

**Biosynthesis of green silver nanoparticles using root and leaf extracts of *Petiveria alliacea* and utilization as nano-insecticides on ants (*Pachycondyla chinensis*)**¹Abubakar S. B., ¹Olaniran O. A., ²Elegbede J. A., ^{2,3}Lateef A., ¹Adebayo T. A., ⁴Gueguim-Kana E.B., ⁴Beukes L.S., ⁵Matyumza N. and ¹Alao F.O¹Department of Crop and Environmental Protection, Ladoke Akintola University of Technology, Ogbomoso, Nigeria.²Laboratory of Industrial Microbiology and Nanobiotechnology, Department of Pure and Applied Biology, Ladoke Akintola University of Technology, Ogbomoso, Nigeria.³Nanotechnology Research Group (NANO+), Ladoke Akintola University of Technology, Ogbomoso, Nigeria.⁴Department of Microbiology, School of Life Sciences, University of KwaZulu-Natal, Private Bag X01, Scottsville, Pietermaritzburg 3209, South Africa.⁵Microscopy and Microanalysis Unit, School of Life Sciences, University of KwaZulu-Natal, Private Bag X01, Scottsville, Pietermaritzburg 3209, South Africa.**Article info****Received:** Dec. 17, 2024**Revised:** Mar. 20, 2025**Accepted:** Mar. 29, 2025**Keywords:**

Biosynthesized AgNPs,

Silver nanoparticles,

Ant,

Pachycondyla chinensis,

Dichlorvos,

Nano-insecticide

Abstract

Ant infestation is a serious concern among the people in both urban and rural areas, hence, the control becomes necessary. Nano-insecticides have been described as one of the alternatives to conventional chemicals due to their environmentally friendly nature. Silver nanoparticles were biosynthesized using *Petiveria alliacea* leaf (PaL-AgNPs) and *Petiveria alliacea* root (PaR-AgNPs) against a species of ants (*Pachycondyla chinensis*) at four concentrations (25%, 50%, 75% and 100% v/v), with different modes of applications (contact, fumigant and feeding deterrent). Synthetic insecticides (Dichlorvos) and control were included in the treatments for comparison. The treatments were arranged in a Complete Randomized Design with 3 replications. Silver nitrate was used in the biosynthesis of AgNPs by *P. alliacea* leaf (PaL-AgNPs) and root (PaR-AgNPs) with maximal absorbance ranged from 426-442 nm. The results clearly showed that nano *P. alliacea* leaf and root exhibited insecticidal potential against the studied insect when compared with untreated ants. However, PaR-AgNPs had significantly higher efficacy than PaL-AgNPs in the studied insect. None of the nano-insecticide formulations was as effective as dichlorvos at the earlier stage of application. However, the mode of application and concentrations applied determined the toxicity of the tested nano-insecticides. Therefore, green silver nanoparticles from *P.alliacea* leaf and root extracts can be used to control ant infestations.

1. Introduction

Ants are one of the most abundant and dominant groups of animals in terrestrial ecosystems, with more than 15,000 described species, and are found in almost all ecosystems [1]. Their presence in the ecosystem has been linked with the spread of human pathogens [2]. Contamination of hospital apparatuses by ants leads to the mechanical transmission of diseases [3], allergies, stings, and bites [4], and food contami-

nation [5]. In the tropics, many ant species have already been incriminated as vectors of pathogens [6]. Food-borne disease pathogens such as *Serratia*, *Citrobacter*, *Klebsiella*, *Enterobacter*, *Proteus*, *Staphylococcus*, and *Yersinia pestis* were isolated from ants [7]. Ants may adversely affect plant health through their mutualistic relationships with honeydew-producing pests, which they defend in return for carbohydrates [8]. The activity of ant workers can contribute to a rise

in Hemiptera populations, which are recognized for causing damage to plants and acting as vectors for a range of plant diseases [9].

Historically, synthetic insecticides have been the dominant method for managing ant infestations [10]. The common types of insecticides are organochlorine, organophosphates, carbamate and synthetic pyrethroids [11]. The prolonged use of these pesticides has raised concerns regarding resistance development in pest populations, as evidenced by studies showing that *Aedes aegypti* larvae exhibit resistance to organophosphates due to enzymatic adaptations [12]. Conventional insecticides have also been scrutinized due to their potential toxicity to non-target species and the environment [13].

Botanical pesticides, which are naturally occurring chemicals derived from plants, have been recognized as an appealing substitute for synthetic chemical insecticides in pest management [14] due to their minimal risk to both the environment and human health [15]. Botanical pesticides are increasingly recognized for their multifaceted properties, which include toxicity to pests, repellency, anti-feedant effects, and insect growth regulatory activities [16]. These characteristics make them essential tools in sustainable agricultural practices, particularly in integrated pest management (IPM) systems [17]. The efficacy of botanical pesticides is largely attributed to their diverse phytochemical compositions, which can target various physiological processes in pests [18]. Recent research has shown that extracts from *P. alliacea* possess significant insecticidal properties, which can be attributed to their phytochemical constituents. For instance, [19] discusses the biopesticidal activities of *P. alliacea* against *Sitophilus oryzae*, indicating that the plant's extracts can effectively reduce pest populations, thereby preserving stored products [19]. This aligns with the findings of [20], who identified dibenzyl trisulfide as the active insecticidal compound in *P. alliacea*, demonstrating its efficacy against various field insect pests. Moreover,

[21] explored the chemical components of *P. alliacea* essential oils, revealing that certain compounds, such as benzenecarbothioic acid, constitute a significant portion of the extract and contribute to its antimicrobial and potentially insecticidal properties [21]. This is further supported by [22], which emphasizes the broad spectrum of antimicrobial activities exhibited by *P. alliacea*, suggesting that its extracts could also target insect pests effectively [22].

Nano-insecticides represent a significant advancement in pest management, leveraging the principles of nanotechnology to create safer and more effective alternatives to traditional synthetic insecticides. Nanotechnology, which involves manipulating materials at the nanoscale (typically 1 to 100 nanometers), allows for the development of pesticide formulations that enhance the bioactivity and stability of active ingredients while minimizing environmental impact and toxicity to non-target organisms [23].

Nano-insecticides from synthesized nanoparticles represent a new frontier in pest control [24]. These nanoparticles, often synthesized through green methods using plant extracts, exhibit unique properties that enhance their insecticidal effectiveness. Studies have shown that silver nanoparticles possess strong antimicrobial and insecticidal activity, making them ideal for use against pest populations that have developed resistance to conventional pesticides [25][26][27].

Therefore, this study aims to evaluate the insecticidal effectiveness of nanobiosynthesized extracts from the root and leaf parts of *P. alliacea* in controlling ant populations with three bioassays (contact, fumigant and feeding deterrent) to establish the best mode of application for pest management.

