanoparticles of most transition elements such as copper – cobalt have been found to be potent as a novel nanolarvicide for mosquito larvae management [7]. These hybrid species are highly desirable for specific technological applications, especially for antimicrobial study [8]. Synthesis of nanoparticles from plant extract using green route is now widely used by most modern day researchers due to its simplicity and environmental friendliness, as opposed to the conventional one such as chemical method [9,10]. More so, biogenic reduction of metal precursors to generate corresponding nanoparticles is environmentally benign, cost effective, free of chemical contaminants for biological and medical applications where purity of nanoparticles is of major concern [11]. In recent times, silver nanoparticles have been investigated extensively owing to their superior physical, chemical, and biological properties, and their superiority stems mainly from the size, shape, composition, crystallinity, and structure of AgNPs compared to their bulk forms [12]. Investigations revealed that nanoparticles are potent against microbes by inhibiting its activities, this therefore, continues to be a topic of great interest to both chemists and biologists alike [13]. In this research, the antimicrobial potency of the green synthesized silver nanoparticles from *Psidium guajava* leaf extract was evaluated.

2. Materials and Methods

2.1. Materials

The materials employed during this work include, *Psidium guajava* leaf, distilled water, Silver nitrate (AgNO_3), Nutrient agar, culture bottle, incubator etc.

2.2. Methods

2.2.1 Sample Collection and Preparation of Plant Extract

Healthy plant samples were collected from the vicinity of Kashere and were washed properly under running tap water. The samples were shade dried and homogenized to fine powder using a mortar and pestle. 10g of powdered *Psidium guajava* Leaves was dissolved in 100ml of distilled water and heated for about 10 minutes at 60°C. The extract was filtered using a whatman No. 1 filter paper and kept for further use.

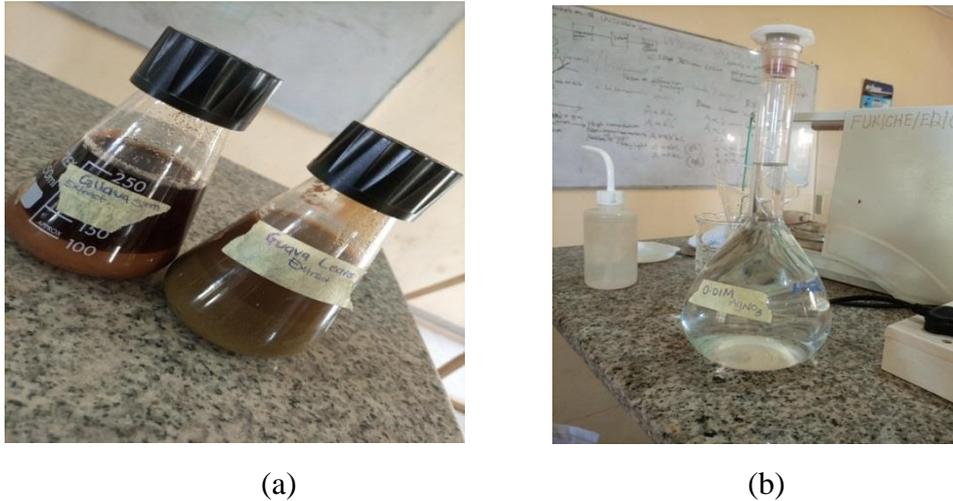


Figure 1. (a) guava leaves extract (b) silver nitrate solution

2.2.2. Synthesis of Silver Nanoparticles using *Psidium guajava* leaf extract:

A solution containing 250 ml of 0.01mol/dm^3 AgNO_3 was gradually mixed with one hundred milliliters of the prepared aqueous leaf extract of *Psidium guajava* (1:5 v/v) on a hot plate at 70°C while stirring for 40 minutes in a 1000 ml beaker. A noticeable change in color of the reaction mixture from light brown to dark brown was conspicuous. The mixture was then stored for about 24 hours after which the nanoparticles settled down. This was evaporated and centrifuged in an oven at 105°C .

