https://dx.doi.org/10.4314/dujopas.v9i3b.7
ISSN (Print): 2476-8316
ISSN (Online): 2635-3490
**Dutse Journal of Pure and Applied Sciences (DUJOPAS),** *Vol. 9 No. 3b September 2023*

# Antibacterial Activities of Biologically Synthesized Silver Nanoparticles from *Alcalypha Wilkesenia* Coated with Some Antibiotics

***[1]Mansur Murtala Musa, [6]Jibril Sani Mohammed, [1]Umar Nasir Mukhtar, [2]Madu Abdulkarim Him, [3]Zandam Nuhu Danladi, [4]Idirisu Jibril Limangba and [5]Makin Tasi'u Yau**

[1]Department of Microbiology and Biotechnology,
Faculty of Science,
Federal University Duste,
Ibrahim Aliyu Bye-Pass,
P.M.B 7156,
Duste Jigawa State,
Nigeria

[2]Department of Biologiocal Sciences
Nigerian Defence Academy, Kaduna,
Nigeria

[3]Department of Science Laboratory Technology,
College of Science and Technology,
Jigawa State Polytechnic Dutse,
Nigeria

[4]Departiment of Petroleum Chemistry
Baze University
Abuja

[5]Departiment of Plant Science and Biotechnology
Federal University Dutsin-ma
Katsina
Nigeria

[6]Department of Biological Sciences
Federal University Dutse.

Email: mansurmurtala1@gmail.com

## Abstract

*Silver nanoparticles and Acalypha wilkesiana have been shown through several lines of evidence to possess antimicrobial properties. Their activities, however, have been hampered by one or more problems ranging from poor to slow absorption, cellular degradation, development of antimicrobial resistance and unwanted oxidation in the case of the Nanoparticles. Antibiotic resistance genes have also been on a constant increase recently. This research was carried out to determine the level of efficacy of the*

**Author for Correspondence*

*combined complex of antibiotics coated with biologically synthesized silver Nanoparticles compared to the orthodox antibiotics individually. The silver Nanoparticles were most active on Klebsiella. pneumoniae (21mm), equal for Escherichia coli and Staphylococcus aureus (14mm) and least for P. aeroginosa (13mm). The activity of the complex on S. aureus was notably highest with the gentamycin (17mm) complex, then ofloxacin (13mm) and finally ampicillin (12mm). That of Pseudomonas aeroginosa ranged from ofloxacin (17mm) to gentamycin (16mm) and then ampicillin (14mm), while that of Escherichia coli was from gentamycin (19mm), ofloxacin (15mm) and then ampicillin (13mm). The activity of the gentamycin (18mm) and ampicillin (18mm) complexes for Klebsiella pneumoniae were found to be equal and a bit higher than that of the ampicillin (11mm) complex. The minimal inhibitory concentration (MIC) and minimal bactericidal concentration (MBC) of the selected organisms against the antibiotic complexes carried out between 2.5mg/L to 25mg/L showed no growth in all the samples. There is a need for further research to explore the pharmacokinetics and dynamics of biologically synthesized nanoparticles.*

**Keywords:** *Acalypha wilkesiana,* Antibiotics, Bacteria, Silver nanoparticles.

## INTRODUCTION

Nanotechnology is an important field of modern research that deals with the design, synthesis, and manipulation of particles structure ranging from approximately 1-100 nm. The tremendous growth in this emerging technology has opened novel fundamental and applied frontiers, including the synthesis of nanoscale materials and the exploration or utilization of their exotic physicochemical and optoelectronic properties (Colvin *et al*., 2004; Mansur *et al*., 2005).

Silver nanoparticles have been used by mankind for about 7000 years, silver metal has been recognized to have a very potent antibacterial agent that is lethal to numerous types of microorganisms causing various infectious diseases (Chernousova and Epple, 2013). However, when a silver metal is applied on moisturized skin surface leads to low effective silver concentration since its oxidation to silver ions, which is required for the bactericidal activity, is a slow process under normal conditions (Zhang *et al*., 2008; Chernousova and Epple, 2013).