2. Materials and Methods

2.1 Study site

The research was carried out at the Crop and Environmental Protection Insects Room, Ladoke Akintola University of Technology,

Ogbomoso North Local Government Area in Oyo state. Ogbomoso is located on the 8°10'1" North of the equator and 4°10'1" East of the Greenwich meridian.

2.2 Insect collection

An insect box measuring 41.5 cm in length, 31.5 cm in width, and 44.5 cm in height, with a glass cover at the top for observation and mosquito netting on the sides for ventilation, was used to collect *Pachycondyla chinensis* from the Apiary section of the Ladoké Akintola University of Technology Teaching and Research Farm. The box was supplied with sugary substances, including pineapple and oranges, and subsequently moved to the insectarium. It was left undisturbed for seven (7) days to allow the *P. chinensis* to acclimate to their new environment, ensuring that only the healthiest ants were selected for the experiment.

2.3 Plant parts collection and preparation

Petiveria alliacea leaf and root were collected from the Ladoké Akintola University of Technology (LAUTECH) Teaching and Research Farm in Ogbomoso, Nigeria, and air-dried for three weeks. The dried materials were then ground into a powdered form using an electric grinder (Silvercrest commercial blender, 7000 watts). 10 grams of each powdered plant material were taken to the Microbiology Laboratory unit at LAUTECH for biosynthesis.

2.4 Biosynthesis and Characterization of Silver nanoparticles.

The nanoparticle solution was prepared following the established procedure by [28], with slight modifications. Powdered samples of each selected plant part (*P. alliacea* root and leaf) were dissolved in 100 ml of distilled water. The mixture was incubated in a water bath at 4000 rpm for 20 minutes. The resulting clear supernatant was used for synthesis by combining it with 1 mM silver nitrate solution. The mixture was then exposed to sunlight for photoactivation and monitored for a colour change, which was allowed to stabilize before further use. The biosynthesized

AgNPs were analyzed using UV spectroscopy and Fourier Transform Infrared Spectroscopy (FTIR).

2.5 Experimental Design and Entomological Bioassays

The small transparent plastic container of 0.78 m³ (4.5 cm X 5.5 cm) was used in carrying out these bioassays; this plastic container was used for easy observation of the studied insect throughout the experiments. The biosynthesized AgNPs as nano-insecticides used include *P. alliacea* and roots (PaL-AgNPs and PaR-AgNPs), which were applied at 100 %, 75 %, 50 % and 25 % v/v using three (3) modes of application (contact, fumigant and feeding deterrent). The experiment was set up in a Completely Randomized Design (C.R.D) with three replicas. The results obtained were compared with the untreated and synthetic insecticides (dichlorvos (DDforce^(r)).

2.5.1 Bioassays

2.5.1.1 Contact toxicity

From the stock solution, 10 ml of each PaL-AgNPs, PaR-AgNPs and dichlorvos solution was measured with a medical syringe (size: 20) and applied to each designated transparent container which contained 4 *P. chinensis* workers while ordinary water was applied as negative control. The Ant behavioural response to each treatment was recorded after the stipulated time. The number of dead and alive were recorded after 2, 4, 6, 8 and 24 hours of exposure.

2.5.1.2 Fumigant toxicity

Cotton wool (1 g) was impregnated with 10 ml of each PaL-AgNPs, PaR-AgNPs, and dichlorvos solution. The treated cotton wool was placed into the transparent container and four active *P. chinensis* and was introduced afterwards. The number of dead and alive insects was recorded after 2, 4, 6, 8 and 24 hours of exposure.

2.5.1.3 Feeding deterrent

Active Ant workers (4) were introduced into a transparent plastic container designated for bioassay. Using an electronic sensitive

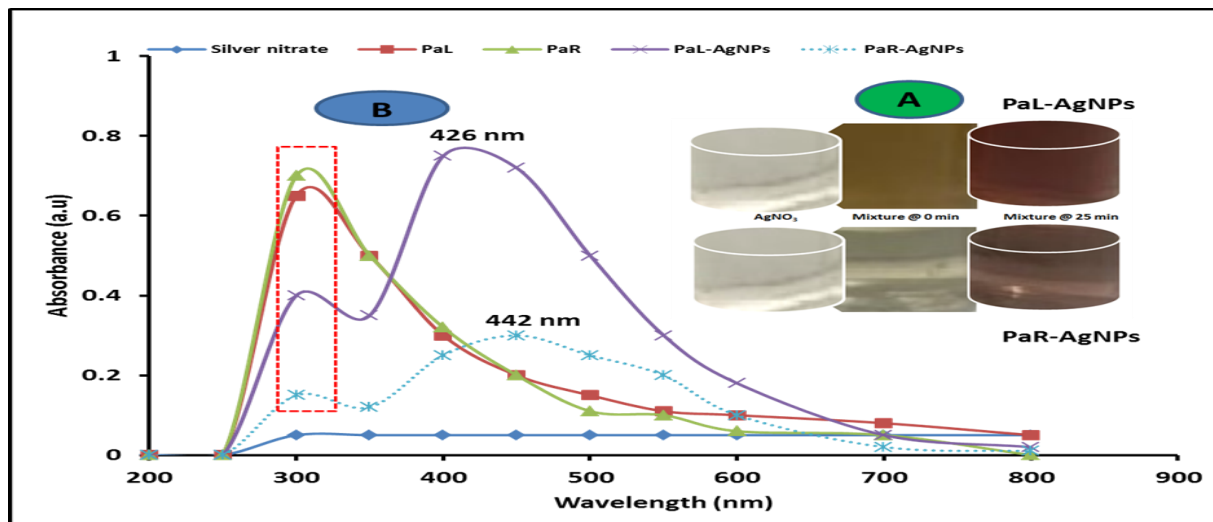


Fig. 1: Course of biosynthesis of Nano *P. alliacea* leaf (PaL-AgNPs) and Nano *P. alliacea* root (PaR-AgNPs) showing (A) biosynthesized AgNPs and (B) silver nitrate solution

scale (Camry sensitive scale), 5 g of sugarcane was chopped and soaked in 10 ml of the respective nano insecticide and dichlorvos solution for 5 minutes. The treated sugarcane was removed and allowed to drain for 5 minutes; the treated sugarcane was then placed inside the respective transparent plastic container.

2.6 Data Collection and Analysis

Ant mortality was determined at 2, 4, 6, 8, and 24 hours and these data were subjected to

analysis of variance (ANOVA) using the complete block design and means were separated at a 5% probability level using software analytical system package (SAS, 2005).

