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EFFECT OF BIOSYNTHESIZED SILVER NANOPARTICLES ON THE DEVELOPMENT OF *ANOPHELES GAMBIAE* SENSU STRICTO

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Abstract

Female Anopheles gambiae sensu stricto, is a vector of *Plasmodium falciparum* accountable for malaria infection in man. This investigation focused on how the developmental stages of the vector can be delayed using Silver Nanoparticles (AgNPs) synthesized with the leaf extracts of *Moringa oleifera* and *Andrographis paniculata* as well as the cobweb extract of *Parasteatoda tepidariorum*. AgNPs synthesized by standard methods were applied at different concentrations on larvae and pupae of *A. gambiae*. Deionized water that does not contain any extract was used as the control. Observations of larvae and pupae developments and mortality at different concentrations were recorded. Data gotten were analyzed using analysis of variance (ANOVA) to identify significance ($p \leq 0.05$) between the means of larvae and pupae still active at different concentrations of AgNPs, after the control had developed to the succeeding stage. Ultra-violet spectra of AgNPs showed a peak at 435.00 nm for *M. oleifera*; 413.5 nm for *P. tepidariorum*; and 424 nm for *A. paniculata*-mediated particles. Development of more than 50 % larvae to pupae were prevented in 20 ml of different concentrations ($\mu\text{g/ml}$) of 42.5 and 21.25 of *M. oleifera* extract-AgNPs; 42.5, 21.25 and 10.63 of *P. tepidariorum* extract-AgNPs; and 10.63 of *A. paniculata* extract-AgNPs, when control had developed to pupae. All inhibited larvae were still alive. Synthesized AgNPs of *P. tepidariorum* was the most active at inhibiting development closely accompanied by *M. oleifera*, and then *A. paniculata*. The activities of the silver nanoparticles at inhibiting the development of larval and pupa stages of *A. gambiae* showed that the particles could be deployed for the control of mosquitoes.

Keywords: King of bitters, Metamorphosis, *Moringa oleifera*, Nanoparticles, Spider cobweb

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Introduction

Mosquito is the most important single group of arthropod of public health importance; therefore its control is equally necessary in order to successfully curtail the spread of mosquito-borne diseases thereby improving ecological and public health (Rapley *et al.*,

2009). Naturally, there are fishes that prey on mosquito larvae; such fishes include *Gambusia patruelis* and *Hemichromis* sp. These fishes feed greatly on diverse developing stages of mosquito and help decrease the occurrence of malaria diseases

(Adesulu and Syndeham, 2007). In the hunt for biological method of controlling malaria vector, eleven larvivorous fish species native to Ethiopia were identified by Fletcher *et al.* (1993); these include five species not previously reported in the country. Verifiable evidences abound that under controlled laboratory conditions the fishes fed seriously on the larvae of *Anopheles gambiae*.

Recently, the application of synthetic insecticides in mosquito control programs has been restricted due to lack of original insecticide, elevated cost of man-made insecticides, and concerns on ecological sustainability. Insecticide's detrimental effect on human health and other non-target populations, their non-biodegradable nature, high rate of biological magnification through ecosystem and increasing insecticidal resistance on a global scale (Brown, 1986) are reasons why alternative sources of control was desirable.

The environmental protection act in 1969 included several rules and regulations to check the application of chemical control agent in nature (Bhatt *et al.*, 2009). This in turns prompt scientists to look for substitute methods ranging from the provision of or promoting the adoption of effective and transparent mosquito management tactics with focus on public education, monitoring and surveillance, source reduction and environmentally friendly least toxic larval control (Bhatt *et al.*, 2009). In view of these, the application of eco-friendly alternatives such as biological control of vectors has been the central focus of the control programme as a substitute to chemical insecticides (Bhatt *et al.*, 2009).

