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BIOSYNTHESIS, CHARACTERIZATION AND ANTIMICROBIAL STUDY OF SILVER NANOPARTICLES (AgNPs)

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ABSTRACT
In this paper, biological synthesis of silver nanoparticles (AgNPs) using Syzygium guineenses stem extract with 1mM, 2mM and 3mM AgNO3 concentrations has been presented. The plant extract was prepared with distilled water. The characterization and morphological composition of the synthesized AgNPs were determined by UV-visible spectroscopy and SEM respectively, while FTIR analysis was performed to identify the presence of the possible functional groups in the synthesized nano particles. It was observed from the UV and SEM analyses that the particles formed have diameters in the range of 23.5nm - 89.3nm, which is the range of nanoparticle size. Antibacterial test was carried out on the sample with six pathogenic microbes (Methicillin Resistant Staphylococus aureas, Vancomycin Resistant Entrococci, Staphylococcus aureas, Bacillus sublitis, Escherichia coli, and Pseudomonas aeruginosa) to ascertain the antimicrobial activity of the synthesized AgNPs. Both the characterization and antimicrobial activity test were very successful and could lead to significant economic viability, as well as being environmentally friendly for treatment of some infectious diseases.
Keywords: Syzygium guineenses, Green Chemistry, Spectroscopy, Optoelectronics, Biomedical Sensors

INTRODUCTION

Nanoscience is one of the critical fields of modern science that has received and still receiving enormous attention by several researchers both in the academia and industries due to its wider range of applications in various aspects of human endeavour (Philip, 2010). It is a branch of contemporary science that deals with the study of particles on nanoscale (research dealing with synthesis, design and development of particles structure of size ranging from approximately 1 -100 nm) (Bindhani*et al.,* 2013). Nanostructure materials have created extreme scientific and technological attention due to their potential applications in diverse areas such as electronics, optics, sensors, information and communication technology, stain-resistant fabrics, transparent sunscreen lotions, scratch free paints for cars, products tagging, textiles, biomedicine, health care and antibacterial activity (Kumar and Yadav, 2009).

Over the last decade, noble metal nanoparticles such as platinum (Pt), Copper (Cu), gold (Au), and silver (Ag) nanoparticles have attracted a considerable interest owing to their imperative applications. Metal nanoparticles such as gold (AuNPs) and silver (AgNPs) have been found to exhibit many inhibitory and bactericidal effects and as such their applications are extended as antimicrobial agents (Sinha and Manjhi, 2015). AgNPs are exceptional elements for the Surface Enhanced Raman Scattering (SERS) to explore single molecules, in addition to their popular function as catalysts for speeding up some chemical reactions. Furthermore, they have been employed in other diverse applications such as electrical conducting, sensing and optical applications.

This pave ways for the fabrication and synthesis of nanoparticles that can be arranged into multifarious architectures. There has been a lot of research works in area of synthesis and fabrication of nanoparticles of different materials. Sylvester *et al*. (2004) synthesized AgNPs with laser ablation of metallic bulk materials in solution. The metallic colloidal particles formed were stable without the presence of any chemical reagent in the solution. Although this method results in the production of pure colloids, which will be useful for further applications in medicine, pharmaceutical industries, etc., however, it could not be economically viable due to the cost implication involved in its implementation and sustenance.

Jung *et al*. (2006) synthesized AgNPs by means of a small ceramic heater that had a local heating area. The result of the experiment showed that the evaporated vapour can cool at a duly faster rate. This is largely due the fact that the ratio of temperature to height (temperature gradient) in the neighborhood of the heater surface is extremely steep in relation to that of a tube furnace.

Consequently, synthesis of small nanoparticles in high concentration was possible. However, this method requires a lot of time and may be associated with toxic by products.

Kumar and Yadav (2009) presented investigative studies of biological synthesis of silver and gold nanoparticles and their applications. An analytic review of the importance of plants and their extracts in the synthesis of silver and gold nanoparticles and their potential applications in various human endeavour were presented. It was shown that the biological process does not require tedious process of maintaining cell cultures and can be efficiently used in the synthesis of gold and silver nanoparticles without causing harm to ecosystem. However, the method of nanoparticle synthesis by process plants is not yet fully developed and well understood.

Dwivedi and Gopal (2010) presented a report on facile and rapid biosynthesis of silver and gold nanoparticles from Chenopodium album, an obnoxious weed. Silver and gold nanoparticles were synthesized from their salt solutions using the aqueous leaf extract of the herb as mild reducing. The dimensions of silver and gold particles in the range of 10 – 30 nm via UV-visible spectrometer with plasmon resonance of 460 and 540 nm respectively were observed. Microscopic analyses: Transmission Electron Microscopy (TEM), X-ray Diffraction (XRD), Electron Dispersive X-ray (EDX) and Fourier Transforms Infrared (FTIR) were carried out in order to characterize the nanoparticles.

Umer*et al*. (2012) carried out an investigative review of various chemical methods for the synthesis of copper nanoparticles; and finally selected the best method amongst them. A thorough research was made before the selection of chemical reducing agent so that an environmentally benign reducing agent was used. Despite the claim that the selected approach was the most suitable chemical method, cost effective and environmentally friendly, scientific justification to such a claim was not explicitly evident.

Annamalai *et al*. (2013) developed a simple biological technique for the synthesis of silver and gold nanoparticles using Chrysopogonzizanioides by the bio-reduction of silver nitrate ($AgNO_3$) and chloroauric acid ($HAuCl_4$) respectively. Silver or gold ions were reduced to Ag or Au particles by the water-soluble organics present in the plant materials. Scanning Electron Microscopic (SEM) analysis was carried out and the results from the analysis confirmed that aqueous gold ions when allowed to be cooled were reduced and lead to the biosynthesis of gold nanoparticles having a range of size 20 - 50 nm.