3. Results

3.1 Biosynthesis and Characterization of AgNPs

The leaf and root extract of *P. alliacea* biosynthesized AgNPs (Nano *P. alliacea* leaf (PaL-AgNPs) and Nano *P. alliacea* root (PaR-AgNPs) in 10 min under ambient conditions

Table 1: Effects of insecticides applied through contact toxicity on Ant

Treatments	Rate	Hours of exposure				
		2	4	6	8	24
Dichlovors		100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
Control		0 ^c	0 ^d	0 ^d	0 ^g	0 ^f
PaR-AgNPs	25	8.3 ^c	8.3 ^d	8.3 ^d	16.7 ^{efg}	33.3 ^e
	50	6.3 ^c	6.3 ^d	31.3 ^c	37.5 ^{cde}	50.0 ^{cde}
	75	0.0 ^c	18.8 ^{cd}	43.8 ^{bc}	56.3 ^{bc}	68.8 ^{bc}
	100	43.8 ^b	50.0 ^b	62.5 ^b	68.8 ^b	81.3 ^{ab}
PaL-AgNPs	25	6.3 ^c	6.3 ^d	6.3 ^d	6.3 ^{fg}	12.5 ^f
	50	0 ^c	0 ^d	6.3 ^d	25.0 ^{def}	43.8 ^{de}
	75	6.3 ^c	12.5 ^d	31.3 ^c	43.8 ^{cd}	56.3 ^{cd}
	100	18.8 ^c	37.5 ^{bc}	56.3 ^b	56.3 ^{bc}	81.3 ^{ab}
Insecticides						
Dichlovors		100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
Control		0 ^c	0 ^c	0 ^c	0 ^c	0.0 ^c
PaR-AgNPs		15.0 ^b	21.7 ^b	38.4 ^b	46.7 ^b	60.0 ^b
PaL-AgNPs		7.8 ^{bc}	14.1 ^{bc}	25.0 ^b	2.8 ^b	48.4 ^b

Means with the same superscript(s) are not significantly different at 5% probability.

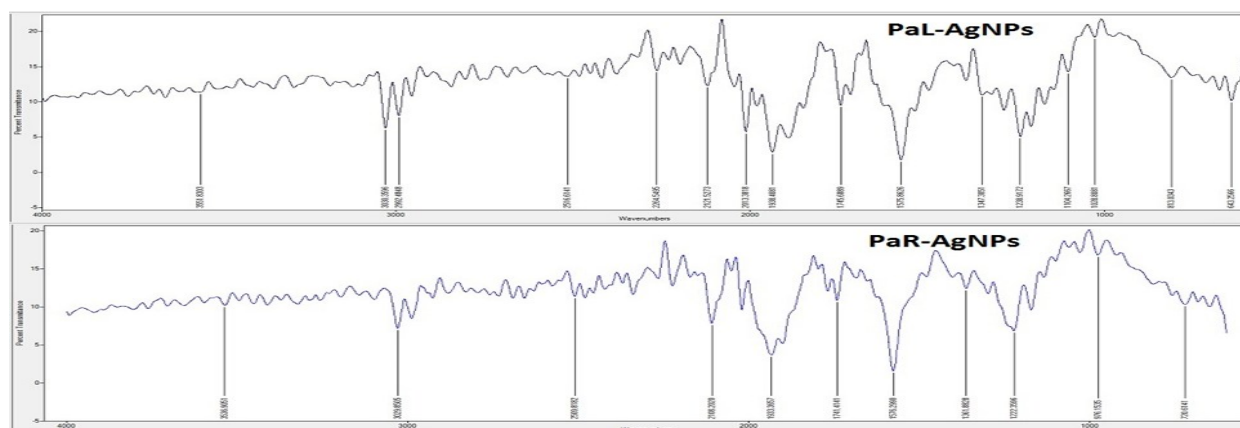


Fig. 2: FTIR spectra of Nano *P. alliacea* leaf (PaL-AgNPs) and Nano *P. alliacea* root (PaR-AgNPs)

of temperature ($30 \pm 2^\circ\text{C}$) with typical dark brown colour for Nano *P. alliacea* leaf and light brown colour for Nano *P. alliacea* root (Fig. 1A) which deepened over time and stabilized eventually after 25 min. The brownish-coloured biosynthesized AgNPs have been extensively reported [30][29][31]. Moreover, maximum absorbance occurred at the wavelengths of 426.32 and 442.11 nm for Nano *P. alliacea* leaf and Nano *P. alliacea* root respectively (Fig. 1B). Absorbance at these wavelengths is due to the unique surface plasmon resonance of the biosynthesized AgNPs and the values here reported are within the range of 391- 460 nm previously established in literature for AgNPs [28][30][31].

Also, FTIR absorption spectra revealed distinct and prominent bands at 3030, 2121, 2013, 1938, 1745, 1575, 1284, and 643 cm^{-1} for Nano *P. alliacea* leaf, which are observed to be close to 3029, 2108, 1933, 1576, and 1222 cm^{-1} obtained for Nano *P. alliacea* root (Fig. 2). Also, other closely related minor peaks obtained were shown at 3551, 2516, 1347, 1104, 1028 and 813 cm^{-1} for Nano *P. alliacea* leaf and 3536, 2509, 1741, 1361, 976 and 720 cm^{-1} for Nano *P. alliacea* root.

Spherical nature of the particles of PaL-AgNPs and PaR-AgNPs was revealed by TEM analysis, coupled with presence of silver (Fig. 3).

As shown in Table 1, there were statistically significant differences among the treatments. At 2 hours after exposure (HAE), Dichlorvos achieved complete ant mortality

(100%) of the studied insects. No significant difference was observed between the effects of *P. alliacea* leaf and root treatments across concentrations, except for the 100% v/v *P. alliacea* root application.

The highest mortality (50.0%) was observed with PaR-AgNPs applied at 100% v/v killed a significantly higher number of ants than other concentrations, while there was no significant difference among ants treated with PaL- AgNPs at 75%, 50%, and 25% v/v, except 100% v/v treatment, which achieved 37.5% mortality(4 HAE). Additionally, PaL-AgNPs at 25% and 50% v/v were as effective as the PaR-AgNPs at the same concentrations at HAE.

PaR-AgNPs and PaL-AgNPs applied at 100% v/v had the same significant effects on the studied insect, while PaR-AgNPs killed a higher percentage number of ants than PaL-AgNPs when applied at 75% and 50% v/v. Meanwhile, the efficacy of PaR-AgNPs at 25% v/v was comparable with PaL-AgNPs at 25% and 50% v/v at 6 HAE.