2. 3. Characterization of the Sample Synthesized

2.3.1. UV-visible spectra analysis

The silver nanoparticles were confirmed by measuring the wavelength of reaction mixture in the UV-vis spectrum at a resolution of 1 nm (from 200 to 800 nm)

2.3.2. FT-IR analysis:

The characterization of the active functional groups on the surface of silver nanoparticles (AgNPs) synthesized from *Psidium guajava* leaf extract was investigated by FTIR analysis and the spectra was scanned in the range of $4000\text{--}400\text{ cm}^{-1}$ at a resolution of 4 cm^{-1} . The sample was prepared by dispersing the silver nanoparticles uniformly in distilled water as a matrix.

2.3.3. SEM Analysis:

The surface morphology of the nanomaterial (AgNPs) was characterized by scanning electron microscope (SEM).

2.3.4. X-ray Diffraction (XRD) Analysis

The Size of the synthesized silver nanoparticles was investigated using X-ray diffractometer operating at a voltage of 45 kV and current of 40 mA with Cu K (α).

2.4. Antimicrobial analysis

The synthesized silver nanoparticles using plant extracts were examined for antibacterial and antifungal potential by agar well diffusion method against some selected gram positive and gram negative bacteria and fungi.

3. Results and Discussion

3.1. Silver Nanoparticles' Formation and UV- Visible Spectrophotometric Analysis

The formation of Silver Nanoparticles first, was identified by color change from brown to orange immediately at the spot and later changed to reddish brown (Figure 2) after the nucleation of the metal ions indicating that phytoconstituents of *Psidium guajava* caused the reduction of Ag into AgNPs in which the phenomenon could be attributed to the surface Plasmon absorption. Similarly, from the UV-Vis spectral analysis, it can be seen that highest absorption peaks appeared at 400 and 500 nm (Figure 3) reflecting the surface plasmon resonance of silver NPs from guava leaves which is characteristic of Silver Nanoparticles. This finding agrees with those of other researchers [14,15].

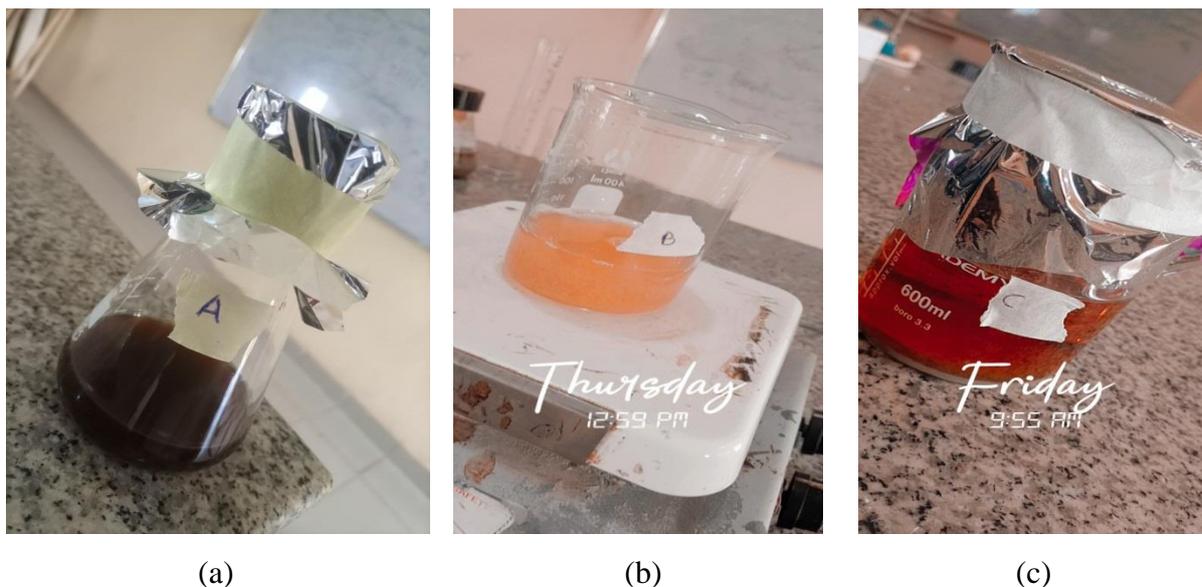


Figure 2. color change of *psidium guajava* leaves extract, before (a), immediately (b), and after addition of silver nitrate (AgNO_3) (c).