Basavegowda *et al*. (2014) studied a plant mediated synthesis of gold nanoparticles using fruit extracts of Ananascomosus. UV-visible absorption spectra were used to characterize the nanoparticles. Morphological, elemental compositions and crystalline phase were found by SEM, energy dispersive X-ray spectroscopy and selected area electron diffraction. The presence of gold nanoparticles in the extracts was confirmed by FTIR analysis. The authors claimed to have presented a first attempt to synthesizing gold nanoparticles (AuNPs) using extracts of *Ananus comosus* fruit extracts.

Bindhani and Panigrahi (2014) reported a novel approach for biosynthesis of gold nanoparticles using Withaniasomnifera as reductants and stabilizers. The study has demonstrated a bio-reductive synthesis of nano-sized gold particles using Withaniasomnifera ethanol leaf extract appears was eco-friendly. These gold nanoparticles produced were characterized by means of UV spectrophotometer, FTIR and TEM. From the results of characterization of AuNPs, it was revealed that the morphology of the AuNPs is dependent on the concentration of the extract and pH of the medium used. However, antimicrobial analysis was not performed on the sample of the plant extract. Unfortunately, infectious diseases still continue to be major health issue worldwide.

Lots of studies and novel approaches have been recently devoted to understanding the channels of disease dissemination for the purpose of manufacturing new drugs. The present research opens a new avenue for the green synthesis of nano materials. Silver has been known for its anti-bacterial properties since ancient times and therefore used for prevention and control of many infections. In line with that, this study seeks to employ biological approach using plant extracts to synthesize & assemble AgNPs. The purpose is to develop a nontoxic and eco-friendly acceptable green chemistry procedure. Another important reason for the choice of plant extract is that the tedious processes such as intracellular synthesis and multiple purification steps or the maintenance of microbial cell cultures are not required.

Therefore, this study is aimed at synthesizing silver nanoparticles (AgNPs) using *Syzygium guineenses* (Malmo in Hausa language) stem bark sample extract with the view to exploring its medicinal importance in curbing some tropical bacterial diseases. In addition to that, the study will demonstrate the viability of using clean and environmentally friendly process (green chemistry) for the synthesis and extraction of nanoparticles. Methicillin Resistant *Staphylococus aureus*, Vancomycin Resistant Entrococci, *Staphylococcus aureus, Bacillus sublitis, Escherichia coli*, and *Pseudomonas aeruginosa*) pathogenic microbes were employed. The objectives of this paper include:

i. Synthesis of AgNPs using *Syzygium guineenses* stem bark sample extract.
ii. Monitoring the formation of the synthesized AgNPsusing UV spectrometer.
iii. Characterizing the synthesized AgNPs using FTIR and SEM
iv. Studying the antimicrobial activity of the synthesized AgNPs

The rest of the paper is organized as follows: In Section 2, details of our approach, methods and materials used are explicitly presented. While analyses of the results obtained, relevance and significance are discussed in Section 3. Finally, conclusion is presented in Section 4.

MATERIALS AND METHODS

The sample of the *Syzygium guineenses* stem bark was obtained from the locally available fully grown plants.

It was kept in the shade for two weeks to dry; and then grinded to obtain the powdered form of the crude stem sample. Silver nitrate ($AgNO_3$) and distilled water were then added. Both the Silver nitrate ($AgNO_3$) and distilled water were collected from the departmental store, Department of Chemistry Ahmadu Bello University, Zaria Nigeria for the purpose of silver nanoparticles synthesis.

50g of the fine powdered stem bark sample was weighed and transferred into Erlenmeyer flask. 300 ml of distilled water was then added to conical flask; and the mixture heated to about 40^0 C - 50^0 C for about 4 - 5 hours. The heated mixture was then allowed to stand for 48 hours. The cooled mixture was then filtered and washed two or three times with distilled water in order to obtain the extract.

The respective masses of $AgNO_3$ solution for 1mM, 2mM and 3mM were calculated as: 0.17g, 0.34g, and 0.51g; and weighed into three separate 1000ml volumetric flask. Distilled water was added and stirred well for complete dissolution, then made up to the mark and kept for the synthesis.

Biosynthesis of the AgNPs

50ml of 1mM, 2mM and 3mM of $AgNO_3$ solution were respectively added into 20ml of the sample extract in three different conical flasks at a temperature of about 50^0C to 60^0C. When plant extract was mixed with $AgNO_3$ solution in some appropriate ratio (1:8 ratio), silver ions (Ag+) were reduced to silver atoms (Ag^0) nanoparticles. This was followed by a sudden change in brown to blackish-brown colour in the aqueous solution of the sample extracts as a result of the excitation of surface Plasmon vibration with AgNPs. The colour changes of the sample when the aqueous plant extract was mixed with an $AgNO_3$ solution is depicted in Fig. 1(a) and (b). The mixture was kept at room temperature for about 24 hours. The formation of a darckish-brown colour confirmed the presence of AgNPs; because AgNPs has been established to exhibit change in colour in aqueous solution due to excitation of surface Plasmon resonance in the AgNPs (Patel *et al.,* 2015).

(a) (b)

Fig. 1: Formation of AgNPs detected by the colour change. (a) Plant sample extract without $AgNO_3$ Solution and (b) Plant Sample extract with $AgNO_3$ Solution**.**

Characterization of the AgNPs

The characterization of AgNPs was done using UV analysis, FTIR, and SEM. The UV and SEM analyses were carried out in the Materials Analysis and Research Laboratory of the Department of Chemical Engineering, Ahmadu Bello University, Zaria while FTIR was done at National Research Institute of Chemical Technology (NARICT), Zaria.

UV-visible Spectroscopic Analysis

By means of a syringe, $1cm^3$ of each of the three concentrations of the mixture was transferred into sample bottles and diluted with $3cm^3$ of distilled water. The samples were monitored via UV-visible machine and recorded the UV-visible spectra of the reaction mixture for 120 min at 20 min interval. The UV-visible spectra of the sample placed in 1 cm plastic cell path length produced AgNPs were recorded between the wavelengths of 350nm -700nm to study spectral absorbance of the AgNPs. The analysis was carried out at room temperature using quartz corvettes (1cm optical path).

