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Green Synthesis, Characterization and Insecticidal Activity of Pelargonium graveolens-Mediated Zinc Oxide Nanoparticles Against the Stored-Product Pest Tribolium confusum

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Abstract

Tribolium confusum Jacquelin du Val (Coleoptera: Tenebrionidae) is one of the most destructive pests of stored grain products worldwide. The present study aimed to synthesize zinc oxide nanoparticles (PG-ZnONPs) using *Pelargonium graveolens* essential oil through an eco-friendly green synthesis approach and to evaluate their insecticidal activity against *T. confusum* adults. The synthesized nanoparticles were characterized using UV–Visible spectroscopy, Fourier-transform infrared spectroscopy (FTIR), scanning electron microscopy (SEM), energy-dispersive X-ray spectroscopy (EDX), and X-ray diffraction (XRD). UV–Vis analysis showed a characteristic absorption peak at 361 nm, confirming the formation of ZnO nanoparticles. SEM observations revealed predominantly quasi-spherical nanoparticles with sizes ranging from 6.7 to 24.6 nm and an average diameter of 14.9 nm. XRD analysis confirmed the crystalline hexagonal wurtzite structure of ZnO, while FTIR spectra demonstrated the involvement of essential oil phytochemicals in nanoparticle stabilization. Contact toxicity bioassays revealed significant insecticidal activity against T. confusum adults, with mortality increasing in a concentration-dependent manner and reaching 86.67% at 150 mg/mL after 24 h of exposure. Probit analysis estimated an LC_{50} value of 5.5 mg/mL. The findings demonstrate that PG-ZnONPs represent a promising eco-friendly alternative to conventional insecticides for the sustainable management of stored-product pests.

1.Introduction

Tribolium confusum (Duval) is one of the most prevalent harmful pests of stored goods and is impervious to a variety of pesticides. (Miloudi, & al 2024). One of the most prevalent pests affecting stored food products on a global scale is the confused flour beetle (*Tribolium confusum* Jacquelin du Val; Coleoptera: Tenebrionidae).

Stored products are subject to numerous attacks by storage pests. Insect coleopterans are common invaders that can cause significant economic losses (Demis and Yenewa, 2022). Despite the frequent utilisation of synthetic pesticides for the management of this pest, the prolonged use of these chemicals has been associated with significant health and environmental concerns (Tiring, G. 2025). Conventional pest control techniques frequently useed chemical pesticides, which present environmental risks and encourage the emergence of resistant pests. (Bhatnagar, S., & al, 2024).

Nanotechnology is a significant field of science with diverse applications, including energy generation, industrial manufacturing processes, and biomedical uses, Nanoparticles, which are the Predominantly preferred nanomaterials that form the basis of nanotechnology, are prepared with different composition, size, shape and surface chemistry to provide new techniques in applications in many different fields (Cimen & al, 2025).

The synthesis of plant-based biological nanoparticles (NPs) is an innovative method in which biological components and their combinations in plant aqueous extracts reduce metal salts, serving a dual function in the manufacturing and stability of metallic NPs (Abdel-Megeed, & al 2025).

The excessive use of chemical pesticides has a detrimental effect on the environment, wildlife and human health,The effectiveness of pesticides has significantly decreased due to insect resistance. Professionals from around the world are working to reduce the usage of dangerous pesticides for disease and pest management by using eco-friendly techniques. (Karthik Raja, & al 2025). Misuse of synthetic pesticides has resulted in insect pest resistance and detrimental environmental effects, emphasising the pressing need for sustainable alternatives in integrated pest management (Martínez-Cisterna, & al 2025). In recent years, nanotechnology has become increasingly important in the fields of environmental and agricultural science. Due to their low cost, availability and simple synthesis, nanoparticles have attracted strong interest for their insecticidal potential. Various inorganic and organic nanoparticles, such as those made from silver, gold, silica, titanium dioxide, zinc oxide and iron, have been successfully tested against numerous harmful arthropods and agricultural pests (Saranya, S., & al 2020).