Nanotechnology is one of the most promising modern methods for pest control in recent years (Rouhani *et al.*, 2012). Understanding the biological processes at

nanoscale level is a strong driving force in the development of nanotechnology (Sareen *et al.*, 2012). This technology is a new method of environmental remediation that could provide a convenient advancement to some of the most inspiring environmental clean-up problems (Chinnamuthu and Murugesu, 2009). AgNPs have its chief applications in the field of biology. Stable AgNPs which have been synthesized by biological method of reduction have come to be one of the wildest growing materials due to their distinctive physical, chemical and biological properties, small size and high specific surface area (Sareen *et al.*, 2012). Attention is given to biological synthesis of nanoparticles due to a rising need to progress eco-friendly technologies in material synthesis (Owolade *et al.*, 2008). Some plant species have been exploited in this regard. The use of environmentally non-threatening materials such as AgNPs offers numerous benefits of eco-friendliness and compatibility for larvicidal application. In recent years, the green synthesis of nanoparticles using plant extracts has emerged as a feasible alternative to traditional chemical procedures and physical methods. In this work, extracts of leaves of *Moringa oleifera*, *Andrographis paniculata* and cobweb extract of *Parasteatoda tepidariorum* were employed to synthesize AgNPs and evaluated for the control of larva and pupa of *Anopheles gambiae*.

Materials and Methods

Collection of Leaves of *Moringa oleifera* and *Andrographis paniculata*

The leaves of *M. oleifera* were collected at Apete Ibadan, Oyo state (Latitude 01°, 13.165' N, Longitude 05°57.500' E), while that of *A. paniculata* were collected along Stadium road Ogbomoso, Oyo state (Latitude 08°, 10' N, Longitude 4°, 16' E),

excluding leaves with visual injury. The leaves were washed gently to remove particles present on the surfaces. The leaves were air dried at room temperature (30 ± 2 °C). The dried leaves were ground to fine powder by using mortar and pestle, sieved (0.5 mm pore size sieve) and kept in a sealed glass container.

Collection of Spider Cobweb (*Parasteatoda tepidariorum*)

The cobwebs of spider were collected from a residential apartment in Ogbomoso, Oyo State by the use of a new clean broom, and taken to the laboratory. These webs were rinsed with distilled water to remove the extraneous materials and dusts. The washed spider webs were air dried at room temperature (30 ± 2 °C) and kept in air tight container for further use.

Preparation of Extracts

Phytochemicals from the leaves were extracted by addition of 50 g of the extract powder into 1 liter of distilled water (WHO, 1996). The mixture was stirred for 45 min by using magnetic stirrer, and stored for three days, for complete extraction of plant material, after which the material was strained and the remaining solid was hand squeezed with glove to remove all the remaining liquid. The obtained extract was clarified by filtration through Whatman number 1 filter paper, and rotary evaporator was used to concentrate the extract at 50 °C. In the case of preparation of the cobweb extract of *Parasteatoda tepidariorum*, the methods of Lateef *et al.* (2016a, b) were employed for the cobweb hydrolysis.

Green Synthesis and Characterization of AgNPs

The green synthesis of AgNPs was carried out using the extracted phytochemicals. One

ml of the leaf extract was added to a reaction vessel containing 30 ml of 1 mM silver nitrate (AgNO_3) solution for the reduction of silver ion. The reaction was carried out in static condition at room temperature of (30 ± 2 °C) for 2 h. AgNP_s formation was monitored by visual observation of change of colour, measurement of absorbance spectrum of the reaction mixture using UV-visible spectrophotometer (Cecil USA), and Fourier transform infrared spectroscopy (FTIR).

Preparation of Mosquito Larvae and Pupae

Isolation of *A. gambiae* eggs

Ten containers (34 cm × 21 cm × 5 cm) were used; each was filled with 1000 ml of deionized water. A little sand and leaf were spread on the surface of the water to attract mosquito; they were placed in different corners of living premises. In 5 days, there were presence of eggs in the water and these were observed with naked eyes. The eggs of *A. gambiae* were identified by single appearance on the water surface and their canoe like shape with a pair of float on either side (Impoinvil *et al.*, 2007). They were then isolated with fine brush.