Fourier Transform Infrared Spectroscopy (FTIR) Analysis

0.5% weight of the sample extract was used to prepare a potassium bromide disc (KBr) by grinding it with the potassium bromide. The entire mixture was compressed into a transparent disc. The compressed mixture was then transferred to the FTIR analyzer and the corresponding spectrum was obtained, depicting clearly the transmittance (in %) against the wave numbers (in cm^{-1}) of various functional groups in the respective samples in the region of 400 - 4000 cm^{-1} at a resolution of 8 cm^{-1} at 25^0 C. These were identified by comparing the empirical values of these parameters with those in the library. Therefore, FTIR measurements are carried out to identify the possible chemical groups present in the AgNPs responsible for reduction, and efficient stabilization of AgNPs.

Scanning Electron Microscopy (SEM) Analysis

Scanning electron microscopic (SEM) analysis was carried out using SEM machine (Hitachi S - 4500). Thin films of the sample were prepared on a carbon coated copper grid by simply adding a very small amount of the sample on the grid. By means of a blotting paper, excess solution from the sample extract was removed. The resulting film on the SEM grid was then kept under mercury lamp for about 5 to 10 min in order to dry.

The dried samples thus obtained were observed at 10000x magnification. The nanoparticle pellet obtained after purification was subjected to energy dispersive X-ray analysis (SEM-EDX) at 20 kV using AZTEC software for the analysis. The SEM analysis is carried out in order to characterize the size, shape, morphology and distribution of synthesized AgNPs.

Antimicrobial study of AgNPs

The antimicrobial activity of Syzygiumguineenses extract was determined with the help some pathogenic microbes. The microbes were gotten from the Department of Medical Microbiology, Ahmadu Bello University Teaching Hospital Zaria. 1000µgml^{-1} concentration of the extract was obtained by dissolving 0.01mg of the extract in 10µL of DIMSO. This was used as the initial concentration of the extract for checking the antimicrobial activities from the plant. The medium that was used as a growth channel of the microbes is Muller Hinton agar; and was prepared according to the manufacturer's specifications (sterilized at 121^{0}C for 15min). It was poured into sterile Petri dishes and was left to cool and solidify.

A diffusion method (Kirby-Baurer) was used for screening the extract. 0.1 of the standard inoculums of the test microbes was seeded with the prepared solution. Sterile swab was used to evenly spread the inoculums over the surface of the medium. Sterile paper disc was saturated with the 1000µgml^{-1}sample extract concentration; and was then placed at the centre of each inoculated medium. The inoculated medium was then wrapped at 37^{0}C for 24 hrs, after which the plate of the medium was observed to prevent the growth of the zone (zone inhibition growth). A transparent ruler (in millimeter) was used to measure the zone.

Minimum Inhibitory Concentration

A broth dilution method based on the clinical laboratory standardization institute was used to determine the minimum inhibitory concentration of the sample extract. Muller Hinton Medium broth was prepared, 10 were put into test tubes and sterile at 121^{0}C for 15 – 20 min and allowed to cool. Normal was prepared, 10 were dispersed into sterile test tube and the test microbe was inoculated and incubated at 37^{0}C for 5 hrs. MC-Farland's turbidity standard scale number 0.5 was used to produce turbid solution. The test microbe was diluted in the normal saline solution continuously until turbidity marched that of the MC-Farland's Scale via visual comparison. At this point, the test microbe concentration was found to be 1.5×10^8 µgml^{-1}. Incubations were made at 37^{0}C for 24 hrs after which each test tube of the broth was observed for turbidity (growth). The lowest concentration of the extract in the broth which shows no turbidity was recorded as the minimum inhibitory concentration.

RESULTS AND DISCUSSION

UV-visible Absorbance

The formation of AgNPs due to the change of colour of the aqueous solution *Syzygium guineense* extract (plant extract) with $AgNO_3$ solution was authenticated by UV-visible spectrometer. The relationship between the absorbance and wavelength for the sample extract, 1mM, 2mM and 3mM $AgNO_3$ solutions for 0 – 120 min at 20 min interval (0, 20, 40, 60 and 80 min) are shown in Figs. 2 – 4 respectively.

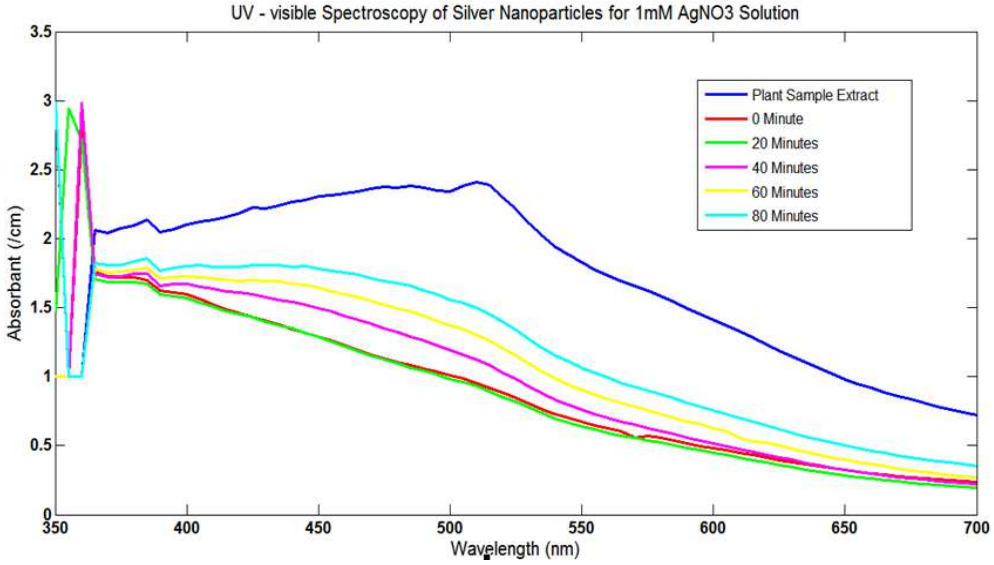

Fig. 2: UV-visible spectra showing absorption as a function of wavelength for 1mM aqueous solution of $AgNO_3$ with *Syzygium guineense* extract.