When silver ions are transformed into a metal silver nanoparticle (AgNPs) (e.g., size and shape depending on optical, electrical, and magnetic properties) that are incorporated into antimicrobial applications, biosensor materials, composite fibres, cryogenic superconducting materials, cosmetic products, and electronic components, through biological and biomimetic methods of synthesis, their toxicity is seen to decrease while their antimicrobial activities get increase markedly (Jain and Pradeep, 2005; Jain and Gauba, 2017). These characteristics make AgNPs wonderful weapons for the clinical management of microbial diseases (Karade *et al*., 2021), especially as their selectivity towards bacterial cells has been proven and no antimicrobial resistance has been so far reported (Zhang *et al*., 2008).

*Acalypha wilkesiana* is a member of the spurge family (Euphorbiaceae) belonging to the genus *Acalypha* and is commonly called copper leaf, Joseph's coat and fire dragon (Makoshi *et al*., 2016). *Acalypha wilkesiana* is a popular outdoor plant native to Fiji and nearby islands in the South Pacific but has spread to most parts of the world, especially the tropics of Africa, America and Asia.

In the northern and Southern parts of Nigeria, expressed juice or boiled decoction of the leaves of *Acalypha wilksenia* is used in traditional healthcare practice, for the management of

gastrointestinal disorders, fungal skin infections, hypertension and diabetes mellitus. The leaf poultice is used in the treatment of headaches swellings, colds and malaria (Akinyemi *et al*., 2005).

Over time man has developed traditional healing methods based on the knowledge of medicinal plants Most of the time, this information is only orally inherited and therefore in danger of being lost in favour of modern medicine (Maggassouba *et al*., 2007). However, it represents a possibility of simple and cheap treatment for the local population. In addition, it is a source of potentially important new pharmaceutical substances. According to the World health organization (W.H.O.), more than eighty percent (80 %) of the world's population relies on traditional medicine for their primary healthcare needs. Vast knowledge of how to use plants against different illnesses may be accepted to have accumulated in areas where the use of plants is still of great importance (Pezzi *et al*., 2020). There have been recorded cases of increasing antibiotic resistance by some pathogenic bacteria as a result, the used of combinations of antibiotics and antibiotic complexes have been already adopted.

This research aims to determine the antibacterial activities of biologically synthesized silver nanoparticles from *acalypha wilkesenia* coated with some antibiotics

## MATERIALS AND METHODS

### Sample Collection

Fresh leaves of *Acalypha wilkesiana* were collected from the Federal University Dutse environment and specimen samples were deposited in the herbarium of the Department of Biological Sciences, Faculty of Science, and Federal University Dutse. The plant was thus authenticated as *Acalypha wilkesiana* LUH 7536 (Copper leaf).

### Preparation and Processing of Plant

The preparation was done according to the procedure described by Kothari and Seshadri (2010). Fresh leaves of *Acalypha wilkesiana* were harvested, properly washed, rinsed in sterile distilled water, and left to air dry at room temperature for two weeks. A dried sample was pulverized using an electric blender. The powdered sample was stored in an air-tight container at room temperature until when required. Using methanol, ethanol and water as solvents, 50g of the dried leaf extracts were dissolved in 250 ml each of the solvents. The concentrated extract was obtained by drying using a rotary evaporator for the methanolic and ethanolic extracts and using a water bath at 100 °C for the water extract. The methanolic and ethanolic crude extracts obtained were reconstituted with 100% Dimethylsulphoxide (DMSO) and distilled water for hot water extract. The extract was stored in amber bottles **(**Lawal and Umar, 2020).