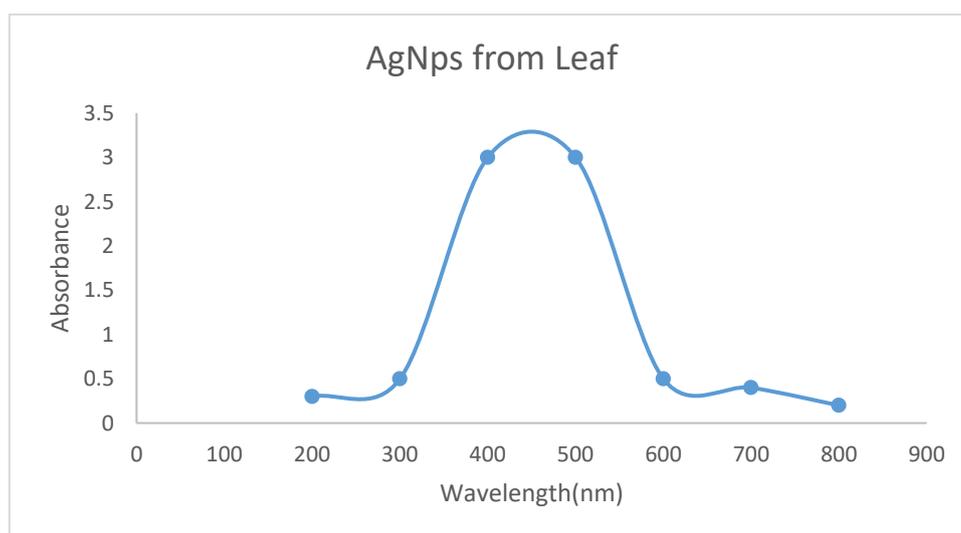


Figure 3. UV-Vis Spectrum of silver nanoparticles (AgNPs) synthesized from *Psidium guajava* leaves

Note: The reduction of Ag was measured periodically at 200-800nm, using distilled water as the blank. A spectrum of NPs was plotted with wavelength on x-axis and absorbance on y-axis.

3.2. FT-IR Interpretation

With the aid of different phytochemicals which would function as reducing, stabilizing and capping agent, FT-IR seeks knowledge about the functional groups present in the synthesized silver nanoparticles for understanding their changes from inorganic silver nitrate (AgNO_3) to elemental silver. From the FT-IR spectrum of the sample under study, the peaks 3416.85 cm^{-1} , 2923.51 cm^{-1} , 1618.95 cm^{-1} , 1384.49 cm^{-1} and 1033.63 cm^{-1} were observed where the absorption band at 3416.85 cm^{-1} corresponds to the stretching due to N-H, while the band at 2923.51 cm^{-1} is associated with C-H stretch of alkane and O-H stretching, 1618.95 cm^{-1} peak shows C=C stretching, 1384.49 cm^{-1} reveals the existence of C-H bending and 1033.63 cm^{-1} depicted C-O stretching. The variations in the FT-IR spectrum indicates the presence of bioactive molecules in plant extracts that participated in the reduction of silver nitrate (AgNO_3) and the formation of silver nanoparticles. Interestingly, this result is in concordance with most of the existing literatures [16-18]