Green-engineered nanoparticles are non-toxic, environmentally benign, biocompatible and biodegradable, and undergo a single-step bioreduction process, unlike their chemically manufactured counterparts. Green technology uses phytoconstituents, such as alkaloids, terpenoids, flavonoids, phenols, tannins and glycosides, to stabilise the nanoparticles by acting as capping or reducing agents (Meena, K., & al 2025). Nanotechnology applications in sustainable agriculture have tremendous potential for managing insect pests, thanks to controlled and targeted release mechanisms. Smaller sizes of nanoparticles ensure proper coverage of the pest surface, resulting in more effective action (Karthik Raja, & al 2025).

Although numerous studies have reported the insecticidal potential of metal nanoparticles against agricultural pests, limited information is available regarding the use of Pelargonium graveolens essential oil-mediated ZnO nanoparticles against *Tribolium confusum*. Moreover, the synergistic contribution of bioactive compounds present in *P. graveolens* essential oil during nanoparticle synthesis and pest control remains insufficiently explored. Therefore, developing environmentally friendly nanopesticides based on essential oils represents a promising strategy for sustainable stored-product protection

This study aims to synthesis green nanoparticles using *Pelargonium graveolens* (geranium) essential oil, and to evaluate their insecticidal toxicity against *T. confusum* In addition to assessing their bioefficacy, the synthesised nanoparticles were fully characterised using physicochemical techniques, including UV–Vis spectroscopy, Fourier-transform infrared spectroscopy (FTIR), X-ray diffraction (XRD) and scanning electron microscopy (SEM), in order to determine their optical properties, functional groups, crystalline structure and morphological features. By combining a plant-derived bioactive compound with an environmentally friendly synthesis approach, this study aims to improve the performance of insecticides while reducing the ecological and health risks associated with conventional pesticides. The study examines the mortality response of *T. confusum* to different concentrations of nanoparticles and exposure periods, highlighting their potential as a sustainable alternative for managing stored-product pests.

2.Material and Methods

2.1. Essential oil extraction

Aromatic plant materials, including *P. graveolens*, was collected from Bordj Bou Arréridj, Algeria, in April 2023. The aerial parts of these plants were naturally dried at 24–25°C for four days. All EOs were obtained from the aerial parts by hydro distillation as described by Elyemni *et al*. (2019)

2.2. GC-MS analysis

The analysis method was performed as described by Amokrane (2023); Chromatographic analyses were performed using an Agilent 6890 Plus gas chromatography (GC) system coupled to a 5975-mass selective detector operating in electron-impact mode. Volatile constituents were separated on an HP-5MS apolar capillary column (30 m × 0.25 mm, 0.25 µm film; 95% dimethylpolysiloxane/5% phenyl). Hydrogen was used as the carrier gas at a constant flow rate of 1 ml/min. Samples (0.02 µL) were injected at 250°C in split mode (1:80). The oven programme began at 60°C (held for eight minutes), then increased by 2°C per minute to 250°C, where it was held for ten minutes, resulting in a total run time of 113 minutes. Mass spectra were acquired using electron ionisation at 70 eV, with a source temperature of 230°C and an interface temperature of 280°C. Compound identification was achieved by matching the recorded spectra with those in the reference libraries (Wiley, NIST and PAST) and with published data. Relative abundances were calculated from normalised peak areas without correction factors.

2.3. Insect Rearing

The experiment was conducted at Bordj Bou Arréridj University, which is located in Algeria. A mass culture of *T. confusum*, was established in the laboratory under insecticide-free conditions. The beetles were reared in 1.5 litre plastic jars containing a mixture of wheat flour and wheat grains (8:2 w/w). The culture was maintained under controlled conditions (27 ± 2°C and 65 ± 5% relative humidity) in complete darkness. Every seven days, newly emerged adults were separated from the culture medium using a 60 mm mesh sieve and transferred to fresh jars. For toxicity and repellency bioassays, adult beetles aged 1–7 days old were used.