### Synthesis of Silver Nanoparticles (AgNPs)

For the synthesis of silver nanoparticles, 1 ml of silver nitrate and leaf extract was taken. For the reduction of $Ag^{+}$ ions, 5 ml of leaf extract was added dropwise to 5 ml of 1 ml silver nitrate solution. A distinct colour change was observed after 24 hours from a yellow to a dark brown solution at room temperature suggesting the formation of silver nanoparticles. The nanoparticles were separated from the mixture by centrifugation (at 4000 rpm for 15 mins) (NCCLS, 2000; Lawal and Umar, 2020).

**Coating of Silver Nanoparticles**

The addition of the silica shell on the AgNPs surface was performed using the seeded polymerization technique by sol-gel reaction described by Graf *et al*. (2003). Initially, the AgNPs previously described were centrifuged at 6000 rpm for 10 minutes in acetone, the supernatant was discarded and the pellet was re-dissolved in 50 mL of ethanol. Then, the solution was centrifuged again at 15000 rpm for 10 minutes. The precipitate was re-dissolved in 20 mL of ethanol and centrifuged again at 15000 rpm for 15 min. The precipitate was resuspended in an ammonia solution (4.2% [v/v] ethanol), and immediately a tetraethoxysilane (TEOS) solution (10% [v/v] ethanol) was added to the mixture under stirring. The reaction was stirred overnight, followed by centrifugation at 8000 rpm for 10 min. The precipitate was washed with ethanol and then dried to obtain an Ag@SiO2 composite (Affonso de Oliveira *et al.,* 2017).

The reaction with 3-aminopropyltriethoxysilane (APTES) was performed in two stages using the same reaction flask. Initially, it was performed the same procedure as previously described and, after overnight stirring, 10.5 µl of APTES was added to the core-shell solution. After APTES addition, the reaction was kept overnight under magnetic stirring, followed by centrifugation at 8000 rpm for 10 min. The precipitate was washed with ethanol and dried to obtain Ag@SiO2-NH2 (Affonso de Oliveira *et al.,* 2017).

Under magnetic stirring, 56.16 µl of EDC was added to an NHS solution, followed by the immediate addition of each antibiotics solution (10µg of each). Stirring was maintained for 1 hour, and then Ag@SiO2-NH2 in MES (0.1 M, pH 5) was added. The reaction was left overnight under magnetic stirring at room temperature, followed by centrifugation at 8000 rpm for 10 min. The precipitate was washed with ethanol and dried to obtain the following solutions of each antibiotic used Ag@SiO2-Ampicillin composite, Ag@SiO2-gentamycin composite and Ag@SiO2-oflaxacin composite (Affonso de Oliveira *et al.,* 2017).

**Standardization of Microbial Cell Suspension**

The cell suspension for the antimicrobial activity was standardized according to 0.5 McFarland standards as described by Mirzajani *et al.,* (2011). Using a micropipette approximately 100 µl of the reconstituted extracts at the various dilutions (100 to 12.5 mg/ml) were dropped into each well which filled them respectively to fullness.

**Antimicrobial Screening of Crude Extracts**

The plant extract was reconstituted in sterile distilled water for the hot water extraction and concentrated DMSO for both methanol and ethanol extracts to give the required dilutions of extracts which ranged from 12.5 to 100 mg/ml the crude extracts were screened for antibacterial activity using the agar well diffusion method (Kutama *et al.,* 2018). Each case will use sterile distilled water and concentrated DMSO as controls. The plates were allowed to stand for one hour on the bench to allow for proper diffusion of extracts into the medium and then incubated uprightly at 37 °C for 24 hours. Zones of inhibition were measured to the nearest millimetre (mm). All experiments were applied in triplicates.

**Determination of Minimum Inhibitory Concentration (MIC)**

Minimum inhibitory concentrations (MIC) were determined by standard agar dilution method procedures as described by Adeniyi *et al*. (2000) and Lawal *et al*. (2020) in a series of doubling extract concentrations.