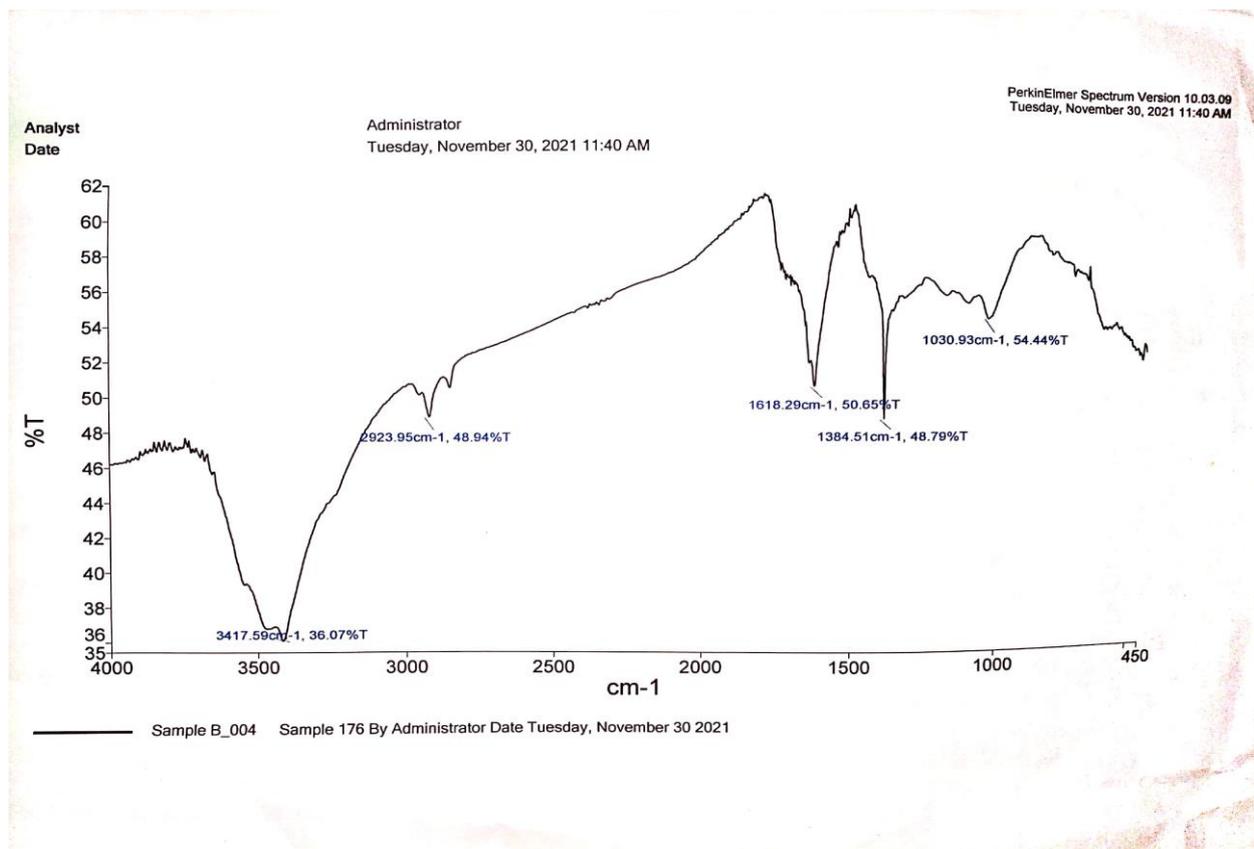


Figure 4. FT-IR Spectrum of Green Synthesized Silver Nanoparticles from *psidium guajava* leaves extract

3.3. Scanning Electron Microscopy (SEM)

The morphology and crystalline structures of bio-prepared Ag NPs were studied via SEM. In Figure 5, the spherical NPs were evidently recognized and dispersed within the size range of 20–80 nm. It is interesting that with increases in the amount of the leaves extract, the SEM images of Ag NPs vary. Therefore, it is significant that the shapes and sizes of Ag NPs depend on the plant extract concentration, which changes the optical and electronic property of NPs. It is also worthy of note that, the reduction of Ag^+ to Ag^0 may be mainly due to the presence of secondary metabolites from the plant extract [19,20].

