

Mosquito larvicidal effect of sonicated biogenic bismuth- Silver oxide nanocomposites

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Abstract

Mosquitoes are a serious menace and they are a vector of pathogens such as malaria, filarial worm, Japanese encephalitis and dengue. *Culex sp.*, a known mosquito species is a vector of the filarial worm causing filariasis. In recent times, there has been a rise in the insecticidal resistance among mosquitoes. This has paved the need for development of alternative insecticides to combat the threat of mosquitoes. Nanotechnology is an interdisciplinary science which involves in the fabrication of materials at nanoscale (one- billionth size). Nanomaterials have a plethora of applications in medical, opto-electronic and environmental cleanup sector. The nanomaterials have also found an application in insecticidal and insect repellent studies. The present work involved in the fabrication of silver oxide- bismuth nanocomposites using ethanolic extract of *Curcuma longa* by sonication method. Structural characterization of the nanomaterials involved the usage of UV- Vis, FT- IR, SEM- EDX and XRD. The size of the nanoparticles as reported from the SEM- EDX was reported to be 123.108 nm. FT-IR analysis reported the OH- stretches of alcohol, alkene, C-O single bond stretch, OH-group of carboxylic acid disturbances and bimetallic metal-metal oxide fingerprint region. The XRD reported an orthorhombic crystal and the grain size was found to be 120.73 nm by using the modified Debye- Scherrer Equation. The larvicidal activity as against *Culex quinquefasciatus.*, was found to be 100 percent at the four tested concentrations viz., 500 ppm, 250 ppm, 125 ppm and 62.5 ppm of the nanocomposites. In *Aedes aegypti* the larvicidal potency was found to be 100 percent at the tested concentrations viz., 500 ppm, 250 ppm and 125 ppm. From the above, it was inferred that the Bismuth- Silver Oxide nanocomposites were found to be highly insecticidal against *A. aegypti* and *C. quinquefasciatus* of mosquitoes.

Keywords: Mosquitoes, *Aedes aegypti*, *Culex quinquefasciatus* nanocomposite, bismuth-Silver Oxide, insecticidal, nanomaterials

Introduction

Nanotechnology is an interdisciplinary science which involves the synthesis and fabrication of materials at nanoscale (10^{-9} m). Particularly, noble- metal nanoparticles are utilized in industries such as opto-electronic, environmental clean- up and healthcare sector. The increase in the rise of mosquito- borne diseases has become a major problem worldwide particularly, malaria, filariasis, Japanese encephalitis and dengue. *Aedes aegypti* is a known vector of dengue and *Culex quinquefasciatus* a species of mosquito is a known vector of filariasis [1]. The need of the hour is to develop an environment- friendly, cost- effective and a virulent insecticide to combat the threat of mosquito- borne diseases as there has been a rise in insecticide resistance amongst them.

Loganathan *et al* have fabricated silver nanoparticles using the aqueous leaf extract of *Knoxia sumatrensis*. The multi-potent bioactivity of these silver nanoparticles was studied. Superior antioxidant, cytotoxic and larvicidal activities against the mosquito vector *C. quinquefasciatus* were observed in the study. The characterization techniques employed in the study included X- Ray Diffraction (XRD), Fourier Transform- Infra Red (FT-IR) and Scanning Electron Microscopy (SEM) analyses and the size and morphology of the nanoparticles were hexagonal and ranged from 7.73 to 32.84 nm [2].

Rajput *et al* have synthesized silver nanoparticles using Indian Belladonna extract. The size of the nanoparticles was revealed to be 15- 20 nm from the SEM micrograph as reported by them. The silver nanoparticles were found to have anticancer, antioxidant and larvicidal activities as

against the vectors of dengue, malaria and filariasis [3]. The XRD revealed the silver nanoparticles to be crystalline in nature. However, the DLS revealed the size of the nanoparticles to be 51 nm.