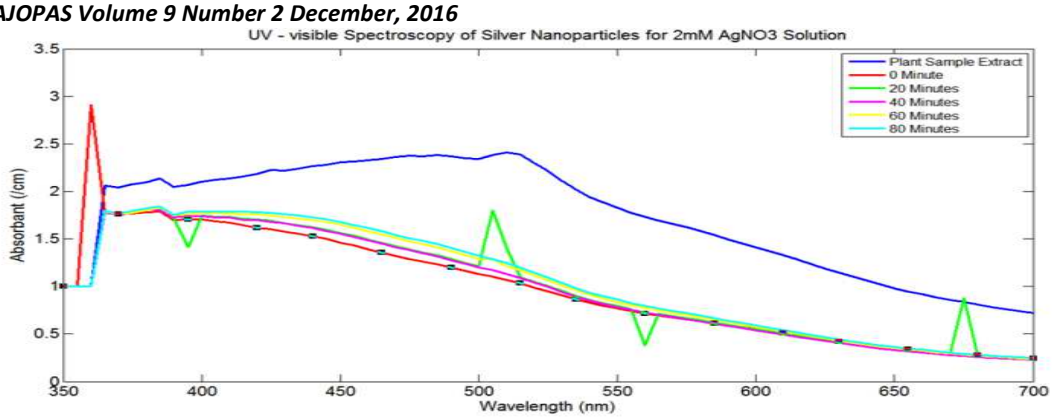

Fig. 3: UV-visible spectra showing absorption as a function of wavelength for 2mM aqueous solution of $AgNO_3$ with Syzygiumguineense stem bark extract.

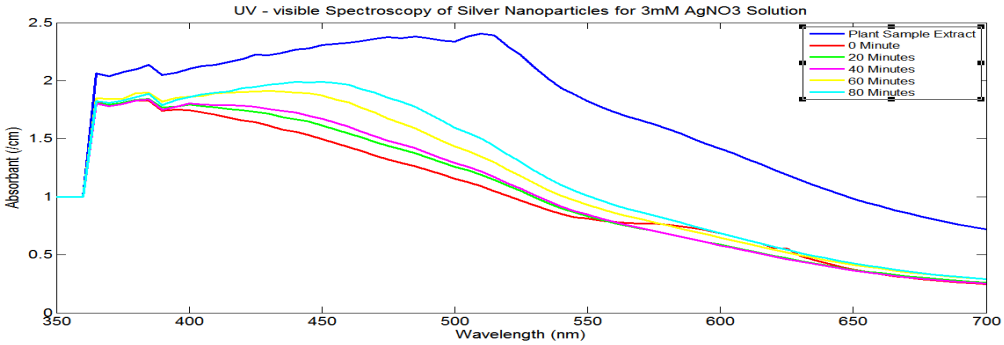

Fig. 4: UV-visible spectra showing absorption as a function of wavelength for 3mM aqueous solution of $AgNO_3$ with Syzygiumguineense extract.

As can be seen from Figs. 2 – 4 above, the absorbance peak occurs in the range of 385nm - 510nm wavelength. For the mixture of the sample extract with 1mM $AgNO_3$ solution, the peak absorbance for 0, 20, 40, 60 and 80 minutes reaction mixture are respectively 355nm, 360nm, 360nm, 385nm and 440nm, and the sample having peak absorbance at 510nm (Figure 2). Similarly, for the mixture of the extract with 2mM $AgNO_3$ solution (Figure 3), peak absorbance for 0, 20, 40, 60 and 80 minutes occurs at 360nm, 505nm, 385nm, 385nm, and 385nm respectively. Peak absorbance at 385nm, 385nm, 385nm, 430nm and 450nm were also observed for 0, 20, 40, 60 and 80 min reaction mixture of the sample extract with 3mM $AgNO_3$ solution.

Based on these results, it can be concluded that the UV- visible spectra of the mixture Syzygiumguineense extract (plant extract) aqueous extract and $AgNO_3$ solution recorded against time of reaction reveals a clear surface Plasmon resonance (SPR) band in the range of 350nm - 700nm. This therefore, becomes apparent after 20 minutes of reaction time and progressively grows to increase in the absorbance during the period 0 - 80 at 20 minutes interval.

FITR Analysis

FTIR measurements were performed in order to identify the presence of possible biomolecules (active chemical functional groups) responsible for efficient stabilization of synthesized AgNPs. Figs. 5 – 8 respectively reveals the absorption bands for the Syzygiumguineense extract without and with 1mM, 2mM and 3mMm $AgNO_3$ solution.

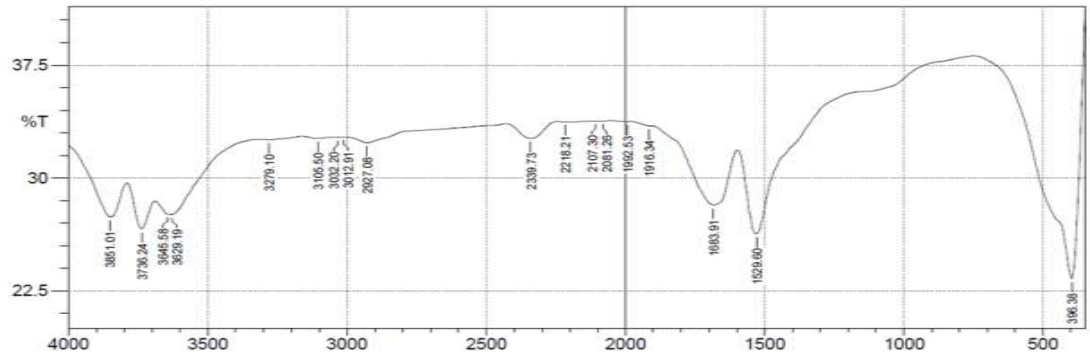

Fig. 5: FTIR absorption spectra of *Syzygium guineense* extract without $AgNO_3$ solution.