Preparation of Mosquito Larvae and Pupae

The isolated eggs were put into ten containers of size 34 cm × 21 cm × 5 cm filled with 1000 ml of deionized water each, and allowed to hatch into larvae. *A. gambiae* developed through four larval sizes of instars before pupating (Foster and Walker, 2009). Larvae were very small in the first instar and increased in size until they reached 5-6 mm by the completion of the fourth instar because the larvae stage is a feeding period, through feeding on organic

matter and algae (Garros *et al.*, 2008). In this study, all the instar stages were regarded as larva and were fed in the laboratory with a mixture of ground rat chow and brewer's yeast (1:1 w/w) daily under standard rearing conditions which were maintained at ~28 °C, 80 % humidity, and photoperiod of 12 h. This procedure was repeated and the larvae were left to develop into pupae. All had developed into pupae within 4-5 days. The identification of *A. gambiae* larvae was done by the method of Foster and Walker (2009). The pupae were identified using the method of Huang *et al.* (2006).

Larval and Pupae Bioassay

Treatment doses were determined by the method of Lateef *et al.* (2015). The Five concentrations used (136, 85, 42.5, 21.25 and 10.63 µg/ml), were prepared from the stock solution by serial dilution method and 20 ml of each concentration was used. Each concentration from the synthesized AgNPs were dispensed in test tubes, indicating the concentration and the extract, after which a plastic pipette was used to pick 10 larvae from the container and transferred them into the various labeled test tubes. The actions of each concentration on the larvae were observed at intervals of 12 h. Mosquito net was used to cover the exit of the test tube and held tight with paper tape to avoid escape of the adult mosquito into the environment if it eventually developed. This procedure was also employed for the introduction of the pupae.

A control experiment was set up using deionized water. Immediately after the introduction of the larvae or pupae into the various concentrations of the synthesized AgNPs, the same numbers of larvae or pupae were also introduced into deionized water in two separate test tubes and the development of the larvae and pupae were monitored at interval of 12 h.

The effect of each concentration of the synthesized AgNPs was monitored on the larvae and pupae at 12 h-intervals for three weeks, because the development of *Anopheles gambiae* from egg to adult is normally within 14 days.

The experiments involved different concentrations of the synthesized AgNPs, for each developmental stage. Each experiment was replicated ten times. The concentration having the highest frequency out of the ten replicates, at which inhibition occurred while the control has developed to the next stage, was recorded as the effective concentration.

The percentage mortality of larvae was calculated thus (Abbott, 1925):

$$\text{Percentage mortality} = \frac{\text{number of larvae}}{\text{number of larvae introduced}} \times 100$$

Lethal dose that killed 50 % of the population of larvae or pupae at different concentrations of the extracts were determined by using Microsoft office Excel.

Statistical Analysis

Analysis of variance was used to compare the means of the inhibited larvae in synthesized AgNPs of the three extracts to detect any significant difference at 95 % confidence interval. This was also repeated for pupae.

Results

Silver Nanoparticles Synthesized using Different Extracts

Moringa oleifera

The observed results indicated the formation of nanoparticles by the characteristic color transformation of the colloids that turned dark brown. The surface plasmon resonance

that was displayed in the UV-Vis absorption spectra of AgNPs synthesized by the extracts of *M. oleifera*, *A. paniculata* and cobweb of *P. tepidariorum* are as shown in Figure 1, while the FTIR absorption spectra are shown in Figures 2-4, respectively.

Figure 1: UV-vis spectra of AgNPs biosynthesized by the extracts of *M. oleifera*, *A. paniculata* and cobweb with SPR obtained at 435, 424 and 413 nm, respectively

The Survival of Larvae on Exposure to AgNPs

The results revealed different numbers of surviving larvae at different concentrations of synthesized AgNPs at 96 h (Table 1). The statistical result using analysis of variance (ANOVA) showed that, there were significant differences in the means of larvae alive at 96 h between the leaf extract of *M. oleifera*, *A. paniculata*, and spider cobweb ($p = 0.002$ at 95 % confidence interval).

Further analysis showed that there was significant difference between the means of larvae alive in (i) synthesized-AgNPs of *M. oleifera* and means of larvae alive in *A. paniculata* synthesized-AgNPs ($p = 0.002$ at 95 % confidence interval); (ii) means of larvae alive in synthesized-AgNPs of *M.*

oleifera and means of larvae alive in synthesized AgNPs of spider cobweb ($p = 0.003$); and (iii) means of larvae alive in spider cobweb synthesized-AgNPs and *A. paniculata* synthesized-AgNPs at $p = 0.05$.