Elumalai *et al* have fabricated silver nanoparticles using an aqueous extract of the leaf of *Blumea mollis*. The NPs were tested against a range of mosquitos and were found to be effective as against the larvae of *Anopheles subpictus*, *Culex vishnui* and *Aedes vittatus* [4]. The characterization of the nanoparticles was carried out with the help of UV- Vis, FT- IR, SEM and XRD analyses and the size of the nanoparticles was reported to be 23- 31 nm.

Prakash Moorthy and Helen Kavitha have demonstrated the multi-potent bioactivity including larvicidal activity of bismuth oxide nanoparticles. The size of the nanoparticles was found to be 20 nm and were found to be amorphous. Larvicidal activity was observed as against *A. aegypti* and *A. albopictus* [5].

A study by M. Prakash and co- workers have demonstrated the antibacterial, larvicidal and photocatalytic activities of gadolinium (Gd)- doped bismuth oxide nanoparticles. The doped nanoparticles were found to demonstrate excellent larvicidal activity as against *A. aegypti* and *A. albopictus*. The NPs were prepared using the *C. procumbens* leaf extract. The morphology of the nanoparticles was found to be agglomerated in nature and the size of the NPs was 30 nm on average [6]. A combination of bismuth and silver oxide nanoparticles and their application in mosquito control has not been visibly reported in the previous literature. Hence the present objective of the study was in the fabrication of a

combination of bismuth and silver oxide nanoparticles and in the study of their potential application in larvicidal activity as against the mosquito vectors *Aedes aegypti* and *Culex quinquefasciatus*.

Materials and methods

Preparation of plant extract: Freshly processed rhizomes of *Curcuma longa* were powdered of which 5 g was taken and extracted by heating with 75 ml of ethanol. The contents were filtered and the filtrate was concentrated in vacuo using a rotary vacuum evaporator. The resulting extract was cooled and further stored in a plastic bottle.

Sonication based synthesis of Bimetallic Bismuth-Silver Oxide Nanoparticles: 1 mL of the obtained extract was taken and dissolved in 99 mL of double distilled water in a 500 mL capacity beaker. Then, 0.1 M of Bismuth nitrate pentahydrate and 0.1 M of silver nitrate solutions were added to it. The contents were mixed thoroughly and were sonicated using a sonication apparatus filled with water. The beaker was fitted to a test-tube holder in such a way that it remained steady and the temperature was maintained at 42 °C for 30 minutes. The contents were observed for any color changes.

Larvicidal activity as against Mosquito larvae: The larvae of *A. aegypti* and *C. quinquefasciatus* larvae was collected from the fresh water and polluted water and brought to the laboratory. The larvae were cultured under the laboratory condition followed by standard protocol. The 3rd instar larvae were used for larvicidal activity. The larvicidal activity against the mosquito larvae was carried out in different concentrations of the nanoparticles at 500 ppm, 250 ppm, 125 ppm, 62.5 ppm. The protocol of Sachin B. Ghar *et al* [7] was followed for larvicidal activity studies. In this study however, 100 mL of distilled water was taken in thirty-two 200 mL capacity plastic cups. 20 cups were selected for *Culex sp.*, and 12 cups were selected for *Aedes sp.* 5x4 model of replication was followed in the study on *Culex sp.*, wherein the 20 cups were segregated into 5 rows and 4 columns each row containing 4 cups and same model of replication was followed for *Aedes sp.* Then, in the 5x4 replication model for *Culex sp.*, 500 ppm, 250 ppm, 125 ppm and 62.5 ppm of the bimetallic NPs were introduced in the first, second, third and fourth container respectively in

each row. This process was further repeated thrice. The similar procedure was followed in *Aedes sp.*, 3x4 model. In the 3x4 model, the cups were arranged in 3 rows and 3 columns, each row containing 3 cups. In the 3x4 replication model, 500 ppm, 250 ppm, 125 ppm and 62.5 ppm of the bimetallic NPs were introduced in the first, second, third and fourth container respectively in each row. The mortality was recorded 24 hrs interval and the data were analyzed with Abbott's formula.

$$\text{Abbott's corrected percent mortality \%} = \frac{\text{Number of larvae dead}}{\text{Total number of larvae introduced}} \times 100$$

Furthermore, ANOVA was used to analyze the significance of different concentrations of silver oxide and bismuth nanocomposites against *A. aegypti* and *C. quinquefasciatus* larvae.