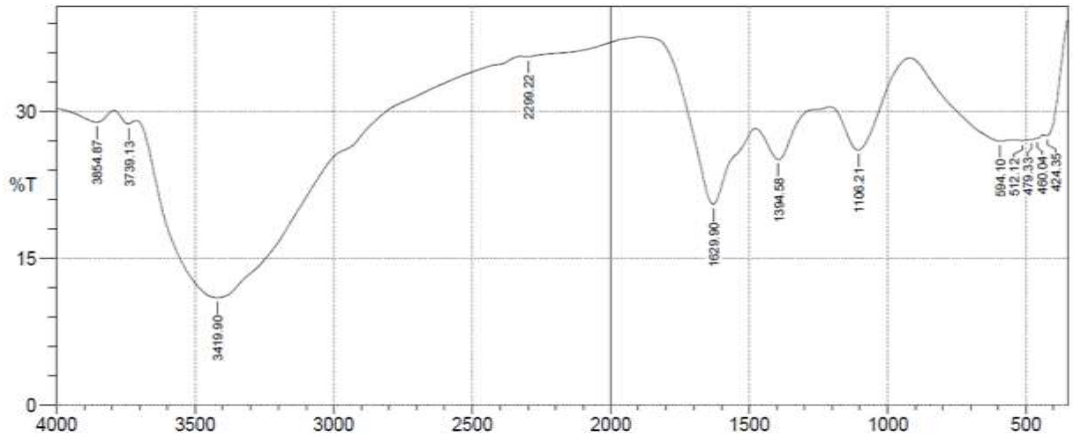

Fig. 6: FTIR absorption spectra of *Syzygium guineense* extract with 1mM $AgNO_3$ solution.

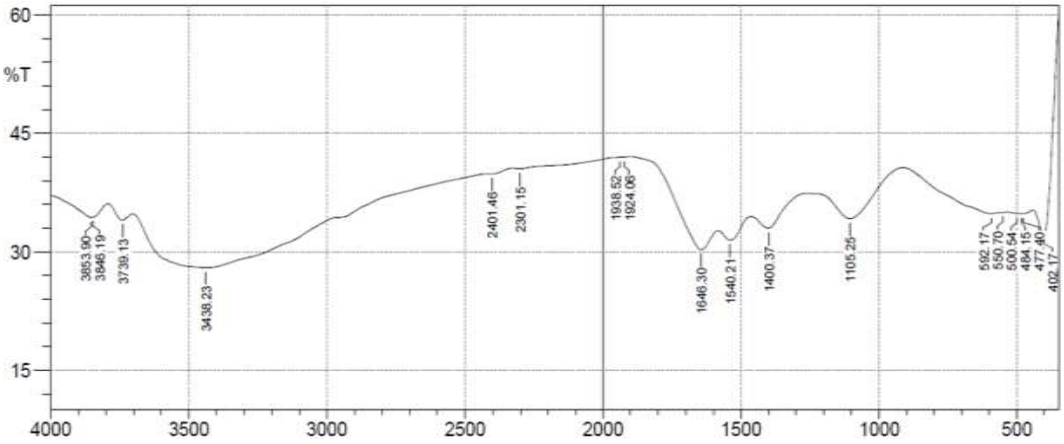

Fig. 7: FTIR absorption spectra of *Syzygium guineense* extract with 2mM $AgNO_3$ solution.

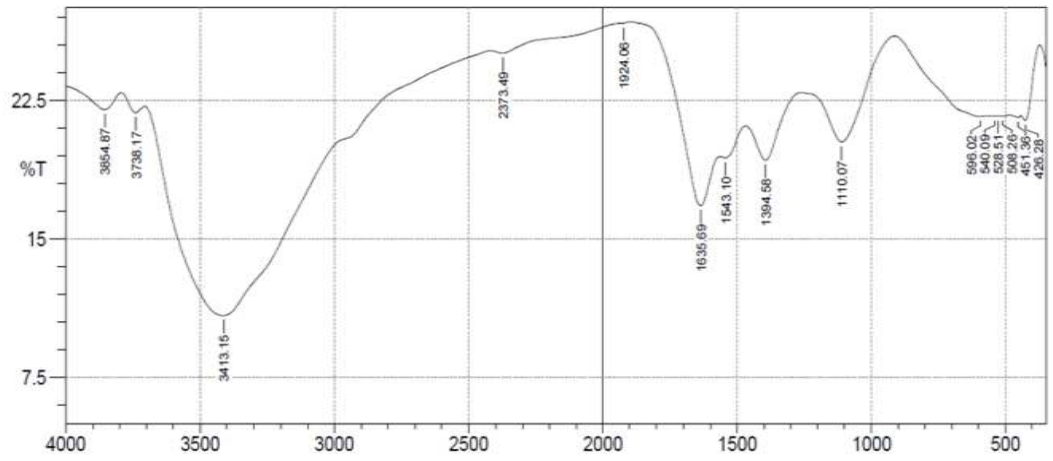

Fig. 8: FTIR absorption spectra of *Syzygium guineense* extract with 3mM $AgNO_3$ solution.

The values of the wave number corresponding to the peaks of the major infrared (IR) bands in Tables 1 - 4 reveal the presence of possible functional groups in the synthesized AgNps.