2.4. Green Synthesis of Nanoparticles

Zinc acetate dihydrate (Zn [OOCCH3]2· 2H2O), (0.1 M) was dissolved in distilled water and heated to 70°C. An essential oil emulsion (3% v/v, dissolved in acetone) was added under drop-by-drop continuous stirring, and the pH was adjusted to 10 using 1 M NaOH to induce precipitation. the final solution was centrifuged for 15 mins at 8000 rpm, the precipitate was cleaned many times with deionized water to remove the impurities dried at 80°C, and calcined at 400°C for 2 h to obtain crystalline ZnO. The PG-ZnONPs are prepared according to the previously described method with some modifications (Selim, Y. A & al 2020).

2.5. Characterization of Nanoparticles

The characterisation of the synthesised zinc oxid nanoparticles (PG-ZnONPs) was conducted through the utilisation of UV-Vis spectroscopy, a technique employed to ascertain the optical properties of the

fabricated PG-ZnONPs across a 200–800 nm scan range. The morphology and size distribution of the nanoparticles were examined by Scanning Electron Microscopy (SEM). The analysis of functional groups presents on the nanoparticle surface, which are responsible for reduction and stabilisation, was conducted using Fourier-transform infrared spectroscopy (FTIR). Energy-dispersive X-ray spectroscopy (EDX) was performed in order to study the morphology and composition of the nanoparticles.

2.6. Contact Toxicity Assay against *Tribolium confusum* J

Contact toxicity on *Tribolium confusum J* was carried out according to the protocol described by BenSlimane et al. (2014), with minor modifications. Twenty individual insects were placed in Petri dishes, which were then sprayed with 500 µl of PG-ZnONPs with different dilutions, namely 150, 125, 100, 75 and 50 mg/ml. five replicates were conducted for every PG-ZnONPs and for each concentration. The control was represented by a spray containing 3% Tween. The death rates were noted 72 hours post-treatment and adjusted according to Abbott's formula (Abbott 1925):

MC%= ((M-Mt) *100) / (100-Mt)

where: *MC* – corrected mortality rate; *M* – mortality rate in treated series; *M*t – mortality rate observed among control series.

The insects were deemed lifeless when no signs of movement were observed. Half-maximal lethal concentration (LC_{50}) values were determined through probit analysis.

2.7. Statistical Analysis

ANOVA for mortality data. at the 5% probabil- ity level, followed by Tukey's post hoc test Post-hoc multiple comparison tests (Tukey/Duncan). Probit analysis for LC_{50} and LT_{50}. Every analysis was carried out with the statistical software ,Version 3.6.1 of RStudio 1.2.5019-R.

3.Results

3.1. Essential oils composition

GC-MS analysis of the EOs extracted from *P. graveolens*, (Fig. 1) revealed their major chemical constituents was primarily composed of β-Citronellol (21.46%) and Trans-Geraniol (20.11%), both monoterpene alcohols, which dominate the aroma as major compounds. Several monoterpene hydrocarbons, such as p-Cymene (1.07%) and α-Pinene dimer (0.68%), were detected as a minor compound. The essential oil also contains several sesquiterpene hydrocarbons, with compounds like β-Bourbonene (0.89%), α-Copaene (0.32%), and Trans-Caryophyllene (0.46%).

3.2. Characterization of green synthesized PG-ZnONPs

All the characterization analyses indicate the evolution of zinc oxide nanoparticles (PG-ZnONPs) synthesized from *P. graveolens* essential oil. UV–Vis spectra of the PG-ZnONPs produced using

P.graveolens essential oil are shown in Fig. 2(a). The strong absorption peak was observed at around 361 nm, which is indicative of the formation of PG-ZnONPs. The efficient synthesis of zinc oxide nanoparticles (PG-ZnONPs) by *Pelargonium graveolens* essential oil was affirmed through several characterisations. It appeared the characteristic absorption peak at 360–380 nm light for UV–Visible spectrum.The morphology and elemental composition of the obtained nanoparticles were examined using scanning electron microscopy (SEM) coupled with energy-dispersive X-ray spectroscopy (EDS) as illustrated in Fig. 4.