Characterization of bimetallic Bismuth-Silver oxide NPs: The bimetallic NPs were characterized using UV- Vis spectrometry, FT- IR, HR- SEM- EDAX and XRD.

Results and discussion

Larvicidal activity: The larvicidal activity was reported for the first time using the bimetallic Bismuth-Ag₂O NPs. The larvicidal potency was observed as against the human mosquito vectors (HVMs) *A. aegypti* and *C. quinquefasciatus*. From the above tables and calculations, it was interestingly noted that a cent percent mortality was recorded in all the tested four concentrations with respect to the negative control which was distilled water (Fig 1&2). On comparison with the study reported by Borase *et al* [8], the activity of the silver oxide and bismuth nanocomposites exceeded in terms of magnitude than the magnitude of larvicidal activity of silver nanoparticles.

ANOVA was conducted across four concentrations against *A. aegypti* and *C. quinquefasciatus* larvae on different days. There is no significant difference in mortality rates between the various concentrations of silver oxide and bismuth nanocomposites against *A. aegypti* and *C. quinquefasciatus* larvae even at the low concentration of 62.5 ppm. The mosquito larvicidal activity of silver oxide and bismuth nanocomposites is highly effective in controlling mosquito larvae (Table 1 & 2).

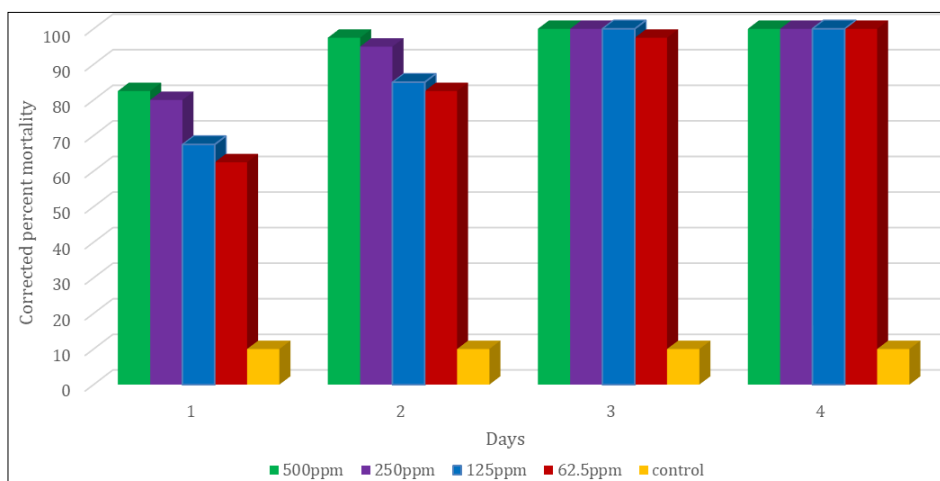


Fig 1: Larvicidal Activity of Bimetallic Bismuth- Silver Oxide NPs (96 h post- treatment) against *C. quinquefasciatus*. *(Post 96 h treatment: Number of larvae introduced in each concentration= 10)

Table 1: ANOVA table for Larvicidal Activity of Bimetallic Bismuth- Silver Oxide NPs (96 h post- treatment) against *C. quinquefasciatus*

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	201.563	3	67.188	.387	.765
Within Groups	2084.375	12	173.698		
Total	2285.938	15			