The Percentage Survival of Pupae at 144h in Varying Concentrations of Synthesized AgNPs

The result revealed different percentages of survival of pupae at different concentrations of synthesized AgNPs of *M. oleifera*, *A. paniculata* and spider cobweb at 144 h (Table 2). Statistically, there were significant differences in the means of pupae alive at 144 h between the leaf extract of *M. oleifera*, *A. paniculata* and spider cobweb ($p = 0.001$ at 95 % confidence interval). Further analysis revealed significant difference between (i) the means of pupae alive in synthesized AgNPs of *M. oleifera* and means of pupae alive in *A. paniculata* synthesized-AgNPs ($p = 0.004$ at 95 % confidence interval); (ii) *M. oleifera* synthesized-AgNPs and synthesized AgNPs of spider cobweb ($p = 0.042$); and (iii) spider cobweb synthesized-AgNPs and *A. paniculata* synthesized-AgNPs ($p = 0.001$).

Discussion

AgNPs Synthesized using *M. oleifera* Extract

The characteristic colour change together with the surface plasmon resonance peak that was displayed in the UV-vis absorption spectrum through biological reduction method with an absorption peak at 435.00 nm. This result fell within the reported range of 391-460 for AgNPs. The FTIR displayed characteristics peaks that are indicators of successful synthesis. The band 3285 cm^{-1} represents the O-H stretch, 1956 cm^{-1} band

represents the aliphatic C = O bond due to aliphatic ketones or esters.

Figure 2: FTIR spectrum of AgNPs synthesized by *M. oleifera* extract

Figure 3: FTIR spectrum of AgNPs synthesized using *A. paniculata* extract

Figure 4: FTIR spectrum of AgNPs synthesized by spider cobweb extract

Table 1: Percentage survival of larvae at 96 h in different concentrations of synthesized AgNPs

Hindered larvae ($\mu\text{g/ml}$)	136.00	85.00	42.50	21.25	10.63	LD ₅₀
<i>M. oleifera</i>	0	30	70	60	20	4.5
Spider web	0	30	60	70	100	71.35
<i>A. paniculata</i>	0	0	0	40	60	2.27
Larvae mortality						
<i>M. oleifera</i>	100	70	30	30	80	
Spider web	100	70	40	30	0	
<i>A. paniculata</i>	100	100	100	60	40	

Table 2: Percentage survival of pupae at 144 h in varying concentration of synthesized AgNPs

Hindered pupae ($\mu\text{g/ml}$)	136.00	85.00	42.50	21.25	10.63	LD ₅₀
<i>M. oleifera</i>	0	50	60	70	60	2.63
Spider cobweb	10	60	70	40	100	71.35
<i>A. paniculata</i>	0	0	0	20	70	2.29
Pupae mortality						
<i>M. oleifera</i>	100	50	20	10	40	
Spider web	90	40	30	20	0	
<i>A. paniculata</i>	100	100	100	80	30	

However, in any sample where hydrogen bond occurs, the number and strength of intermolecular interaction varies greatly within the sample, causing bond in the sample to be particularly broad (Coates, 2000). Most of the frequencies are group frequencies which is the presence or absence of a certain functional group in the sample.

AgNPs Synthesized using the Extract of *A. paniculata*

The absorption peaked at the wavelength of 424.00 nm. The bands of 3292 and 1636 cm^{-1} in the FTIR spectrum refer to the bonding vibration of the N–H bond of amines, and C=C stretch of alkenes or C=O stretch of amides, respectively (Shankar *et al.*, 2014).

AgNPs Synthesized using Extract of Spider Cobweb

The characteristic colour change, with the absorption that peaked at 413.5 nm fell within the reported range of 391–460 for AgNPs. The bands 3277 and 1635 cm^{-1} recorded in the FTIR are attributed to the bonding vibration of the amides (N–H group) of protein and the deformation atoms, respectively indicating protein as the capping and stabilizing biomolecules in the

synthesis of AgNPs as reported by Shankar *et al.* (2014). Thus, the presence of protein in the cobweb extract was responsible for the capping and stabilization of the AgNPs.