SEM micrographs recorded at a magnification of 60,000× revealed the formation of predominantly quasi-spherical ZnO nanoparticles with a relatively homogeneous nanoscale distribution. A slight degree of particle agglomeration was observed, which is commonly reported for metal oxide nanoparticles synthesized via green methods. This behavior can be attributed to the presence of bioactive compounds from P. graveolens essential oil, which act as reducing and stabilizing agents and form an organic capping layer on the nanoparticle Surface (Fig. 3).

The crystalline nature and average grain size of green synthesized (PG-ZnONPs) were confirmed using the X-Ray diffraction technique (XRD) using Cu-Kα radiation (Fig. 5). The XRD spectrum showed peaks at 31.7, 34.4, 36.2, 47.5, 56.6, 62.8, degrees 2θ attributed to the 100, 002, 101, 102, 110, 103, 112, crystal planes, respectively, confirms the formation of hexagonal wurtzite Zno phase (JCPDS No. 36-1451) without any secondary phases, indicating high purity of the synthesized material. The mean crystalline size of (PG-ZnONPs) ranged from 6.7 to 24.6 nm with an average value of approximately 14.9 nm. Fourier transform infrared spectroscopy (FT-IR) revealed the presence of a characteristic peak at approximately 520 cm − 1 which indicated the formation of Zn□O nanostructure illustrated in Fig. 2 *(b)*, The peaks observed at around 3400 cm − 1 could be corresponding to O–H and N–H functional groups indicating the groups originating from biomolecules compounds, the peak projected around 2920 cm − 1 corresponds to the C-H stretching, How- ever the peak projected around 1630 cm − 1 corresponds to the C = O stretching vibration indicting the indolent of proteinaceous biomolecules, the band detected around 1400 cm − 1 is assigned to the C − N stretching vibration of carboxylate .FTIR analysis confirmed successful interaction/adsorption of essential oil compounds onto ZnO nanoparticle surface.

3.3. Contact Toxicity Assay of green synthesized PG-ZnONPs

The mortality rates induced by the five dilutions of PG-ZnONPs against *T. confusum* DuVal individuals is determined using the contact test. (Fig. 6). The intensities of the mortality rates varied according to the duration of exposure (F (4) = 16,291; P = 0.0000*), and the applied concentration (F (4) = 6,541; P = 0.0000*). After 24 hours, CM rates of 20% at 50mg/ml to 86.67% at 150 mg/ml. The corrected mortality rate increased gradually with increasing PG-ZnONPs concentrations were obtained 76,67% to 100 mg/ml and 86,67% à150 mg/ml. After 72 hours. Probit analysis identified two lethal concentration levels: CL_{50}=5,5mg/ml (χ^2=2,671).