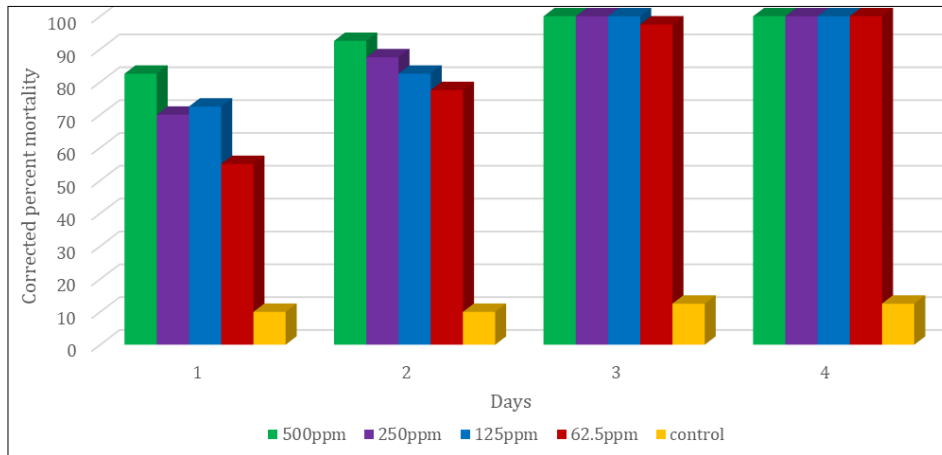


Fig 2: Larvicidal activity of bimetallic Bismuth- Silver Oxide Nanoparticles (NPs) against *A. aegypti*. *(Post 96 h treatment: Number of larvae introduced in each concentration= 10)

Table 2: ANOVA test for Larvicidal activity of bimetallic Bismuth- Silver Oxide Nanoparticles (NPs) against *A. aegypti*

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	257.422	3	85.807	.384	.766
Within Groups	2679.688	12	223.307		
Total	2937.109	15			

Characterization of NPs: The bimetallic nanoparticles were found to be brown in color. As from the UV- Vis Spectrograph, the nanoparticles were formed at the region at 333 nm indicating bathochromic shift. The FT- IR Spectrograph revealed the presence of alcohols (-OH stretch), carboxylic acids, (-C-O single bond stretch), and the presence of fingerprint region at 428, 577 cm^{-1} revealed the presence of metal- metal oxide bonding. The SEM Micrograph revealed the size of the NPs to be 123. 108 nm on average. The XRD diffractogram revealed the presence of Bismuth and silver oxide and it is in corroboration with the EDAX mapping. The crystallographic orthorhombic nature was observed and the grain size of the NPs was found

to be 120. 8 nm. The XRD was in corroboration with the crystallographic open database (COD No: 00- 100- 7165).