Bioactivities of Synthesized Nanoparticles on Metamorphosis in *A. gambiae*

The inhibiting effects of synthesized AgNPs of the three extracts on the larvae at different concentrations were shown in Table 1. While in the development from pupa to adult, the effects were shown in Table 2. The control experiment showed all larvae had metamorphosed to pupae at 96 h and pupa to adult at 144 h. These hours were taken as the time expected for all larvae in the test experiments to metamorphose to pupae (96 h), and pupae to adult (144 h).

These results showed that the inhibiting properties were concentration dependent and the spider cobweb synthesized-AgNPs was the most effective followed by *M. oleifera*-AgNPs, and the least active was *A. paniculata*-AgNPs in inhibiting the development of the larvae to pupa and pupa to adult in *A. gambiae*.

Gilbert *et al.* (1977) suggested that the brain hormone, prothoracicotrophic hormone

(PTTH), acts directly on the prothoracic glands (PGs) to stimulate the synthesis of the steroid pro-hormone ecdysone. Hydroxylation of ecdysone at the C-20 will form ecdysterone (20-hydroxyecdysone) in tissues peripheral to the PGs, which in turn initiates the moulting process. It is probable, that the nanoparticles probably provided an easy passage for the phytochemicals in the extracts into the insect. The presence of these phytochemicals might have interfered with the production of ecdysterone, most probably at the point of the addition of hydrogen ion at C-20 (preventing hydroxylation), thereby resulting in the failure of the larvae to metamorphose to pupae.

Safranek and Williams (1984), were of the opinion that juvenile hormone (JH) secreted by the corpora allata is responsible for preventing metamorphosis. As long as JH is present, the hydroxyecdysone-stimulated moults result in a new larval instar. The secretory cells of the corpora allata are active during larval moults but are inactive during the metamorphic moult.

In the last larval instar, the medial nerves from the brain to the corpora allata inhibit the gland from producing a juvenile hormone, and there is a simultaneous increase in the body's ability to degrade existing JH, resulting in metamorphosis into pupae. The phytochemicals might have acted by increasing the production of JH by the corpora allata, and the reduction in the body's ability to degrade existing JH, thereby retaining the larval form from metamorphosing to pupae.

Torchilin (2006) suggested that the cells penetrating peptides modified nanoparticles has various pharmaceutical nano-carriers (like liposomes and nanoparticles) and have been modified with cell penetrating peptides

(CPPs) which increase the rate of skin penetration. CPPs have the potential to carry lipids load such as nanoparticles, liposome and nano-structure lipid carriers (NLC) across the skin layer, it also function as an intra-cellular delivery of peptides, proteins, nucleic acid, anti-bodies imaging agent and nanoparticle system. The inhibitive properties in this study may be as a result of deactivation of certain neuro-secretory cells which are to respond by secreting brain hormones in the process of ecdysis.

Larvicidal and Pupacidal Activities of Synthesized Nanoparticles

The result (Table 1) also showed that the LD₅₀ for synthesized AgNPs of *A. paniculata* was the most effective. Lateef *et al.* (2015) were of the opinion that probably the particles denatured/interfered with or interrupted the interaction of phosphorous containing compounds like DNA or sulphur containing protein which leads to denaturation of enzymes and organelles or even death of the cell. These results also conform to the submission of Sap-Iam *et al.* (2010) that were of the opinion that, decreases in cellular membrane permeability and the reduction in ATP syntheses has a final point on the cellular function and cell death. The efficacy of the pupicidal property in the synthesized extracts was shown in Table 2. The result showed presence of active pupacidal properties in the synthesized nanomaterial, which are responsible for pupae mortality. The synthesized AgNPs of *A. paniculata* was the most effective, however the mortality encountered was strictly found to be concentration dependent. The mechanisms, at which the death occurs, could be the ability of synthesized nanomaterial to penetrate through the pupae membrane (Sap-Iam *et al.*, 2010). This result was in consonance with the submission of Sareen *et*

al. (2012) with the report that AgNPs in the intracellular space can bind to sulphur-containing proteins or to phosphorus containing compounds like DNA, which leads to the denaturation of some organelles and enzymes. Subsequently, the decrease in membrane permeability and disturbance in proton motive force causes loss of cellular function and finally cell death. Some studies have shown different degrees of potency of nanoparticles against larvae of mosquito (Lateef *et al.*, 2016b, c, d; Azeez *et al.*, 2017; Aina *et al.*, 2019).