4.Discussion

The aim of this study was to develop a green and eco-friendly method or the synthesis of zinc oxide nanoparticles using of essential oils in the context of green chemistry has been demonstrated to function as a reducing and stabilising agent. and to evaluate their insecticidal activity against *Tribolium* confusum. The physicochemical analyses (UV–Vis, FTIR and GC–MS) results it can be confirmed that *P. graveolens* oil essential consists of several phytochemicals including; β-Citronellol (21.46%) and Trans-Geraniol (20.11%), both monoterpene alcohols which acted as reducing and stabilizing agent for successful phytosynthesis of PG-ZnONPs. Also (XRD, SEM, etc.) confirmed the formation of predominantly quasi-spherical ZnO nanoparticles with a relatively homogeneous nanoscale distribution, indicating that the bioactive compounds present in the essential oil played a significant role in nucleation and growth control. which was confirmed with the results of researchers (Naiel& al 2022). The insecticidal activity of PG-ZnONPs may be attributed to their small particle size and high surface-area-to-volume ratio, which facilitate penetration through the insect cuticle and enhance cellular interactions. ZnO nanoparticles are known to induce the generation of reactive oxygen species (ROS), leading to oxidative stress, membrane disruption, protein denaturation, and eventual insect mortality. Furthermore, the presence of bioactive monoterpene alcohols such as citronellol and geraniol from P. graveolens essential oil may contribute synergistically to the observed toxicity through neurotoxic and repellent effects.The morphological properties observed in this study, characterized by small particle size ranged from 6.7 to 24.6 nm, with an average value of approximately 14.9 nm, confirming the nanocrystalline nature of the synthesized ZnO,are comparable to those reported by other authors who used plant extracts of Lavender (Limonium pruinosum L.for ZnO nanoparticles. (ZnO Nps) synthesis was showed that the obtained ZnO NPs are crystalline in nature. The EDX results confirmed the purity of the obtained ZnO NPs and the presence of Zinc in its oxide (Naiel, & al 2022). Regarding The results further show that, the biosynthesized ZnO nanoparticles exhibited significant toxicity against *Tribolium castaneum* the mortality rates varied according to the duration of exposure ($F(4) = 16,291$; $P = 0.0000^*$), and the applied concentration ($F(4) = 6,541$; $P = 0.0000^*$), showing a clear dose-dependent increase in mortality. These results are consistent with previous studies demonstrating, that High mortality of Bemisiatabaci was observed in the100 lg/mL of the Solanum melongena (SM-AgNPs) treated concentrations (Pushparaj & al 2023). The LC50 values of 0.246 and 0.215 g/kg, respectively, indicate that C. sinensis essential oil and its EC formulation have insecticidal action against T. castaneum. In comparison to ZnO NPs alone or bulk ZnO, the ZnO with essential oil NPs formulation resulted in 100% mortality at lower concentrations and shorter exposure durations (El-Bakry& al 2025) .These results could be due to their smaller size, larger surface area, and higher reactivity, which can cause oxidative stress and insect cell damage(Król& al 2017). Our results showed that CM levels of *Tribolium confisum* adult varied from 20% at 50 mg/ml to 86.67% at 150 mg/ml of PG-ZnONPs after 24h .In contrast, some authors have reported insecticidal effects when using plant extracts alone or when synthesizing Copper nanoparticles derived from Acacia cornigera exhibited significantly greater insecticidal efficacy against *Tribolium castaneum*, achieving a 90% mortality rate, compared to the 76.6% mortality rate resulting from copper nanoparticles sourced from Annona purpurea (Solorzano ,2024).

Conclusion

This study successfully demonstrated the green synthesis of zinc oxide nanoparticles (PG-ZnONPs) using Pelargonium graveolens essential oil as a natural reducing, capping, and stabilizing agent. Physicochemical characterization confirmed the successful formation of highly crystalline hexagonal wurtzite ZnO nanoparticles with a predominantly quasi-spherical morphology and an average particle size of approximately 14.9 nm. The presence of phytochemical functional groups identified by FTIR suggests that essential oil constituents contributed to nanoparticle stabilization during biosynthesis.

The biosynthesized PG-ZnONPs exhibited remarkable insecticidal activity against Tribolium confusum, with mortality increasing in a concentration-dependent manner and reaching 86.67% at the highest tested concentration after 24 h of exposure. The low LC_{50} value further demonstrates the high toxic efficiency of the nanoparticles, highlighting their potential as an effective alternative to conventional synthetic insecticides.

Overall, the integration of nanotechnology with plant-derived bioactive compounds offers a promising and environmentally sustainable strategy for stored-product pest management. Green-synthesized PG-ZnONPs could contribute to reducing reliance on synthetic pesticides while supporting safer grain preservation and improved food security. Nevertheless, additional investigations on their long-term stability, persistence under storage conditions, safety toward non-target organisms, and field-scale performance are recommended before large-scale practical application.

Declarations

Conflicts of interest

The authors declare no conflict of interest.

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Contributions of authors:

“A.H. Investigation, Writing – Original Draft; N.B. Writing – Review & Editing, Funding Acquisition, Resources; A.S. Conceptualization, Methodology, Data Curation; Investigation, Formal Analysis, Software; S. Validation, Formal Analysis; NG. B. Resources; H.G. Supervision, Visualization ;F.B. Project Administration. All authors reviewed the manuscript.”