However, the highest mortality of the studied insect was observed in the ant treated with PaR-AgNPs 100% v/v, while PaR-AgNPs at 75 % compete effectively with PaL-AgNPs at 100% v/v at 8 HAE. Meanwhile, a significantly higher number of dead ants was recorded from the ants treated with PaL-AgNPs at 75% than PaR- AgNPs at 50% v/v at 8 HAE.

At 24 HAE, no significant difference was observed between the 100% v/v of PaR-

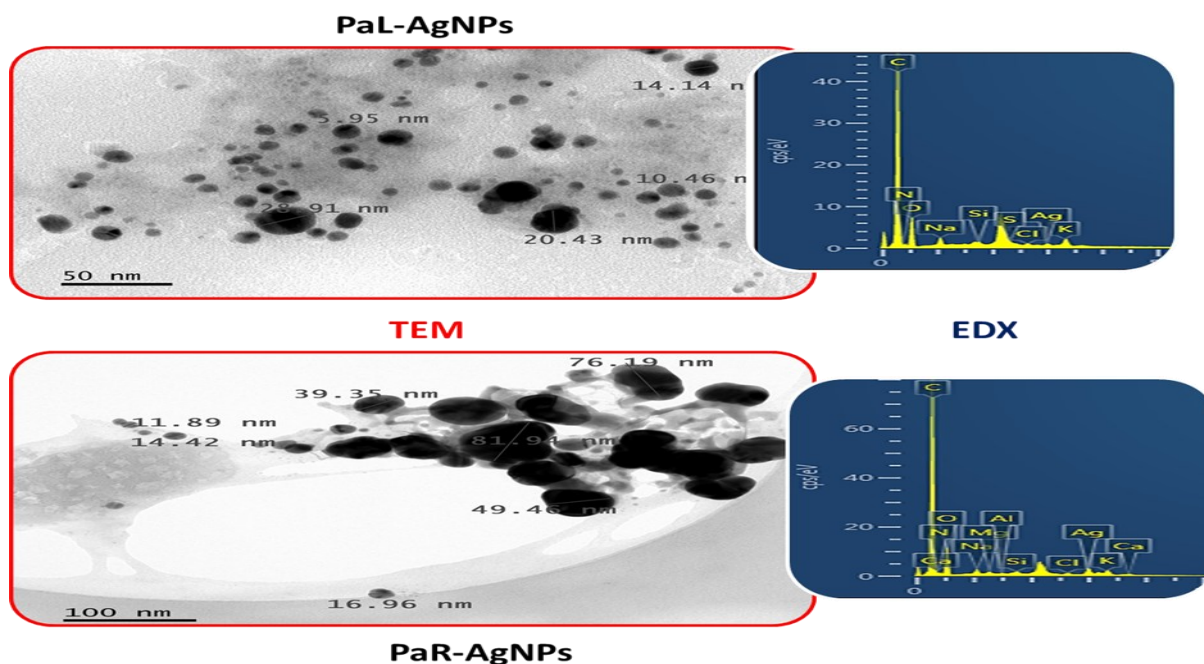


Fig. 3: The TEM micrographs (a) Field Emission from Scanning Electron Micrography of biosynthesized *Petiveria alliacea* root and leaf (b) EDX spectra of the biosynthesized AgNPs

AgNPs and PaL- AgNPs. PaR- AgNPs tested at 25% 75%v/v killed a significantly higher number of ants than PaL- AgNPs applied at the same concentration.

As shown in Fig 4, 25 v/v resulted in a mortality rate of 21.43%, which was the least effective among the treatments. Concentration applied at 50% v/v had a higher biocidal

effect (46.9%) than 255 v/v. At 75% v/v, 62.5% mortality was observed, while 87.5% mortality was recorded in the ant treated with 100% v/v.

3.2 Effect of insecticides applied through fumigant toxicity on ant

Table 2 shows statistically significant dif-

Table 2: Effect of insecticides applied through fumigant toxicity on Ant

Treatments	Rate	Hours of application				
		2	4	6	8	24
Dichlovors		100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
Control		0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^c	0.0 ^f
PaR-AgNPs	25	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^c	8.3 ^{ef}
	50	0.0 ^b	0.0 ^b	0.0 ^b	6.3 ^{bc}	18.8 ^{def}
	75	0.0 ^b	0.0 ^b	0.18 ^b	25.0 ^b	43.8 ^{bc}
	100	0.0 ^b	0.0 ^b	18.8 ^b	25.0 ^b	56.3 ^b
PaL-AgNPs	25	0.0 ^b	0.0 ^b	0 ^b	0.0 ^c	12.5 ^{ef}
	50	0.0 ^b	0.0 ^b	6.3 ^d	6.3 ^{bc}	25.0 ^{cde}
	75	0.0 ^b	0.0 ^b	12.5 ^b	25.0 ^b	31.3 ^{cde}
	100	0.0 ^b	0.0 ^b	12.5 ^b	25.0 ^b	37.5 ^{bcd}
Insecticides						
Dichlovors		100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
Control		0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b
PaR-AgNPs		0.0 ^b	0.0 ^b	10.0 ^b	15.0 ^b	0.0 ^b
PaL-AgNPs		0.0 ^b	0.0 ^b	7.8 ^b	14.1 ^b	15.6 ^b

Means with the same superscript(s) are not significantly different at 5% probability.

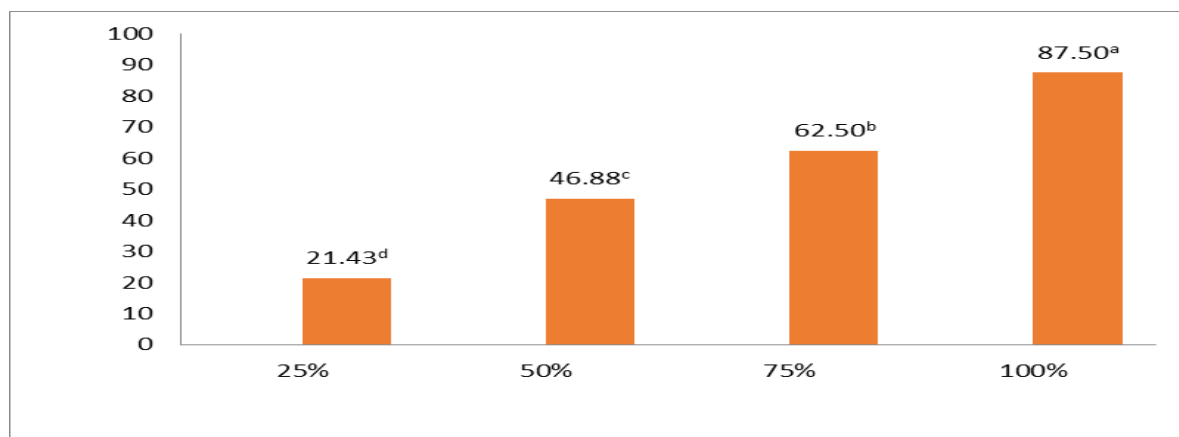


Fig. 4: Effect of rates of application of insecticide as contact toxicity

ferences among the treatments. At 2 hours after exposure (HAE), Dichlorvos resulted in 100% ant mortality in the treated container. However, no significant differences were observed between nano formulations of PaR-AgNPs and PaL-AgNPs regardless of the concentration. This trend remained consistent at 4 and 6 hours after application.