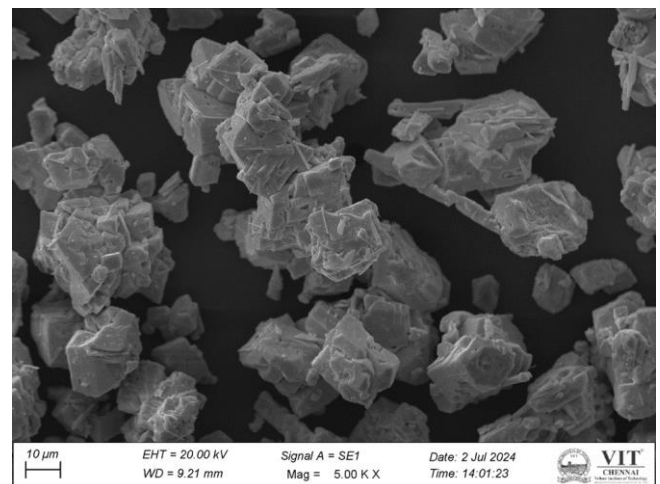


Fig 3: SEM Micrographs of Bismuth- Silver Oxide Nanoparticles

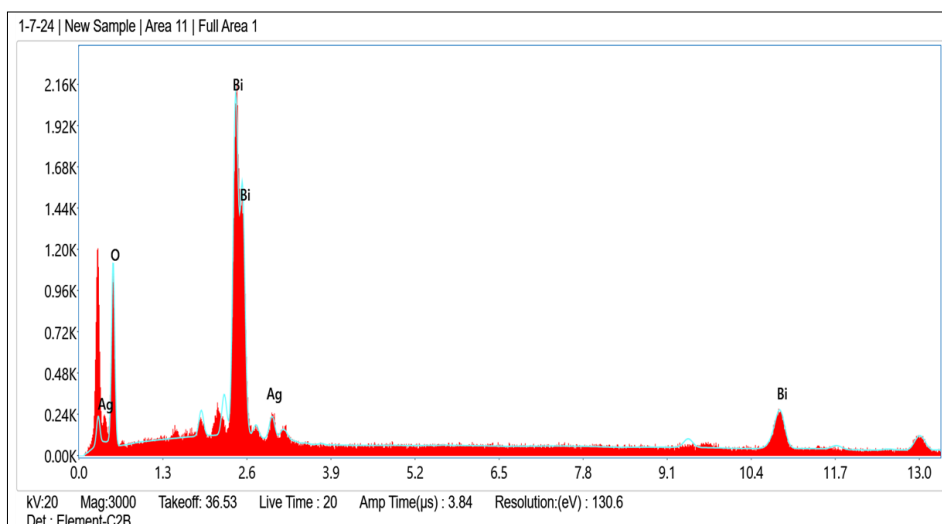


Fig 4: EDAX Spectrograph of Bismuth- Silver Oxide NPs

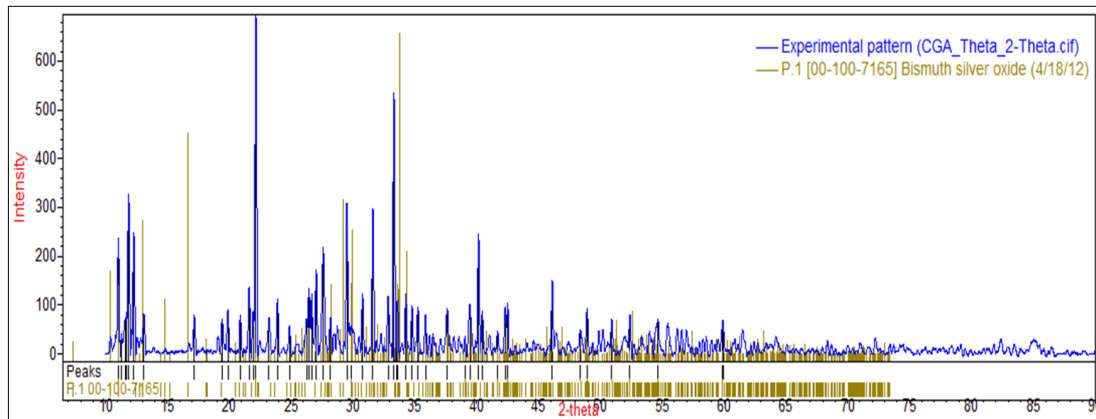


Fig 5: XRD (X- Ray Diffractogram) of Bismuth- Silver Oxide Nanoparticles. Smoothened peaks could be found in this figure