Table 1: Wave number of the peaks of the major IR bands for Raw Sample Extract

S/N	Wave Number (cm^{-1})	Bond type	Functional Group
1	396.38	C – H	Aromatic
2	1529.6	C = C	Aromatic
3	1683.91	C = C	Alkene
4	1916.34	C = C	Alkene
5	1992.53	C = C	Alkene
6	2081.26	N = C = N	Carbodiimide
7	2107.3	C ≡ C	Alkyne
8	2218.21	C ≡ N	Nitride
9	2339.73	C = N	Nitrile
10	2927.08	C – H	Alkane
11	3012.91	C – H	Alkene
12	3032.2	= C - H	Alkene
13	3105.5	N – H	Amide
14	3279.1	O – H	Alcohol
15	3629.19	N – H	Amide
16	3645.58	N – H	Amide
17	3736.24	N – H	Amide
18	3851.01	N – H	Amide

Table 2: Wave number of the peaks of the major IR bands for 1mM Sample

S/N	Wave number (cm^{-1})	Bond type	Functional Group
1	424.35	C – H	Aromatic
2	460.04	C – H	Aromatic
3	479.04	C – H	Aromatic
4	512.12	C – Cl	Alkyl Halide
5	594.1	C – Cl	Alkyl Halide
6	1106.21	C – O	2^0 Alcohol
7	1394.58	C – H	Alkane
8	1629.9	C = C	Alkene
9	2299.22	C ≡ N	Nitride
10	3419.9	O – H	Acid
11	3739.13	N – H	Amide
12	3854.87	N – H	Amide

Table 3: Wave number of the peaks of the major IR bands for 2mM Sample

S/N	Wave number (cm^{-1})	Bond type	Functional Group
1	402.17	C – H	Aromatic
2	477.4	C – H	Aromatic
3	484.15	C – H	Aromatic
4	500.54	C – Br	Alkyl Halide
5	550.7	C – Br	Alkyl Halide
6	592.17	C – Br	Alkyl Halide
7	1105.25	O – H	Alcohol
8	1400.37	C = C	Aromatic
9	1540.21	C = C	Aromatic
10	1646.3	O – H	Acid
11	1924.06	C = C	Alkene
12	1938.52	C = C	Alkene
13	2301.15	C = N	Nitrile
14	2401.46	C = N	Nitrile
15	3438.23	N – H	Amine
16	3739.13	N – H	Amide
17	3846.19	N – H	Amide
18	3853.9	N – H	Amide

Table 4: Wave number of the peaks of the major IR bands for 3mM Sample

S/N	Wave number (cm^{-1})	Bond type	Functional Group
1	426.28	C – H	Aromatic
2	451.36	C – H	Aromatic
3	508.26	C – Br	Alkyl Halide
4	528.1	C – Br	Alkyl Halide
5	540.09	C – Br	Alkyl Halide
6	596.02	C – Br	Alkyl Halide
7	1110.07	C - O	Alcohol
8	1394.58	C = C	Alkane
9	1543.1	C = C	Aromatic
10	1635.69	O – H	Acid
11	1924.06	C = C	Alkene
12	2373.49	C = N	Nitrile
13	3413.15	N – H	Amine
14	3738.17	N – H	Amide
15	3854.87	N – H	Amide

The FTIR analysis reveals the presence of active functional groups in the synthesized AgNPs. The band observed in the raw sample extract at say 1529.6 cm^{-1} and 3279.1 cm^{-1} peaks in Table 1 shows the presence of C=C and O-H functional groups respectively. The peaks were shifted when $AgNO_3$ solution was added. The change in the intensity of the peak depends on the concentration of the $AgNO_3$ solution. For example for C=C functional group, the peak shifted from 1529.6 cm^{-1} to 1629.9 cm^{-1} when 1mM of the solution was added. Further shift to 1924.06 cm^{-1} was observed with 2mM concentration of the stabilizer. This value was maintained even with increase in the concentration to 3mM as presented in Tables 2, 3 and 4 respectively. Similarly for O-H functional group, increase in peaks from 3279.1 cm^{-1} to 3419.9 cm^{-1} with addition of 1mM of $AgNO_3$ solution was observed. For 2mM and 3mM $AgNO_3$ solutions, a constant sharp decrease in the peaks was noticed increases to 1. This is in agreement with the results presented in Reddy *et al.* (2015) and Anandalkshim*et al.* (2016).

SEM Analysis

SEM analysis on the sample extract was performed to determine the morphological composition of AgNPs in terms of their shapes and sizes. The morphological structures of the AgNPs showing different sizes of nanoparticles from Syzygiumguineense sample extract for 1mM, 2mM and 3mM are shown in the SEM images with average diameter of 23.5nm to 89.3nm: Figs 9 (a), (b) and (c) respectively.

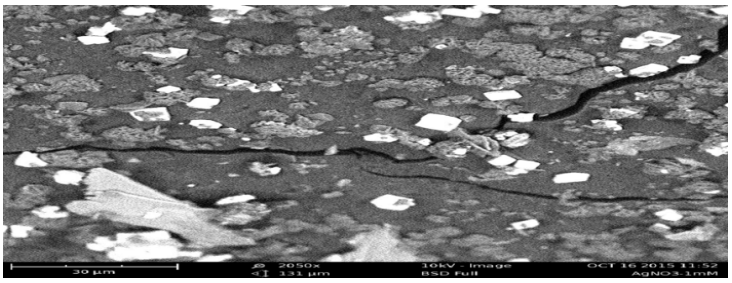

Fig. 9(a): SEM image of AgNPs synthesized form *Syzygium guineense* Extract with 1mM $AgNO_3$ solution.

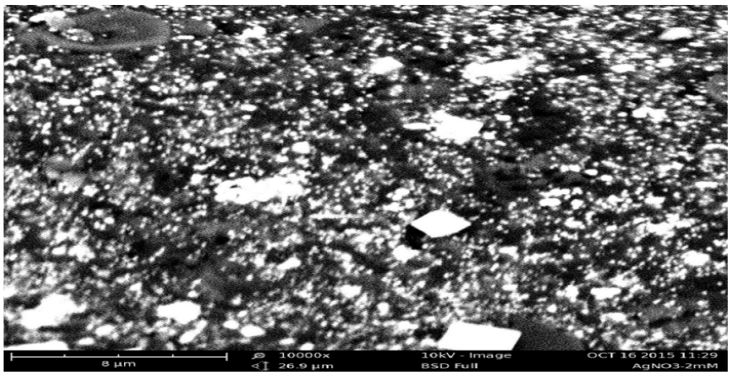

Fig. 9(b): SEM image of AgNPs synthesized form *Syzygium guineense* Extract with 2mM $AgNO_3$ solution.