**Determination of Minimum Bactericidal Concentration (MBC)**

The Minimum Bactericidal Concentration (MBC) was determined by first selecting the tubes that showed no growth during the MIC determination. One loop full from each of these tubes was subcultured onto the surface of nutrient agar and incubated for 24 hours at 37°C. The lowest concentration at which no growth was observed on the agar was noted as the MBC (Lawal and Umar, 2020).

**RESULTS**

The ethno pharmacological claims of biosynthesized silver nanoparticles and biosynthesized silver nanoparticles coated with some antibiotics (Ampicillin, Gentamycin and Ofloxacin) from *Acalypha wilkesenia* were investigated which revealed the efficacy and potency of biosynthesized silver nanoparticles and biosynthesized silver nanoparticles coated with some antibiotics (Ampicillin, Gentamycin and Ofloxacin) against bacterial isolate: zones of inhibition were determined and are represented in Tables 1and 2.

The minimum inhibitory concentration of biologically synthesized silver nanoparticles and biologically synthesized silver nanoparticles coated with antibiotics against the isolates expressed in milligrams per millilitre (mg/ml) is indicated in Table 3. The MIC values for the AgNPs and AgNPs coated with each ampicillin, gentamycin, Ofloxacin were identical for all the tested bacteria. Thus the information is represented in one table for all. The minimum bactericidal concentration of biologically synthesized silver nanoparticles and biologically synthesized silver nanoparticles coated with antibiotics against the isolates expressed in milligrams per millilitre (mg/ml) is indicated in Table 4. The MBC values ranged from 20mg/ml to 5mg/ml showing that there was no activity in biologically synthesized Silver nanoparticles coated with all the 3 antibiotics in all the isolates.

Table 1: Antimicrobial susceptibility of test isolates to Biosynthesized silver nanoparticles

| Organism | $Ag^+$ (25mg/ml) | Amp | Gen | Ofl |
|---|---|---|---|---|
| *Staphylococcus aureus* | 14 | 0.0 | 4 | 3 |
| *Pseudomonas aeruginosa* | 13 | 0.0 | 0.0 | 4 |
| *Escherichia coli* | 14 | 0.0 | 0.0 | 0.0 |
| *Klebsiella pneumonia* | 21 | 0.0 | 3 | 2 |

**NOTE**: All readings are average of triplicate experiments and are represented in millimetres (mm);
**KEY**: Ag+ = Silver nanoparticles

Table 2: Antimicrobial susceptibility of test isolates to Biosynthesized silver nanoparticles coated with antibiotics

| Organisms | Ag+(25mg/ml) | Camp(25mg/ml) | Cgen(25mg/ml) | Cofl(25mg/ml) |
|---|---|---|---|---|
| *S. aureus* | 14 | 12 | 17 | 13 |
| *P. aeruginosa* | 13 | 14 | 16 | 17 |
| *E. coli* | 14 | 13 | 19 | 15 |
| *K. pneumoniea* | 21 | 11 | 18 | 18 |

**NOTE**: All readings are average of triplicate experiments and are represented in millimetres (mm)

Table 3. The MIC values for the AgNPs and AgNPs coated with antibiotics

| **Organism** | **MIC (mg/ml)** | **Concentration of AgNPs and AgNPs with Antibiotics (mg/ml)** | | | |
|---|---|---|---|---|---|
| | | 20 | 10 | 5 | 2.5 |
| ***Staphylococcus aureus*** | 2.5-20 | - | - | - | - |
| ***Pseudomonas aeruginosa*** | 2.5-20 | - | - | - | - |
| ***Escherichia coli*** | 2.5-20 | - | - | - | - |

| | | | | | |
|---|---|---|---|---|---|
| ***Klebsiella pnemoniae*** | 2.5-20 | - | - | - | - |

**NOTE**: All readings are average of triplicate experiments; **KEY**: - No Bacterial growth; + Bacterial growth; MIC Minimum inhibitory Concentration; R resistance