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Figures

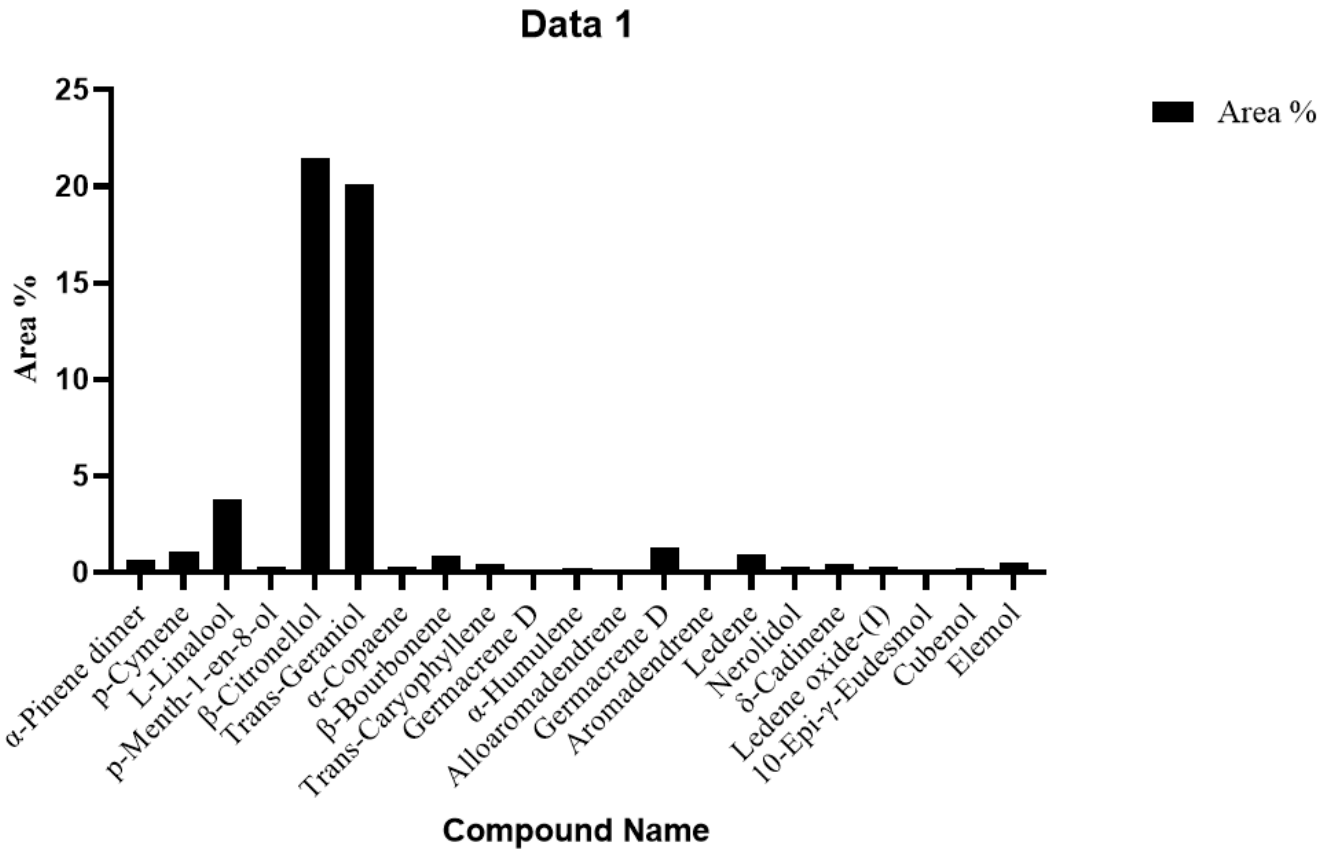


Figure 1

Compounds detected in the essential oil obtained from P. graveolensleaves