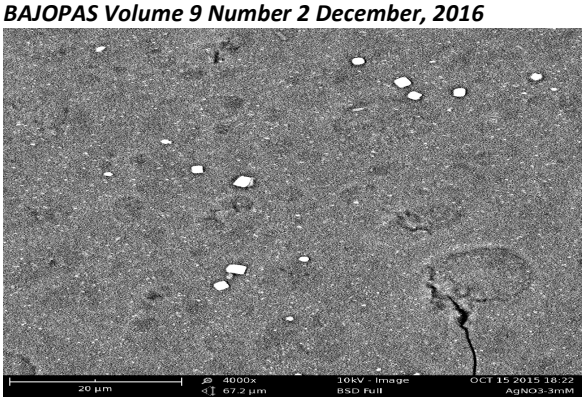

Fig. 9(c): SEM image of AgNPs synthesized form Syzygiumguineense Extract with 3mM $AgNO_3$ solution.

Figures 9(a) through9 (c) show different shapes (triangle, rectangle, tetrahedral, round surfaces, 5 or 6 diagonal, cubic, pyramid, spheres, etc)and sizes of the synthesized AgNPs for 1mM, 2mM and 3mM$AgNO_3$ solution. As it can be observed from Figure 9 (a), the shapes are mostly combination of different geometries and sizes for 1mM concentration of $AgNO_3$. As the concentration increases to 2mM, the shapes tend to become more spherical with proportional decrease in size (see Figure 9 (b)). Similarly, for 3mM concentration of $AgNO_3$,the shapes are nearly spherical with minimum surface given rise to a more stable AgNPs, which is in agreement with the result obtained in Khodashenas and Ghorban (2015). It can therefore be concluded that increase in concentration of $AgNO_3$solution (stabilizer) results in a more thermodynamically stable AgNPs.Furthermore,the analysis verifies that the particles formed are of a nano-size.

Antimicrobial Analysis of the Sample Extract

The antimicrobial activity of the AgNPs was tested against six microbe pathogens: Methicillin Resistant StaphylococusAureas, Vancomycin Resistant EntroCoci, Staphylococcus Aureas, Bacillus Sublitis, Escherichia Coli, and Pseudomonas Aerigynosa (Table 5). The results agree with other studies which have shown the efficacy of AgNPs antibacterial activity (Annamalai et al., 2013).

Table 5: Antibacterial Activity of Plant Extract and AgNPs showing Zones of Inhibition (mm)

Microorganism	Plant Extract (μgml^{-1})	NPs- 01 (μgml^{-1})	NPs -02 (μgml^{-1})	NPs-03 (μgml^{-1})	Standard Drug1 (Conc.)	Standard Drug2 (Conc.)
Methicillin Resistant Staphylococus Aureus	21	29	28	30	35	34
Vancomycin Resistant Enro Coci	0	0	0	0	0	0
Staphylococus Aureus	20	27	26	29	37	35
Bacillus Sublitis	0	0	0	0	34	32
Escherichia Coci	21	30	27	30	37	38
Pseudomonas Aeriginosa	20	29	26	32	32	31

The highest zone of inhibition was observed in Methicillin Resistant Staphylococus Aureus and Escherichia Coci for plant extract. For 1mM AgNPs, the maximum zone of inhibition was observed in Escherichia Coci. The highest zone of inhibition for 2mM AgNPs was associated Methicillin Resistant Staphylococus Aureus. Similarly, for 3mM AgNPs, the highest zone of inhibition was observed in PseudomanasAuriginosa. It can be observed that all AgNPs and plant raw extract had no effect on Vancomycin Resistant Entrococci and *Bacillus Sublitis*

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Table 6: Minimum Inhibitory Concentration (MIC) of the Plant Extract against the Microorganism.

Microorganism	Plant Extract (μgml^{-1})	NPs- 01 (μgml^{-1})	NPs -02 (μgml^{-1})	NPs-03 (μgml^{-1})	Standard Drug1 (Conc.)	Standard Drug2 (Conc.)
Methicillin Resistant Staphylococus Aureus	250	62.5	125	62.5	35	34
Vancomycin Resistant Entro Coci	0	0	0	0	0	0
Staphylococus Aureus	250	125	125	62.5	37	35
Bacillus Sublitis	0	0	0	0	34	32
Escherichia Coci	250	62.5	125	62.5	37	38
Pseudomonas Aeriginosa	250	62.5	125	62.5	32	31

From Table 6, the minimum inhibitory concentration of the raw plant extract for all the pathogenic microbes was calculated as 250µgml-1. AgNPs for 1mM $AgNO_3$ had a minimum inhibitory concentration of 62.5μgml^{-1} for Methicillin Resistant Staphylococus Aureus, Escherichia Coci and Pseudomonas Aeriginosa; and 125μgml^{-1} for Staphylococus aureus. For 2mM $AgNO_3$ sample extract, the minimum inhibitory concentration was found to be 125μgml^{-1} for Methicillin Resistant Staphylococus Aureus, Staphylococus Aureus, Escherichia coli and Pseudomonas aeriginosa. While for 3mM $AgNO_3$ sample extract, inhibitory concentration was 62.5μgml^{-1} for Methicillin Resistant Staphylococcus Aureus, Staphylococus Aureus, Escherichia Coli and Pseudomonas Aeriginosa. It can be seen that all AgNPs and plant raw extract had no effect On Vancomycin Resistant EntroCoci and Bacillus Sublitis. The images of antimicrobial activity of the plant sample extracts for varying $AgNO_3$ concentration (1mM, 2mM and 3mM) is shown in Figure 10.

Figure 11 shows the bar chart of antimicrobial activity of the raw Syzygiumguineenses, extract and with 1mM, 2mM and 3mM of $AgNO_3$ solutions exhibiting the highest zones of inhibitions in six different pathogenic microbes, while Figure 12 depicts the minimum inhibitory concentration (MIC) of the raw plant extract against the microorganisms.