Table 4 MBC of AgNPs and AgNPs coated with antibiotics against selected isolates

| **Organism** | **MBC (mg/ml)** | **Concentration of AgNPs and AgNPs with Antibiotics (mg/ml)** | | | |
|---|---|---|---|---|---|
| | | 20 | 10 | 5 | 2.5 |
| ***Staphylococcus aureus*** | 2.5-20 | - | - | - | - |
| ***Pseudomonas aeruginosa*** | 2.5-20 | - | - | - | - |
| ***Escherichia coli*** | 2.5-20 | - | - | - | - |
| ***Klebsiella pnemoniae*** | 2.5-20 | - | - | - | - |

**NOTE**: All readings are average of triplicate experiments; **KEY**: - No Bacterial growth; + Bacterial growth; MBC Minimum Bactericidal Concentration; R resistance

## DISCUSSION

Many studies have established the usefulness of medicinal plants as a great source for the isolation of active principles for drug formulation (Banso 2000; El-Mahmud 2007; Falodun *et al*., 2006). Several species of the genus *Acalypha* have been studied and it has been demonstrated that they have antioxidant, wound healing, post-coital antifertility, neutralization of venom, antibacterial, antifungal and antitrypanosomal activities (Parez *et al*., 2006, Marwah *et al*., 2007 and Shurwalker *et al*., 2004). Similarly, this study supports the antibacterial activities of biologically synthesized silver nanoparticles coated antibiotics from *wilkesiana* as a broad-spectrum antimicrobial agent since it inhibited the growth of Gram-positive (*S. aureus*) and gram-negative bacteria *(E. coli,* K. *pneumoniae and P. aeruginosa*) (Shurwalker *et al*., 2004; Marwah *et al*., 2007).

The silver nanoparticles synthesized from *Acalypha wilkesiana* leaf extract act as a reducing agent and the antibacterial activities of the extract were increased in all the organisms, most notably in *K. pneumoniae* which showed an increase of up to 16.5% in activity from the 8mm zone of inhibition at 25mg/ml to 21mm. This was followed by E. *coli* with a 55.5% increase from 9mm to 14mm at 25mg/ml. *S. aureus* had a 27.3%increase from 11mm to 14mm, meanwhile P. *aeruginosa* showed a 44.4% increase from 9mm to 13mm when the synthesized extract was used. This is similar to the report by Sajeshkumar *et al*., 2015.

The biologically synthesized silver nanoparticles from the plant extract were tested against the organisms that were standardized to 0.5 McFarland at a concentration of 25mg/ml. the biologically synthesized silver nanoparticle showed a great character in increasing the general activity of the extract. This suggested that there is an increase in the uptake and delivery of the extract into the microbial cells, this also supported the report of Adekola *et al*., 2019 which revealed that biologically synthesized silver nanoparticles inhibited the growth of many bacterial isolates.

Table 2 also shows the activity of the biosynthesized silver nanoparticles of the extracts coated with each ampicillin (Camp), gentamycin (Cgen) and ofloxacin (Cofl). The organisms showed varying sensitivity to the Camp, with an increase of 7.6% from the 13mm in the biosynthesized

silver nanoparticles coated without the antibiotics to 14mm with the antibiotic and 3mm with only the antibiotic in P. *aeruginosa*, there were however lessened activity of the biosynthesized silver nanoparticles coated with the ampicillin compared to those without the antibiotics. For *S. aureus* there was a 14.2% decrease from 14mm to 12mm, a 7.14% decrease from 14 to 13mm in E. coli and most notably from 21mm to 11mm in K. *pneumoniae*, representing a 47.6% decrease in activity (Jene *et al*., 2013). The activity of the Cgen was however better than that of the Camp, there were significant increases in all the isolates except for K. *pneumoniae* which recorded a 14.2% decrease from 21mm to 18mm. For *S. aureus* the increase was 21% from 14mm to 17mm and 35.7% from 14mm to 19mm for E. *coli*. The effect on *P. aeruginosa* was also quite pronounced with an increase in inhibition diameter from 13mm to 16mm for the Cgen, representing a 23% increase. For the Cofl there was a lessened activity against *S. aureus* and K. *pneumoniae* with a decrease from 14mm to 13mm and 21mm to 18mm, representing a 7.1% and 14.3% decrease respectively supported by (Kaviya *et al.,* 2011).