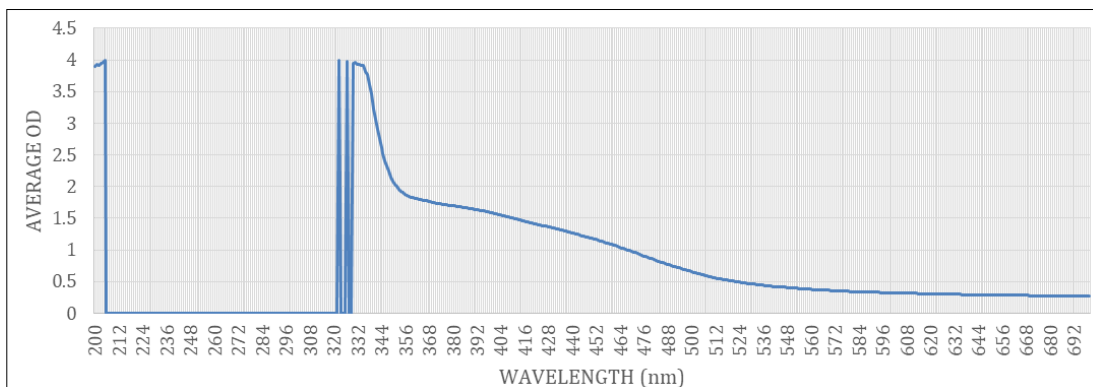


Fig 6: UV- Vis spectrograph of Bimetallic Bismuth- Silver Oxide Nanoparticles. Bathochromic shift is observed

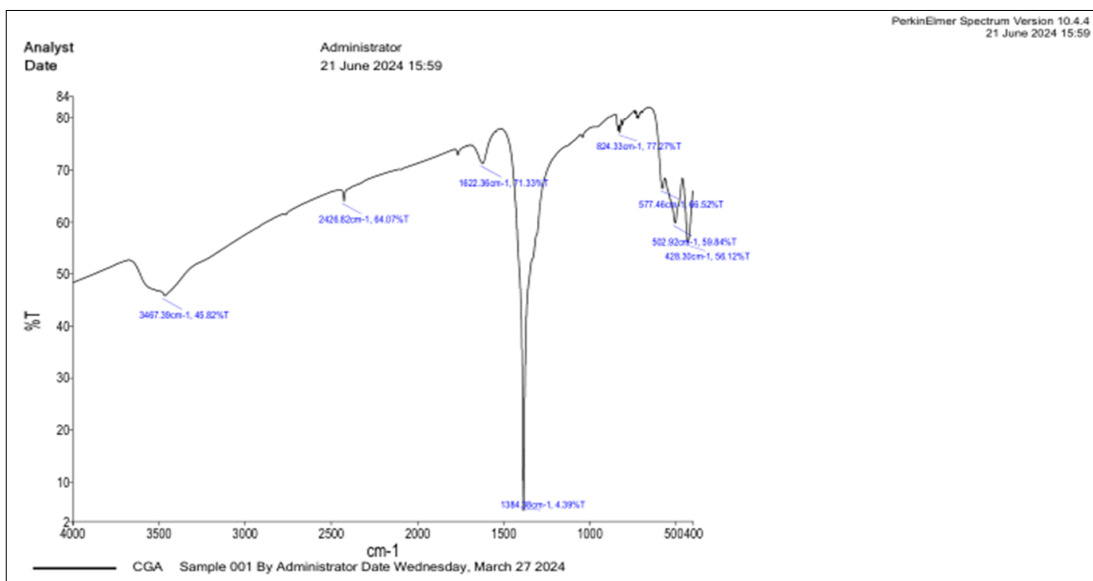


Fig 7: FT- IR spectrograph of bimetallic Bismuth- Silver Oxide NPs

Conclusion

From the above study, it could be thus concluded that the bimetallic Bi- Ag₂ O NPs were found to possess cent percent larvicidal activity of *A. aegypti* and *C. quinquefasciatus* larvae after 96 h of treatment. This could pave the way for development of a novel insecticide either via mosquito repellent fumes, organically made cones or mosquito repellent ointments.

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Conflict of interest

The authors do not have any conflict of interest.

Authors Contribution

Adithya C.G. and Jisha H.S. performed the assays. Dr. M. Raja supervised the experiment and performed the statistical analyses. Dr. M.F. Valan helped in editing the manuscript. Adithya C.G. wrote the manuscript.

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