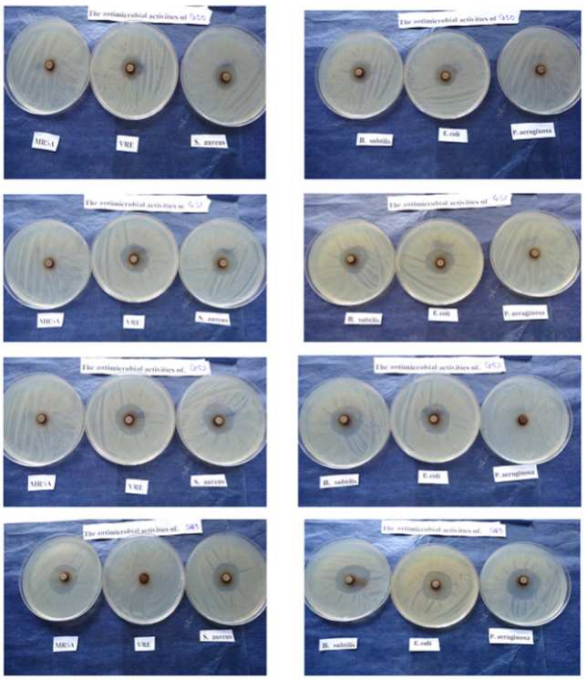

Figure 10: Antimicrobial studies of the *Syzygium guineense* sample extract with varying concentrations of AgNPs samples (1mM, 2mM and 3mM).

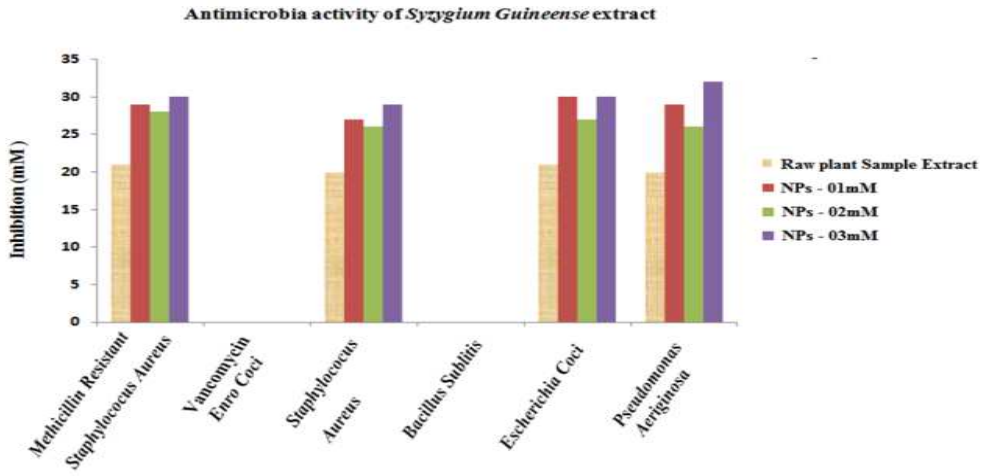

Figure 11: Microbial Activity of Raw Plant Extract and with different pathogenic microbes.

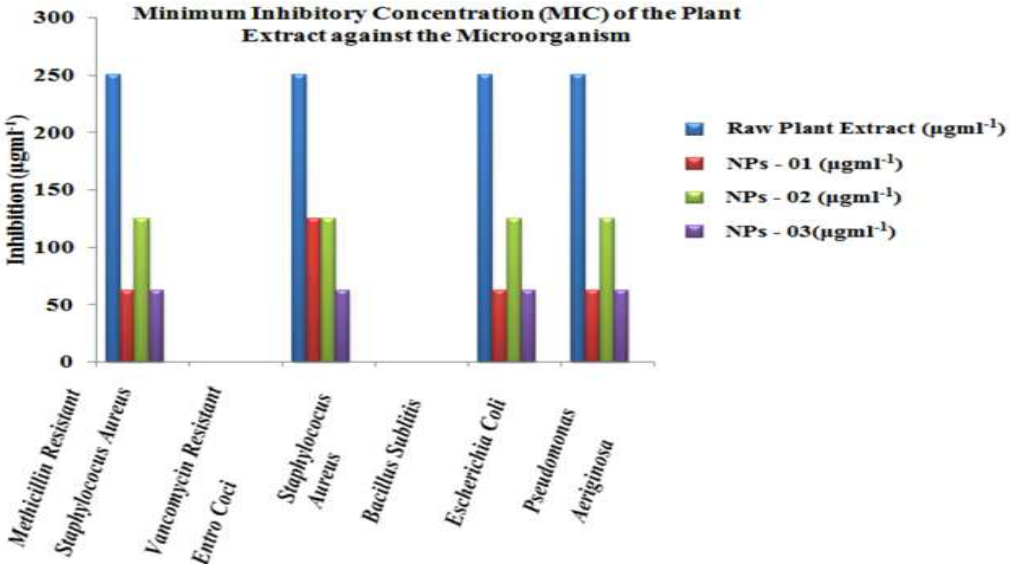

Figure 12: Minimum Inhibitory Concentration (MIC) of the Plant Extract against the Microorganism at different concentration.

CONCLUSION

A green technique to synthesize silver nanoparticles (AgNPs) using Syzygiumguineenses stem extract has been presented. In this paper, morphological compositions of AgNPs from Syzygiumguineenses stem extract were determined and characterized by UV-visible spectroscopy, FTIR and SEM. The average size of the synthesized particles was in the range of 18.5nm - 89.3nm, which clearly showed that they were on nano-scale size (1nm-100nm). For the production of nanoparticles in a large scale, the characterization is very vital and could lead to significant economic viability, as well as being environmentally friendly for treatment of some infectious diseases (e.g. cancer, skin diseases, etc), drug delivery, biomedical sensors, optoelectronics and other electronic and medical applications. The study of the effect of silver nanoparticles on human pathogens presents novel approaches for investigating a new range of its antibacterial activity.

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