At 8 HAE, PaR-AgNPs applied at 100% and 75% v/v exhibited similar efficacy to PaL-AgNPs applied at the same concentrations. The effectiveness of PaR-AgNPs at 50% v/v was also comparable to that of PaL-AgNPs at the same concentration. Likewise, there was no significant difference between treatments

using PaR-AgNPs and PaL-AgNPs at 25% v/v.

However, the highest mortality of the studied insect was observed in the ant treated with PaR-AgNPs applied at 100% v/v at 24 HAE. The mortality of ants treated with PaL-AgNPs at 75% v/v and PaR-AgNPs at 50% v/v were not significantly different. Furthermore, the mortality recorded for PaR-AgNPs at 75% v/v was higher than that at 50% v/v.

As presented in Fig. 5, 64.58% mortality was observed at 100 v/v rate of application, there was a significant decrease in efficacy at 75 v/v with 37.50, and the downtrend continues at 50 and 25 v/v with 21.88 and 10.71

Table 3: Effect of insecticides applied through feeding deterrent toxicity on Ant

Treatments	Rate	Hours of exposure				
		2	4	6	8	24
Dichlorvos		100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
Control		0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b
PaR-AgNPs	25	0.0 ^b	0.0 ^b	0.0 ^b	6.3 ^b	12.5 ^{bc}
	50	0.0 ^b	0.0 ^b	0.0 ^b	18.8 ^b	18.8 ^{bc}
	75	0.0 ^b	0.0 ^b	6.3 ^b	6.25 ^b	6.3 ^c
	100	6.3 ^b	6.25 ^b	6.3 ^b	12.50 ^b	13.8 ^{bc}
PaL-AgNPs	25	0.0 ^b	0.0 ^b	8.3 ^b	8.3 ^b	8.3 ^c
	50	0.0 ^b	0.0 ^b	0.0 ^b	6.3 ^b	6.3 ^c
	75	0.0 ^b	0.0 ^b	0.0 ^b	12.5 ^b	18.8 ^{bc}
	100	0.0 ^b	8.33 ^b	0.0 ^b	18.8 ^b	31.3 ^b
Insecticides						
Dichlorvos		100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
Control		0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^c
PaR-AgNPs		0.0 ^b	1.67 ^b	1.7 ^b	11.7 ^b	16.7 ^b
PaL-AgNPs		1.56 ^b	1.56 ^b	3.1 ^b	10.9 ^b	14.1 ^{bc}

Means with the same superscript(s) are not significantly different at 5% probability.

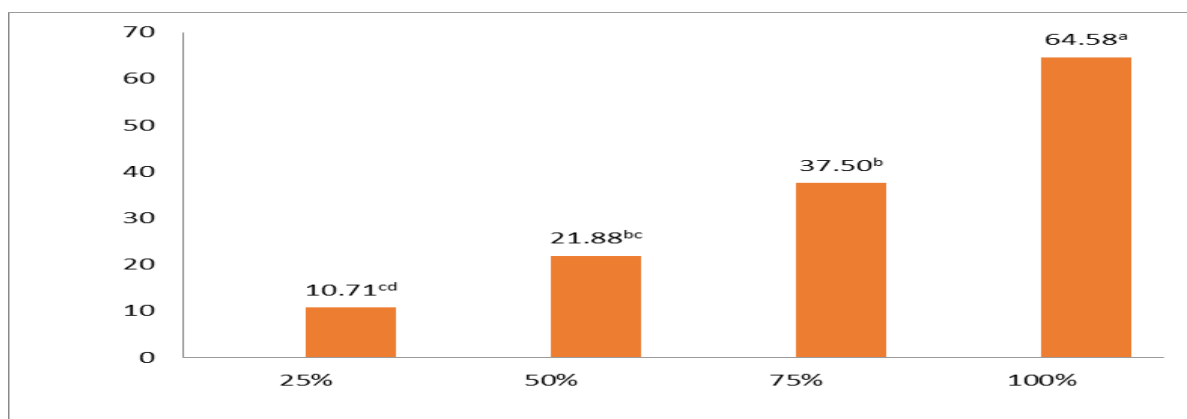


Fig. 5: Effect of rates of application of insecticide as fumigant toxicity

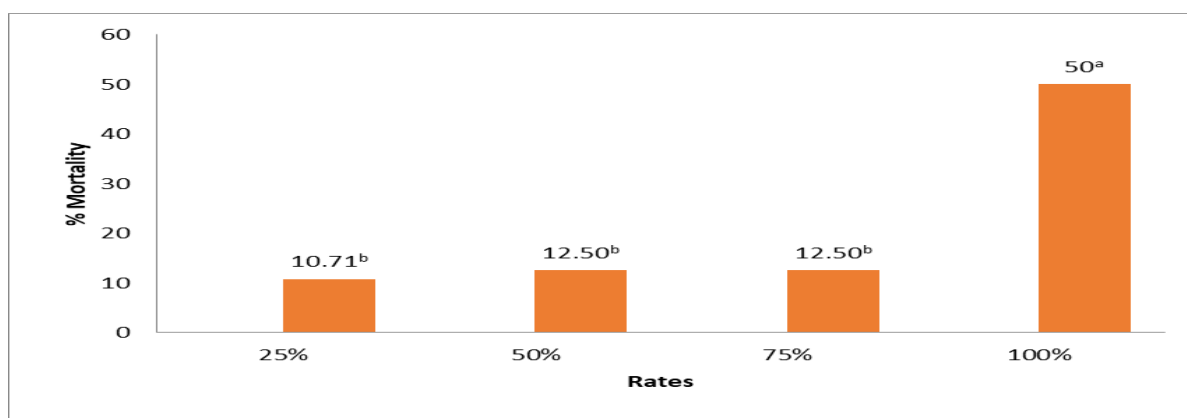


Fig. 6: Effect of rates of application of insecticide as feeding deterrent toxicity

respectively.

3.3 Effect of insecticides applied through feeding deterrent on ant

As presented in Table 3, there was a statistical difference among the treatments. At 2 hours after exposure (HAE), 100% of the ants were killed in the container treated with Dichlovers. Meanwhile, there was no significant difference between the nano insecticides irrespective of the v/v. Similar results were observed at 4, 6, and 8 hrs of application.