The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of biologically synthesized silver nanoparticles and biologically synthesized silver nanoparticles coated with antibiotics against the isolates expressed in milligram per millilitre (mg/ml) is indicated in Tables 3a, b and c and tables 4a, b, c and d above. The MIC value ranged from 20mg/ml to 5mg/ml showing that there was no activity in biologically synthesized Silver nanoparticles coated with all the 3 antibiotics in all the isolates. Sajeshkumar *et al*., (2015) whose report that the Antibacterial property of S. *aureus,* P. *aeruginosa,* E. *coli* and K. *pneumoniea* was increased by the use of nanoparticles when compared to antibiotics gentamycin, ofloxacin and ampicillin. For E. *coli*, antibacterial activity was greater when the antibiotic Carbenicillin was used compared to silver nanoparticles. For Staphylococcus species and Bacillus species antibacterial activity are in the same range when both nanoparticles and antibiotics were used separately (Sajeshkumar *et al*., 2015).
This resistant nature was also seen in the organism's response to the test antibiotics. Also, this may be due to their ability to produce extracellular enzymes that help such organisms to degrade and metabolize substrate such that the extract becomes a source of food to the bacteria instead of inhibiting their growth after they have been rendered nontoxic due to degradation (Tortora *et al*., 2002). The MIC of this study has the same result as MBC (Tortora *et al*., 2002). The MIC and MBC of the biologically synthesized silver nanoparticles and biologically synthesized coated with gentamycin, ampicillin and ofloxacin there were susceptible to all the isolates at all the concentrations of 20mg/ml, 10mg/ml, 5mg/ml and 2.5mg/ml. the MIC is equivalent to the MBC.

The result also showed that there was a disparity between the biologically synthesized silver nanoparticles and biologically synthesized coated with gentamycin, ampicillin and ofloxacin and standard antibiotics as the former inhibited the growth of organisms (S. *aureus,* P. *aeruginosa,* E. *coli* and K. *pneumoniea*) that some of the concentrations and standard antibiotics failed to inhibit. The disparity between the activities of the extracts and the standard antimicrobial drug may be due to the mixtures of bioactive compounds present in the extract or its fractions compared to the pure compound contained in the standard antibiotics (Gatsing *et al*., 2010). This demonstration of activity against such test bacteria may form the scientific bases for the local dependence on this plant in the treatment of various ailments.

## CONCLUSION

The demonstration of activities against both gram-negative and gram-positive bacteria is an indication that the plant can be a source of bioactive substances that could be of a broad spectrum of activity.

The search for new drugs to counter the challenges posed by resistant strains of bacteria might have started yielding results as the investigation of this plant and biologically synthesized silver nanoparticles has demonstrated enormous therapeutic potential. It can serve the desired purpose with lesser side effects that are often associated with synthetic antimicrobial agents. Plants and biologically synthesized silver nanoparticles.

It is important to scientifically investigate those plants which have been used in traditional medicines as potential sources of novel antimicrobial compounds. . Also, the pharmaceutical industries should use these as an inexhaustible source of natural drugs that may be employed in combating ailments and inconveniences resulting from microbial attacks

**REFERENCES**

Adekola, F. A.; Dada, Fehintoluwa, E.; Adelani-Akande, A. T.; (2019). Silver nanoparticle synthesis by Acalypha wilkesiana extract: phytochemical screening, characterization, influence of operational parameters, and preliminary antibacterial testing. *Heliyon*, **5**(10): 02517

Adeniyi, B. A., Fong, H. S., Pezzito, J. M., Luyengi, L. and Odelola, H. A. (2000). Antibacterial activity of diospyrin, isodiospyrin and bisisodiospyrin from *Diospyros piscatorial* (Gurke) (Ebenaceae). *Phytotherapy Research,* **14**: 112 – 117.