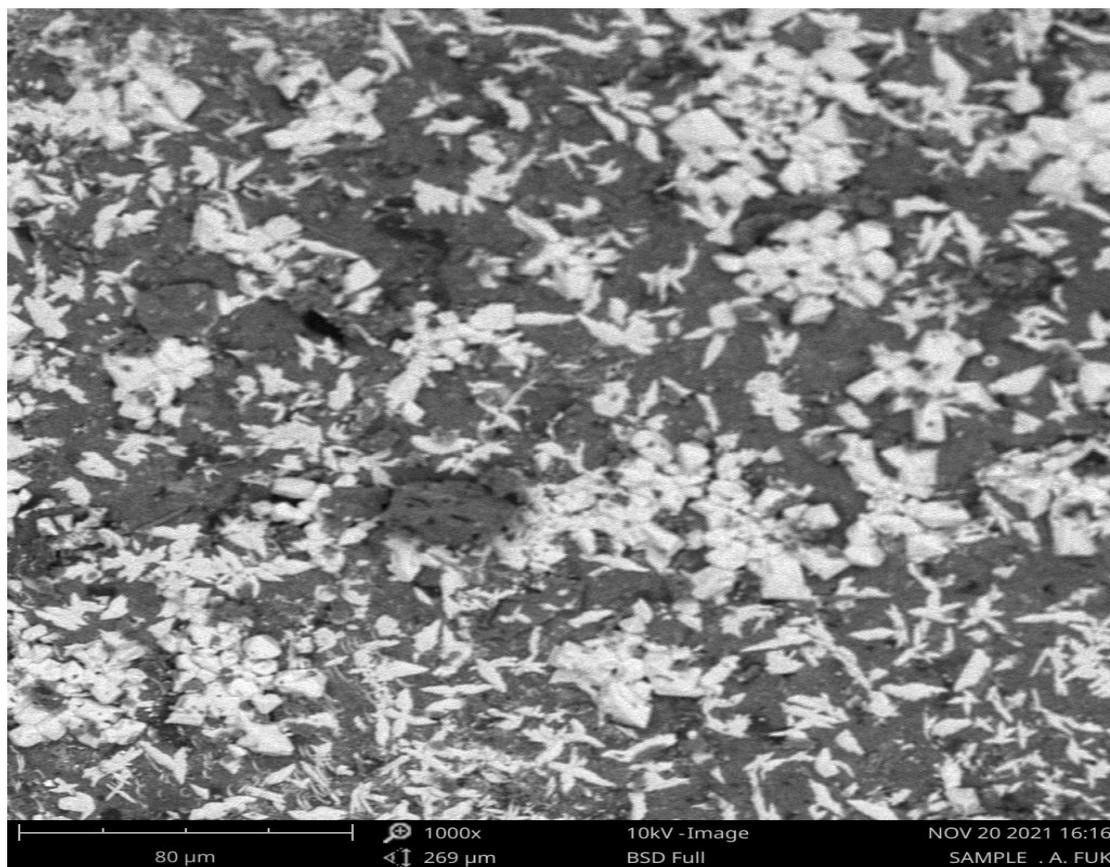


Figure 5. SEM Spectrum of Silver Nanoparticles from *Psidium guajava* leaves extract.

3.4. XRD Analysis

The X-ray diffraction patterns of green synthesized silver nanoparticles is shown below, and a careful study of the spectrum showed that the structure of AgNPs under investigation has a face-centred cubic (fcc) structure. For the synthesized silver nanoparticles of *Psidium guajava* leaves extract, the average size of the green synthesized Ag NPs was obtained to be 45.5 nm.

using the Scherrer equation: $D = K\lambda / \beta \cos\theta$, where

K is a constant equal 1,

λ is the Xray source wavelength

β is the full width half maximum,

θ is the corresponding diffraction angle to the lattice plane and finally,

D denotes the diameter of silver nanoparticles

This finding corresponds to the earlier literatures [21,22].