Conclusion

It can be concluded that the synthesized AgNPs of *M. oleifera*, *A. panicula* and spider cobweb inhibited the process of development and also resulted in mortality in the larval and pupa stages of *Anopheles gambiae* sensu stricto. This can be a promising control of mosquitoes.

References

- Abbott, W.S. 1925. A method of computing the effectiveness of an insecticide, *Journal of Economic Entomology* 18: 265-267.
- Adesulu, E. A. and Sydenham, D.H.J. 2007. The freshwater fishes and fisheries of Nigeria. Macmillan Nigeria Publishers Limited, Ibadan.
- Aina, D.A., Owolo, O., Lateef, A., Aina, F.O., Abbas, S.H., Adeoye-Isijola, M., Okon, V., Asafa, T.B., Elegbede, J.A., Olukanni, O.D. and Adediji, I. 2019. Biomedical applications of *Chasmanthera dependens* stem extract mediated silver nanoparticles as antimicrobial, antioxidant, anticoagulant, thrombolytic, and larvicidal agents. *Karbala International Journal of Modern Science* 5: 71-80.
- Azeez, M.A., Lateef, A., Asafa, T.B., Yekeen, T.A., Akinboro, A., Oladipo, I.C., Gueguim-Kana, E.B. and Beukes, L.S. 2017. Biomedical applications of cocoa bean extract-mediated silver nanoparticles as antimicrobial, larvicidal and anticoagulant agents. *Journal of Cluster Science* 28: 149-164.
- Bhatt, K.C., Pandey, A., Dhariwal, O.P., Panwar, N.S. and Bhandari, D.C. 2009. Tum-thang" (*Crotalia tetragona* Roxb. Ex Andr.). A little known wide edible species in the north eastern hill of India. *Genetic Resources and Crop Evolution* 56: 729-733.
- Brown, A.W. 1986. Insecticide resistance in mosquito. A pragmatic review. *Journal of American Mosquito Control Association* 2(2): 123-140.
- Chinnamuthu, C.R. and Murugesu Boopathi, P. 2009. Nanotechnology and Agroecosystem. *Madras Agricultural Journal* 96: 17-31.
- Coates, A. 2000. Interpretation of infrared spectra, a practical approach. *Encyclopedia of Analytical Chemistry* R.A. Meyers (Ed.) John Wiley and Sons Ltd.
- Fletcher, M., Tecklehaimanot, A., Yemane, G., Kassahun, A., Kiddane, G. and Beyene, Y. 1993. Prospects for the use of larvivorous fish for malaria control in Ethiopia: search for indigenous species and evaluation of their feeding capacity for mosquito larvae. *The American Journal of Tropical Medicine and Hygiene* 96: 12-21.
- Foster, L. and Walker, A. 2009. African Malaria mosquito, *Anopheles gambiae*. Medical and Veterinary Entomology, 2nd Ed. Academic Press, Burlington MA.

Garros, C., Ngugi, N., Githeko, A.E., Tuno, N. and Yan, G. 2008. Gut content identification of larvae of the *Anopheles gambiae* complex in Western Kenya using a barcoding approach. *Molecular Ecology Resources* 8(3): 512-518.

Gilbert, P., Colliers, P.J. and Brown, M.R.W. 1977. Influence of growth rate on susceptibility to antimicrobial agent: biofilm cell cycle, dormancy and stringent response. Antimicrobial agent chemother. *Koolman Journal (Thieme, stuttgart)* 34: 1865-1868.

Huang, J., Walker, E.D., Vulule, J. and Miller, J.R. 2006. Daily temperature profiles in and around Western Kenyan larval habitats of *Anopheles gambiae* as related to egg mortality. *Malaria Journal* 5: 87.

Impoinvil, D.E., Cardenas, G.A., Gihture, J.I., Mbogo, C.M. and Beier, J.C. 2007. Constant temperature and time period effects on *Anopheles Gambiae* egg hatching. *Journal of American Mosquito Control Association* 23(2): 124-130.