At 24 (HAE), PaL-AgNPs applied at 100% v/v killed significantly the highest number of ants than other applied nano insecticides. However, PaR-AgNPs applied at 100% v/v significantly perform better than other concentrations. PaL-AgNPs applied at 75% v/v had the same significant effects as PaR-AgNPs applied at 50% and 25% concentrations.

As shown in fig 6, 50% mortality was observed at 100 v/v in respect to feeding toxicity, there was no significant difference in another level of exposures.

4. Discussion

Synthetic insecticides are used to control both field and storage insect pests due to their rapid action on the target insects [32, 33]. However, synthetic insecticides have been implicated in causing environmental hazards, and their unavailability at critical times is a significant concern [34, 35]. Therefore, nano-insecticides were formulated using silver nanoparticles (AgNPs) with *P. alliecia* leaf and *P. alliecia* root applied at different concentrations (25%, 50%, 75% and 100%) v/v. The products were analyzed using UV spectrometry and FTIR analysis. The biosynthesized AgNO₃ were used as nano-insecticides against *Pachycondyla chinensis* ant workers

as indices with different bioassays (contact toxicity, fumigant toxicity and feeding deterrent bioassay). The tested nano-insecticides exhibited insecticidal action against the studied ants when compared with untreated ants. This observation is in line with the earlier study by [36], who reported that *P. alliacea* effectively controls ant infestation. Similarly, insecticidal activities of crude extracts of *P. alliacea* have been reported against ants [37] [20]. This suggests that the tested plant extract possesses an insecticidal chemical compound. However, the insecticidal effect of *P. alliacea* extract can be attributed to dibenzyltrisulphide [38][39].

The bioefficacy of nano-insecticide formulations had delayed effects compared with synthetic insecticides (dichlorvos) against *Pachycondyla chinensis*. For instance, 100% mortality of the studied insect was observed in the container treated with dichlorvos within 2 hours after the application of the insecticides. This delayed effect displayed by PaL-AgNPs and PaL-AgNPs is a major characteristic feature of botanical insecticides [40][41].

The rate of toxicity of nano-insecticide depends on the dosage applied. For instance, 100% concentration killed the studied ant faster than 75% v/v, while the least mortality of the ant was recorded on the ant treated with 25% v/v. This observation concurred with earlier researchers who reported that the effectiveness of nano-insecticides depends on the tested concentration [42][43][44]. Also, the effectiveness of nano *P. alliacea* increased with the hours of application.

However, it was observed that the mode of application also determined the effectiveness of nano-insecticide formulations. The application of nano-insecticides through contact killed a higher percentage of *pachycondyla chinensis* than nano-insecticides applied through fumigant. This is an indication that biosynthesized AgNPs should be applied through contact bioassays. This study collaborated with [45][46], who reported that plant-based silver nano-insecticides effectively control ants when applied through contact. Contrarily, [47][48] reported that nano-

insecticides applied through fumigants perform better than when applied through contact against ants.

Comparatively, the application of nano-insecticides through contact had higher ant mortality ranging from 12.5% - 81% followed by the application of nano-insecticides through fumigant which had the number of mortality of ants at 8% - 56% while the least number of ant mortality was recorded on ant treated with feeding toxicity which had 31%. This observation is in line with the earlier research work by [49][50], who reported that the application of biosynthesized AgNPs through contact killed faster than other modes of application.

5. Conclusion

In conclusion, this study has demonstrated that biosynthesized silver nanoparticles (AgNPs) derived from *Petiveria alliacea* extracts possess significant effects as nano-insecticides in the control of ants, particularly when applied at higher concentrations ranging from 75% to 100% v/v. The findings highlight the efficacy of *P. alliacea*-based nano-insecticides as a promising alternative to conventional chemical pesticides, offering an environmentally friendly and targeted approach to pest management. Incorporating these nano-insecticides into integrated pest management (IPM) programs could provide a sustainable solution for controlling ant infestations while reducing the ecological footprint associated with traditional pesticides. Further research could explore these nano-insecticide formulations against other insects and for environmental safety purposes.

References

- [1] Fayle, T. M., & Klimeš, P. (2022). Global perspectives on ants' roles in ecosystems. *Annual Review of Ecology, Evolution, and Systematics*, 53, 123–148. <https://doi.org/10.1146/annurev-ecolsys-011922-022546>
- [2] Kraemer, S. A., Ramachandran, A., & Perron, G. G. (2019). Antibiotic pollution in the environment: from microbial ecology to public policy. *Microorganisms*, 7(6), 180. <https://doi.org/10.3390/microorganisms7060180>
- [3] Cruz, F. M., Silva, J. M., Oliveira, A. P., & Almeida, R. M. (2020). Ants in a hospital environment and their potential as mechanical vectors of