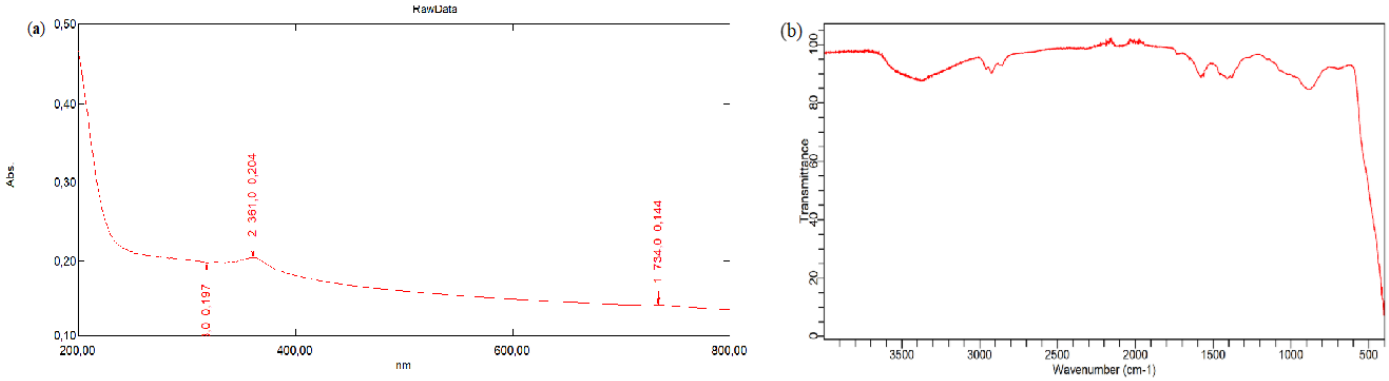


Figure 2

(a) UV–Vis spectrum; (b) FT-IR of PG-ZnONPs

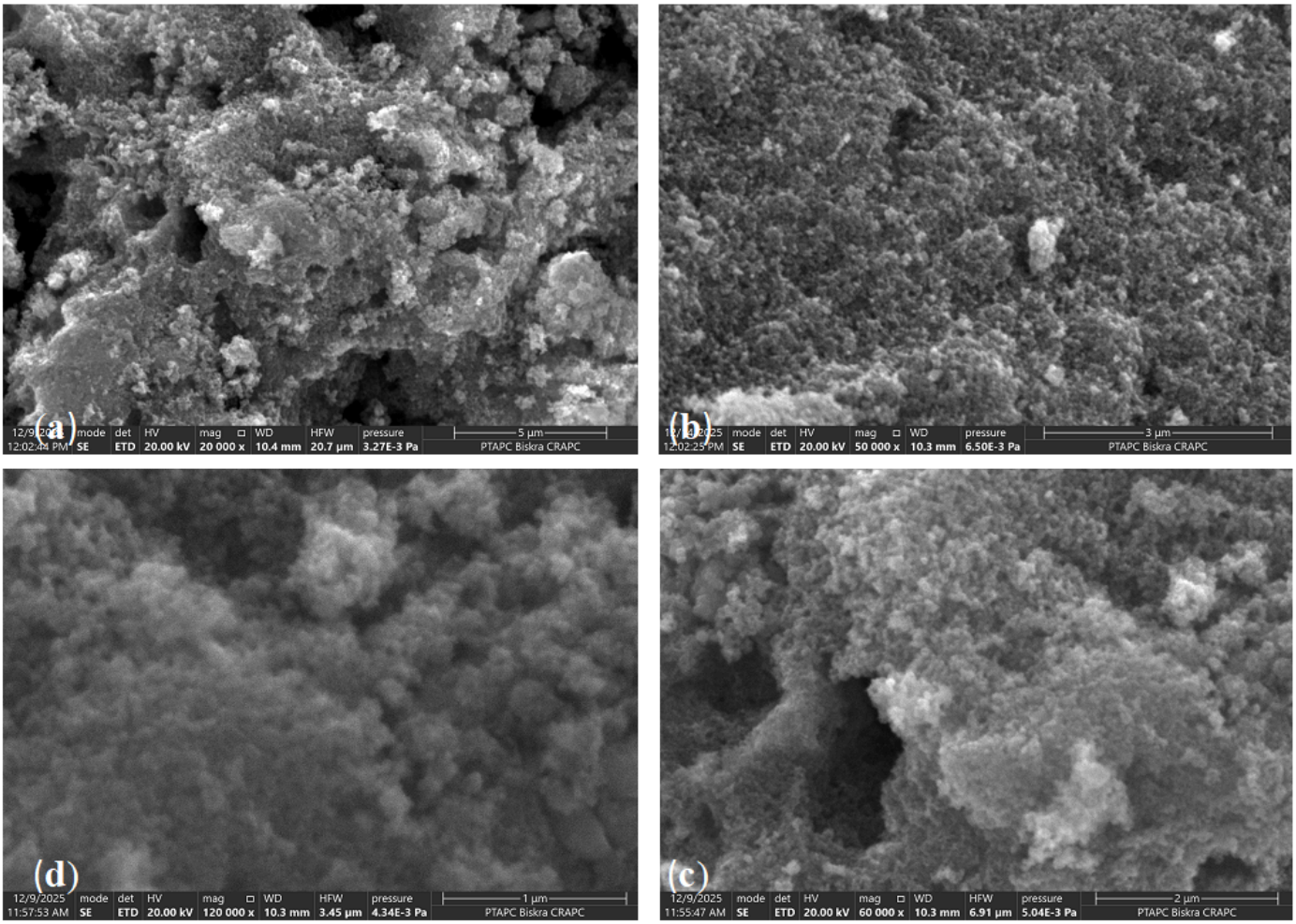


Figure 3

SEM image of biosynthesized PG-ZnONPs