Affonso de Oliveira, J.F., Saito, A., Bido, A.T., Kobarg, J., Hubert Karl Stassen, H.K., and Cardoso, M.B. (2017) Defeating Bacterial Resistance and Preventing Mammalian Cells Toxicity Through Rational Design of Antibiotic-Functionalized Nanoparticles. *Scientific Reports,* **7:** 1326.

Akinyemi, K. O., Oladapo, O., Okwara, C. E., Ibe, C. C. and Fasure, K. A. (2005). Screening of crude extracts of six medicinal plants used in South-West Nigerian unorthodox medicine for antimethicillin resistant *Staphylococcus aureus* activity. *BMC Complement Alternat Med,* **5**: 6.

Banso, A. and Mann, A. (2000). Antimicrobial alkaloid fraction from Commiphora *africana*. *J.Pharm. Biores*., **3**(2): 98-102.

Chernousova, S., & Epple, M. (2013). Silver as an antibacterial agent: ion, nanoparticle, and metal. *Angewandte Chemie International Edition*, **52**(6), 1636-1653.

Colvin, V.L.; Schlamp, M.C. and Alivisatos, A. (2004). Light-emitting diodes made from cadmium selenide nanocrystals and a semiconducting polymer. *Nature,* **370**: 354-357.

El-Mahmood, A. M., and Ameh, J. M., (2007) In vitro antibacterial activity of Parkia biglobosa (Jacq) root bark extract against some microorganisms associated with urinary tract infections. *African journal of Biotechnology*, **6**(11):1272-1275.

Falodun, A., Okunrobo, L. O. and Uzoamaka, N., (2006). Phytochemical screening and anti-inflammatory evaluation of methanolic and aqueous extracts of Euphobia heterophylla Linn (Euphobiaceae). Afr. J. *Biotechnol.* ; **5**(6):529-531.

Gatsing, D., Nkeugoauapi, C. N., Nkah, B. N., Kuiate, J. R., Tchouanguep, F. M.; (2010); Int, *J. Pharmacol*. (2010), **6:**173-182.

Graf, C., Vossen, D. L. J., Imhof, A. and van Blaaderen, A. (2003). A General Method To Coat Colloidal Particles with Silica. *Langmuir*, **19:** 6693–6700

Jain, J., and Gauba, P. (2017). Heavy metal toxicity-implications on metabolism and health. *Int J Pharma* Bio Sci, **8**(4): 452-460.

Jain, P. and Pradeep, T. (2005). Potential of silver nanoparticle-coated polyurethane foam as an antibacterial water filter. *Biotechnol Bioeng*, **90:** 59-63.

Jena, J., Pradhan, N., Dash, B. P., Sukla, L. B. and Panda, P. K. (2013). Biosynthesis and characterization of silver nanoparticles using microalga Chlorococcum humicola and its antibacterial activity. *Int J Nanomater Biostruct*, **3**(1): 1-8.

Karade, V. C., Patil, R. B., Parit, S. B., Kim, J. H., Chougale, A. D. and Dawkar, V. V. (2021). Insights into shape-based silver nanoparticles: a weapon to cope with pathogenic attacks. *ACS Sustainable Chemistry & Engineering*, **9(**37): 12476-12507.

Kaviya, S., Santhanalakshmi, J., and Viswanathan, B. (2011). Green synthesis of silver nanoparticles using Polyalthia longifolia leaf extract along with D-sorbitol: study of antibacterial activity. *Journal of nanotechnology*, **2011**: 1.