- bacteria. *Journal of Hospital Infection*, 106(2), 246–252. <https://doi.org/10.1016/j.jhin.2020.01.012>
- [4] Guillet, C., Martin, O. Y., Meincke, C., Joerg, L., & Schmid-Grendelmeier, P. (2022). Part I: Insect stings and bites—Beyond the realm of bee and wasp allergies: A survey of the literature and our own cases. *Allergo Journal International*, 31(6), 183–193. <https://doi.org/10.1007/s40629-022-00199-5>
- [5] Abebe, G. M. (2020). The role of bacterial biofilm in antibiotic resistance and food contamination. *International journal of microbiology*, 2020(1), 1705814. <https://doi.org/10.1155/2020/1705814>
- [6] Akinwande, K. L., Atunwa, O. B., & Adewale, P. O. (2024). Anthropization drives in-door establishment, dispersal and food-borne pathogens' carrying capacity of ants (Hymenoptera: Formicidae). *Proceedings of the Indian National Science Academy*, 1–10. <https://doi.org/10.1007/s43538-024-00313-x>
- [7] Pesquero, M. A., Carneiro, L. C., & Pires, D. D. J. (2012). Insect/Bacteria association and nosocomial infection. *Salmonella, A Diversified Superbug*, 449–468.
- [8] Poelman, E. H., Cusumano, A., & De Boer, J. G. (2022). The ecology of hyperparasitoids. *Annual Review of Entomology*, 67(1), 143–161. <https://doi.org/10.1146/annurev-ento-060921-072718>
- [9] McCalla, K. A., Milosavljević, I., & Hoddle, M. S. (2023). A low-toxicity baiting program precipitates collapse of Argentine ant and ant-associated hemipteran pest populations in commercial citrus. *Biological Control*, 177, 105105. <https://doi.org/10.1016/j.biocontrol.2022.105105>
- [10] Lin, P. A., Paudel, S., Lai, P. C., Gc, R. K., Yang, D. H., & Felton, G. W. (2024). Integrating water and insect pest management in agriculture. *Journal of Pest Science*, 97(2), 521–538. <https://doi.org/10.1007/s10340-023-01701-w>
- [11] Kannan, M., Bojan, N., Swaminathan, J., Zicarelli, G., Hemalatha, D., Zhang, Y., Ramesh, M., & Faggio, C. (2023). Nanopesticides in agricultural pest management and their environmental risks: a review. *International Journal of Environmental Science and Technology*, 20(9), 10507–10532. <https://doi.org/10.1007/s13762-023-04795-y>
- [12] Viana-Medeiros, P. F., Bellinato, D. F., & Valle, D. (2018). Laboratory selection of *Aedes aegypti* field populations with the organophosphate malathion: negative impacts on resistance to deltamethrin and to the organophosphate temephos. *PLoS neglected tropical diseases*, 12(8), e0006734. <https://doi.org/10.1371/journal.pntd.0006734>
- [13] Mužinić, V., & Želježić, D. (2018). Non-target toxicity of novel insecticides. *Archives of Industrial Hygiene and Toxicology*, 69(2), 86–102. <https://doi.org/10.2478/aiht-2018-69-3111>
- [14] Damalas, C. A., & Koutroubas, S. D. (2020). Botanical pesticides for eco-friendly pest management: Drawbacks and limitations. *Pesticides in Crop Production: Physiological and Biochemical Action*, 181–193. <https://doi.org/10.1002/9781119432241.ch10>
- [15] Nefla, Y., & Kalariya, G. B. (2024). Nano-Enhanced Essential Oils as Insecticide. *Indian Journal of Entomology*, 1–7. <https://doi.org/10.55446/IJE.2024.1962>
- [16] Olanrewaju, O., Edamaku, P., Nwite, O., Umoru, U., & Murtala, M. (2023). Pesticidal effects of GMELINA ARBOREA (roxb) *powders on beans weevil (Callosobruchus maculatus)* at vom.. <https://doi.org/10.1101/2023.04.28.538680>
- [17] Khodijah, S. (2024). The efficacy of botanical pesticides in controlling coffee berry borer, *Hypothenemus hampei* (ferrari, 1867) (coleoptera: curculionidae): a meta-analysis. *Jurnal Hama Dan Penyakit Tumbuhan Tropika*, 24(1), 58–65. <https://doi.org/10.23960/jhptt.12458-65>
- [18] Mao, G., Tian, Y., Sun, Z., Ou, J., & Xu, H. (2019). Bruceine d isolated from *Brucea javanica* (l.) merr. as a systemic feeding deterrent for three major lepidopteran pests. *Journal of Agricultural and Food Chemistry*, 67(15), 4232–4239. <https://doi.org/10.1021/acs.jafc.8b06511>
- [19] Ayodeji, O. (2023). Biopesticidal activities of three botanicals (*Ageratum conyzoides* l., *Petiveria alliacea* l. and *Hyptis suaveolens* l. poit) against *sitophilus oryzae* l. *Open Access Journal of Science*, 6(1), 61–66. <https://doi.org/10.15406/oajs.2023.06.00194>
- [20] Alao, F., Adebayo, T., & Olaniran, O. (2012). On-farm evaluation of natural toxicants from *Tephrosia vogelii* and *Petiveria alliacea* on megalurothrips *sjoestedti* and apion varium of cowpea (*vigna unguiculata* (l) walp). *Bangladesh Journal of Agricultural Research*, 36(4), 575–582. <https://doi.org/10.3329/bjar.v36i4.11745>
- [21] Fan, Y., Xin, X., Liu, L., Feng, H., Wang, P., Zhang, Y., ... & Gao, D. (2020). Diversity analysis and associated antimicrobial activity of essential oil in *Pyrrosia petiolosa*. *Chemistry & Biodiversity*, 17(12). <https://doi.org/10.1002/cbdv.202000666>
- [22] Khadka, G. (2023). Isolation and characterization of fungal endophytes from *petiveria alliacea* and their antimicrobial activities in south florida. *Microbiology Research*, 14(3), 1470–1482. <https://doi.org/10.3390/microbiolres14030100>
- [23] Hou, Q., Zhang, H., Bao, L., Song, Z., Liu, C., Jiang, Z., & Zheng, Y. (2021). NCs-delivered pesticides: A promising candidate in smart agriculture. *International journal of molecular sciences*, 22(23), 13043. <https://doi.org/10.3390/ijms222313043>
- [24] Sabbour, M. M., & Shadia, E. (2015). Nanotechnology's role in modern pest management. *Nanotechnology for Environmental and Agricultural Applications*, 32–45.
- [25] Azeez, A. M., Lateef, A. A., & Yusoff, M. M. (2017). Insecticidal Activity of Green Synthesized Silver Nanoparticles from *Pelargonium citroneolum* against Citrus Mealybug, *Planococcus citri*. *Plant Protection*, 4(1), 1–10. <https://doi.org/10.5281/zenodo.3952355>
- [26] Lateef, A. A., Azeez, A. M., & Yusoff, M. M. (2018). Synthesis and characterization of silver nanoparticles using *Azadirachta indica* leaf extract and their insecticidal activity against *Aedes aegypti*. *International Journal of Tropical Insect Science*, 38(3), 128–138. <https://doi.org/10.1007/s42690-018-0005-3>
- [27] Kumar, S., Singh, P., & Sharma, R. (2020). Green