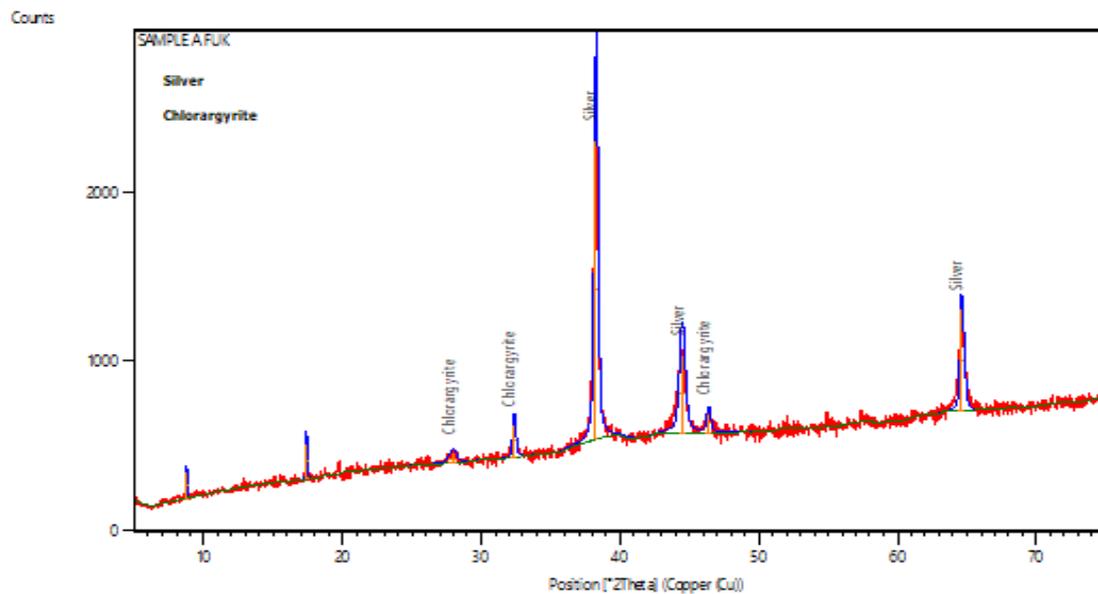


Figure 6. XRD Spectrum of Green Synthesized Ag NPs

3.5. Antimicrobial Activity

Presented below (Table 1) is the result of antimicrobial investigation of Silver Nanoparticles against *Staphylococcus aureus*, *Candida albicans*, *Escherichia coli*, and *Aspergillus niger*. Throughout the studies, Augmentin was used as control at concentration of 300 μ g/L. Different concentrations of 100, 200, 300, 400 and 500 μ g/L of Silver Nanoparticles was tested against each pathogen. With increase in concentrations of Silver Nanoparticles of all the pathogens, there generally appeared to be increase in inhibition zone. Interestingly, this finding validates the report by the earlier researcher [21]. At higher concentration of 500 μ g/L, the zones of inhibition were in the following order; 22.50 mm, 17.00mm, 15.44mm, and 13.23mm for *E. Coli*, *S. aureus*, *C. albicans* and *Aspergillus niger* respectively. For each concentration investigated, *E. coli*, demonstrated higher zone of inhibition as opposed to all other pathogens under investigation. The results of this research therefore indicated that Silver Nanoparticles synthesized from *Psidium guajava* leaves extract demonstrated effective antimicrobial activity on the selected Pathogenic microbes.

Table 1. Antimicrobial activity of Ag nanoparticles

AgNPs Test Organism	Concentration(mm)					Control (Augmentin)
	100 μ g/L	200 μ g/L	300 μ g/L	400 μ g/L	500 μ g/L	
<i>S.aureus.</i>	11.33mm	11.44mm	13.33mm	14.00mm	17.00mm	29.00mm
<i>E.Coli</i>	12.32 mm	13.55mm	15.66 mm	17.33mm	22.50 mm	25.33mm
<i>C.albican</i>	11.33mm	11.66mm	12.33mm	14.33mm	15.44mm	28.00mm
Aspergillus Niger	10.13mm	11.10mm	12.33mm	12.99mm	13.23mm	23.33mm

4. Conclusion

Silver Nanoparticles were green synthesized from *Psidium guajava* leaves and different Characterization techniques such as UV-Visible, FT-IR, SEM and XRD were all employed to ascertain the absorption peaks, functional group, surface morphology and crystalline size of the nanoparticles in question. These nanoparticles green synthesized were

applied against four different pathogens namely, *S. aureus*, *E. coli*, *C. albican* and *Aspergillus niger* and the investigation showed that the Silver nanoparticles synthesized were potent against the selected pathogens.

Authors' Contributions: This work was carried out in collaboration among all authors. Author MY designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author AG managed the analyses of the study. Authors JJ and AI managed literature searches. All authors read and approved the final manuscript.

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Conflict of Interest: The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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