Kothari, V., and Seshadri, S. (2010). In vitro antibacterial activity in seed extracts of Manilkara zapota, Anona squamosa, and Tamarindus indica. *Biological research*, **43**(2): 165-168

Kutama, R. M., Abdulkadir, S., Kwalli, S. A., & Chiroma, G. (2018). Phytochemical compositions in some Nigerian medicinal plants and their pharmacological properties: A review. *International Journal of Anesthesia and Clinical Medicine*, **6**(1): 15.

Lawal D. and Umar A. (2020) antibacterial efficacy of some standard antibiotics discs coated with biologically synthesized silver nanoparticles from cassia occidentalis leaves. *Journal of Medicine and Health Research,* **5**(1): 47-53.

Maggassouba, F. B., Diallo, A., Kouyate. M. Mara, F. and Bangoura. O. (2007). Ethno botanical survey and antibacterial activity of some plants used in Guinean traditional medicine. *Journal of Ethnopharmacology*. **114:** 44-53.

Makoshi, M. S., Oladipo, O. O., Gotep, J. G., Forcados, G. E. and Shu, M. L. (2016).Safety evaluation of *Acalypha wilkesiana* in albino rats and BHK-21 cell line.*Comparative Clinical Pathology* **25**:1618-5641.

Mansur, H. S.; Grieser, F.; Marychurch, M. S.; Biggs, S.; Urquhart, R. S. and Furlong, D. (2005). Photoelectrochemical properties of 'q-state' cds particles in arachidic acid langmuirblodgett films. *J Chem Soc Faraday Trans*, **91**:665-672.

Marwah, R. G., Fatope, M. O., Mahrooqi, R. A., Varma, G. B., Abadi, H. A. and Al- Burtamani, S. K. S. (2007). Antioxidant capacity of some edible and wound healing plants in Oman. *Food Chem*, **101**:465-470

Mirzajani, Fateme, Alireza Ghassempour, Atousa Aliahmadi, and Mohammad Ali Esmaeili. (2011). Antibacterial effect of silver nanoparticles on Staphylococcus aureus. *Research in microbiology,* **162**(5): 542-549.

NCCLS (2000). Performance standards for antibacterial disk susceptibility tests: Approval standard M2-A7 7th edition. Pennsylvania: *Clinical and Laboratory Standards Institute*.

Oladunmoye, M. K. (2006) Comparative evaluation of Antimicrobial Activities and Phytochemical Screening of two varieties of *Acalypha Wilkesiana*. *Trends Appl. Sci. Res***, 1**:538-541.

Perez Gutierrez, R. M., and Vargas, S. R. (2006) Evaluation of the wound healing properties of *Acalypha langiana* in diabetic rats. *Fitoterapia*, **77**:286-289.

Pezzi, L., Diallo, M., Rosa-Freitas, M. G., Vega-Rua, A., Ng, L. F. P., Boyer, S. and Siqueira, A. M. (2020). GloPID-R report on chikungunya, o'nyong-nyong and Mayaro virus, part 5: *Entomological aspects. Antiviral research*, **174**: 104670.

Sajeshkumar, N. K., Prem Jose Vazhacharickal, Jiby John Mathew, and Anupa Sebastin. (2015). Synthesis of silver nanoparticles from curry leaf (Murraya koenigii) extract and its antibacterial activity. *CIB Tech J Pharm Sci,* **4**: 15-25

Shirwaikar, A. Rajendran, K., Bodla, R. and Kumar, C.D. (2004). Neutralization potential of Viper *Russelli russelli* (Russell's viper) venom by ethanol leaf extract of *Acalypha indica*. *J. Ethnopharmacol*. **94**:267-273.

Tortora, J. G., Funke, R. B., and Case, L. C. (2002) *Microbiology: An Introduction*. 7th ed., Person Education, Inc. Publishing; 912.

Zhang, Y., Peng, H., Huang, W., Zhou, Y. and Yan, D. (2008). Facile preparation and characterization of highly antimicrobial colloid Ag or Au nanoparticles. *J Colloid Interface Sci*, **325:**.371-376.