- synthesis of silver nanoparticles: An eco-friendly approach for pest management in agriculture. *Journal of Nanobiotechnology*, 18(1), 1-15. <https://doi.org/10.1186/s12951-020-00699-5>
- [28] Lateef, A., Folarin, B. I., Oladejo, S. M., Akinola, P. O., Beukes, L. S., & Gueguim-Kana, E. B. (2018). Characterization, antimicrobial, antioxidant, and anticoagulant activities of silver nanoparticles synthesized from *Petiveria alliacea* L. leaf extract. *Preparative biochemistry and biotechnology*, 48(7), 646-652. <https://doi.org/10.1080/10826068.2018.1479864>
- [29] Sobi, M. A., Usha, D., Umadevi, M., Bindhu, M. R., Sureshkumar, S., Al-Dosary, M. A., ... & Hatamleh, A. A. (2022). Size-dependent antimicrobial activity of *Boerhaavia diffusa* leaf mediated silver nanoparticles. *Journal of King Saud University-Science*, 34(5), 102096. <https://doi.org/10.1016/j.jksus.2022.102096>
- [30] Elegbede, J. A. and Lateef, A. (2019). Green Nanotechnology in Nigeria: The Research Landscape. *Annals of Science and Technology*, 4(2), 6–38. doi:10.2478/ast-2019-0008
- [31] Elegbede, J. A., Lateef, A., Azeez, M. A., Asafa, T. B., Yekeen, T. A., Oladipo, I. C., Adebayo, E.A., Beukes, L.S. & Gueguim-Kana, E. B. (2018). Fungal xylanases-mediated synthesis of silver nanoparticles for catalytic and biomedical applications. *IET nanobiotechnology*, 12(6), 857-863. <https://doi.org/10.1049/iet-nbt.2017.0299>
- [32] Obembe, O. and Ojo, D. (2018). Toxicity and oviposition inhibitory effect of extract and powder of *momordica charantia* leaf against *callosobruchus maculatus* fab. (Coleoptera: chrysomelidae) on stored cowpea seed. *Journal of Bioscience and Biotechnology Discovery*, 3(3), 65-70. <https://doi.org/10.31248/jbbd2018.067>
- [33] Ananda, A. and Rustam, R. (2023). The several of jengkol peel extract (*pithecellobium lobatum* benth) against corn armyworm (*Spodoptera frugiperda* j. e. smith) in the laboratory. *International Journal of Science and Research Archive*, 9(1), 577-583. <https://doi.org/10.30574/ijrsra.2023.9.1.0458>
- [34] Babarinde, S. A., Adeyemi, I. A., & Olatunji, G. B. (2018). The challenge of synthetic insecticide availability in agricultural pest control. *Nigerian Agricultural Journal*, 15(2), 45–62.
- [35] Olaitan, A. F., Abiodun, A. T., & Abiodun, O. O. (2020). Bioactivity and effects of spraying interval of selected plant extracts for control of pre-flowering insect pests of watermelon (*Citrulus lanatus* [Thunb.] Matsum. & Nakai). *Acta agriculturae Slovenica*, 116(1), 107-114. <https://doi.org/10.14720/aas.2020.116.1.1583>
- [36] García-Mateos, M. R., Elizalde Sánchez, E., Espinosa-Robles, P., & Álvarez-Sánchez, M. E. (2023). Toxicity of *Petiveria alliacea* L. on greenhouse whitefly (*Trialeurodes vaporariorum* West.). *Agronomy*, 13(6), 1450. <https://doi.org/10.3390/agronomy13061450>
- [37] Elizalde Sánchez, E., Espinosa-Robles, P., Álvarez-Sánchez, M. E. (2020). Toxicity of *Petiveria alliacea* L. on greenhouse whitefly (*Trialeurodes vaporariorum* West.) under laboratory and greenhouse conditions. *Revista Mexicana de Ciencias Agrícolas*, 11(2), 271-297.
- [38] Cruz-Estrada, A., Gamboa-Angulo, M., Borges-Argáez, R., & Ruíz-Sánchez, E. (2013). Insecticidal effects of plant extracts on immature whitefly, *bemisia tabaci* genn. (hemiptera: aleyroideae). *Electronic Journal of Biotechnology*, 16(1). <https://doi.org/10.2225/vol16-issue1-fulltext-6>
- [39] Adesanya, E., Oyesiku, O., Adesanya, O., Ogunlakin, A., Odugbemi, A., & Egieyeh, S. (2023). Phytochemical components and gc–ms analysis of *petiveria alliacea* l. fractions and volatile oils. *Physical Sciences Reviews*, 9(6), 2239-2251. <https://doi.org/10.1515/psr-2022-0311>
- [40] Mansour, A. H., & Al-Mansoori, A. (2024). Insecticidal activities of biosynthesized silver nanoparticles using *Petiveria alliacea* extracts. *Insects*, 15(7), 469
- [41] Olaniran, O. A., Elegbede, J. A., Lateef, A., Adebayo, T. A., Gueguim-Kana, E. B., Beukes, L. S., Matyumza, N. & Alao, F. O. (2024). Anti-cockroach Activities of Biosynthesized Silver Nanoparticles using *Petiveria alliacea* Extracts. <https://doi.org/10.21203/rs.3.rs-5253110/v1>
- [42] Hesham, H. A., et al. (2023). Concentration-specific effects of nano-based pesticides on pest control. *Journal of Pest Management*, 45(3), 245–260.
- [43] Awad, A. M., El-Sayed, A., & El-Shafey, H. A. (2022). Current and future prospects of “all-organic” nanoinsecticides for sustainable agriculture and global food security. *Frontiers in Nanotechnology*, 3, 1082128. <https://doi.org/10.3389/fnano.2022.1082128>
- [44] Yan, X., Wang, Y., & Zhang, Y. (2019). Nano-enabled pesticides for sustainable agriculture and global food security: A review. *Nano Today*, 28, 100758. <https://doi.org/10.1016/j.nantod.2019.100758>
- [45] Rajna, R., et al. (2019). Efficacy of nano-pesticides: A comparative analysis of application methods. *International Journal of Pest Science*, 14(2), 167–174.
- [46] Sharma, A., Kumar, S., & Tripathi, P. (2019). A facile and rapid method for green synthesis of *achyranthes aspera* stem extract-mediated silver nano-composites with cidal potential against *Aedes aegypti* l.. *Saudi Journal of Biological Sciences*, 26(4), 698-708. <https://doi.org/10.1016/j.sjbs.2017.11.001>
- [47] Wang, J., Zhang, Y., Li, X., & Chen, H. (2022). Applications of nano-insecticides: Contact vs. fumigant methods. *Pest Control Science*, 47(5), 92–107. DOI: 10.1007/s11356-022-18544-7.
- [48] Hashimoto, Y., Sakamoto, H., Asai, H., Yasoshima, M., Lin, H., & Goka, K. (2020). The effect of fumigation with microencapsulated allyl isothiocyanate in a gas-barrier bag against *solenopsis invicta* (hymenoptera: formicidae). *Applied Entomology and Zoology*, 55(3), 345-350. <https://doi.org/10.1007/s13355-020-00684-9>
- [49] El-Saadony, F. M., et al. (2023). Comparative efficacy of biosynthesized silver nanoparticles in pest control. *Nanotechnology in Agriculture*, 15(1), 67–81.
- [50] Gaddam, S. A., Kotakadi, V. S., Subramanyam, G. K., Penchalaneni, J., Challagundla, V. N., Dvr, S. G., & Pasupuleti, V. R. (2021). Multifaceted phyto-genic silver nanoparticles by an insectivorous plant *Drosera spatulata* Labill var. *bakoensis* and its potential therapeutic applications. *Scientific Reports*, 11(1), 21969. <https://doi.org/10.1038/s41598-021-01281-8>