Lateef, A., Ojo, S.A., Akinwale, A.S., Azeez, L., Gueguim-Kana, E.B. and Beukes, L.S. 2015. Biogenic synthesis of silver nanoparticles using cell-free extract of *Bacillus safensis* LAU 13: antimicrobial, free radical scavenging and larvicidal activities. *Biologia* 70(10): 1295-1306.

Lateef, A., Ojo, S.A., Azeez, M.A., Asafa, T.B., Yekeen T.A., Akinboro, A., Oladipo, I.C., Gueguim-Kana, E.B. and Beukes, L.S. 2016a. Cobweb as novel biomaterial for the green and eco-friendly synthesis of silver nanoparticles. *Applied Nanoscience* 6: 863-874.

Lateef, A., Akande, M.A., Ojo, S.A., Folarin, B.I., Gueguim-Kana, E.B. and Beukes, L.S. 2016b. Paper wasp nest-

mediated biosynthesis of silver nanoparticles for antimicrobial, catalytic, anti-coagulant and thrombolytic applications. *3Biotech.* 6: 140.

Lateef, A., Azeez, M.A., Asafa, T.B., Yekeen, T.A., Akinboro, A., Oladipo, I.C., Azeez, L., Ojo, S.A., Gueguim-Kana, E.B. and Beukes, L.S. 2016c. Cocoa pod extract-mediated biosynthesis of silver nanoparticles: Its antimicrobial, antioxidant and larvicidal activities. *Journal of Nanostructure in Chemistry* 6(2): 159-169.

Lateef, A., Ojo, S.A., Folarin, B.I., Gueguim-Kana, E.B. and Beukes, L.S. 2016d. Kolanut (*Cola nitida*) mediated synthesis of silver-gold alloy nanoparticles: antifungal, catalytic, larvicidal and thrombolytic applications. *Journal of Cluster Science* 27(5): 1561-1577.

Owolade, O.F., Ogunleti, D.O. and Adenekan, M.O. 2008. Titanium dioxide affects disease development and yield of edible cowpea. *Electronic Journal of Environmental, Agricultural and Food Chemistry* 7(50): 2942-2947.

Rouhani, M., Samih, M.A. and Kalantari, S. 2012. Insecticidal effect of silica and silver nanoparticles on the cowpea seed beetle, *Callosobruchus maculatus* F. (Col.: Bruchidae). *American Nano Science* 4(4): 297-305.

Rapley, L.P., Johnson, P.H., Williams, C.R., Silcock, R.M., Larkman, M., Long, S.A., Russell, R.C. and Ritchie, S.A., 2009. A lethal ovitrap-based mass trapping scheme for dengue control in Australia: II. Impact on populations of the mosquito *Aedes aegypti*. *Medical and Veterinary Entomology* 23(4): 303-316.

Safraneck, L. and Williams, C.M. 1984. Critical weights for metamorphosis in the tobacco hornworm, *Manduca sexta*. *Biological Bulletin* 167: 555-567.

Sap-Iam, N., Homklinchan, C., Larpeudomlert, R., Warisnoicharoen, W. 2010. Evaluation of anti-bacteria efficacy of biological synthesised nanoparticle. *Journal of Applied Sciences* 10(23): 3132-3136.

Sareen, S.J., Pillai, R.K., Chandramohanakumar, N. and Balagopalan, M. 2012. Larvicidal potential of biologically synthesised silver nanoparticles against *Aedes albopictus*. *Research Journal of Recent Sciences* 1: 52-56.

Shankar, S., Jaiswal, L., Aparna, R.S.L. and Prasad, R.G.S.V. 2014. Synthesis, characterization, *in vitro* biocompatibility, and antimicrobial activity of gold, silver and gold silver alloy nanoparticles prepared from *Lansium domesticum* fruit peel extract. *Materials Letters* 137: 75-78.

Torchilin V.P. 2006. Multifunctional Nanocarriers. *Advance Drug Delivery Review* 158(14): 1532-1555.

World Health Organization. 1996. World Health Organization. Report of the WHO informal consultation on the evaluation on the testing of insecticides, CTD/WHO PES/IC/96.1. Geneva: WHO; 1996. 69p. IRIS (Institutional for repository sharing). 9241208393.