nanoparticles at different magnifications (a) 10000× , (b) 50 000× , (c) 60 000× , (d) 120 000×.

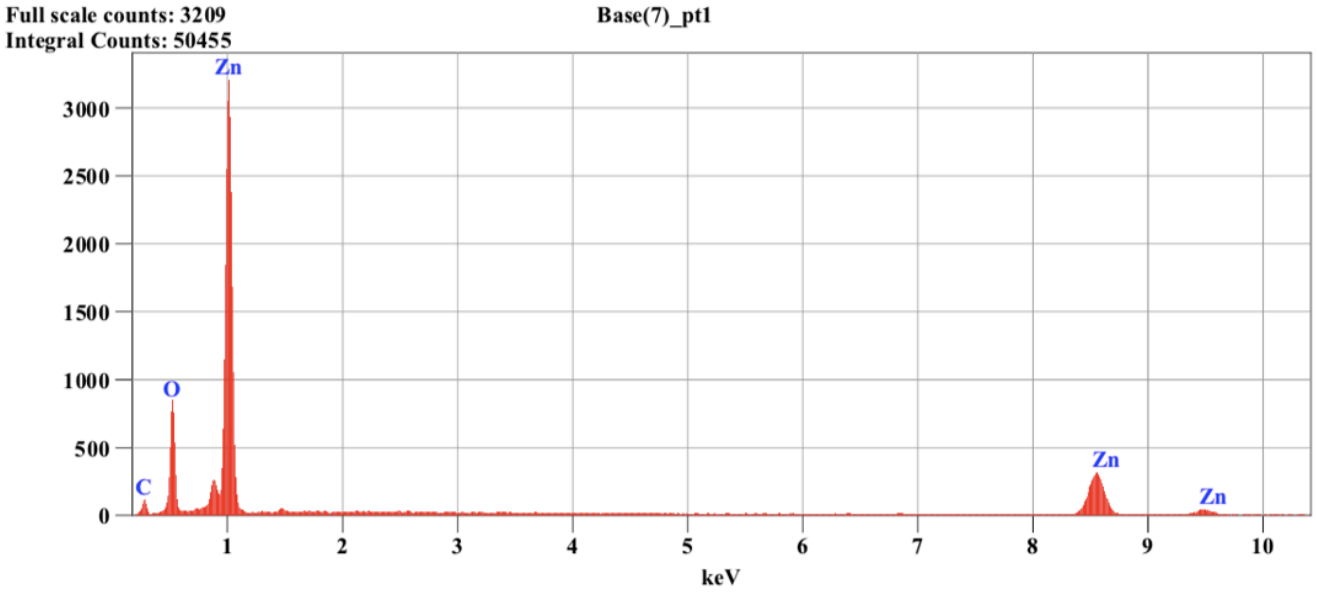


Figure 4

EDX of the phytosynthesized PG-ZnONPs

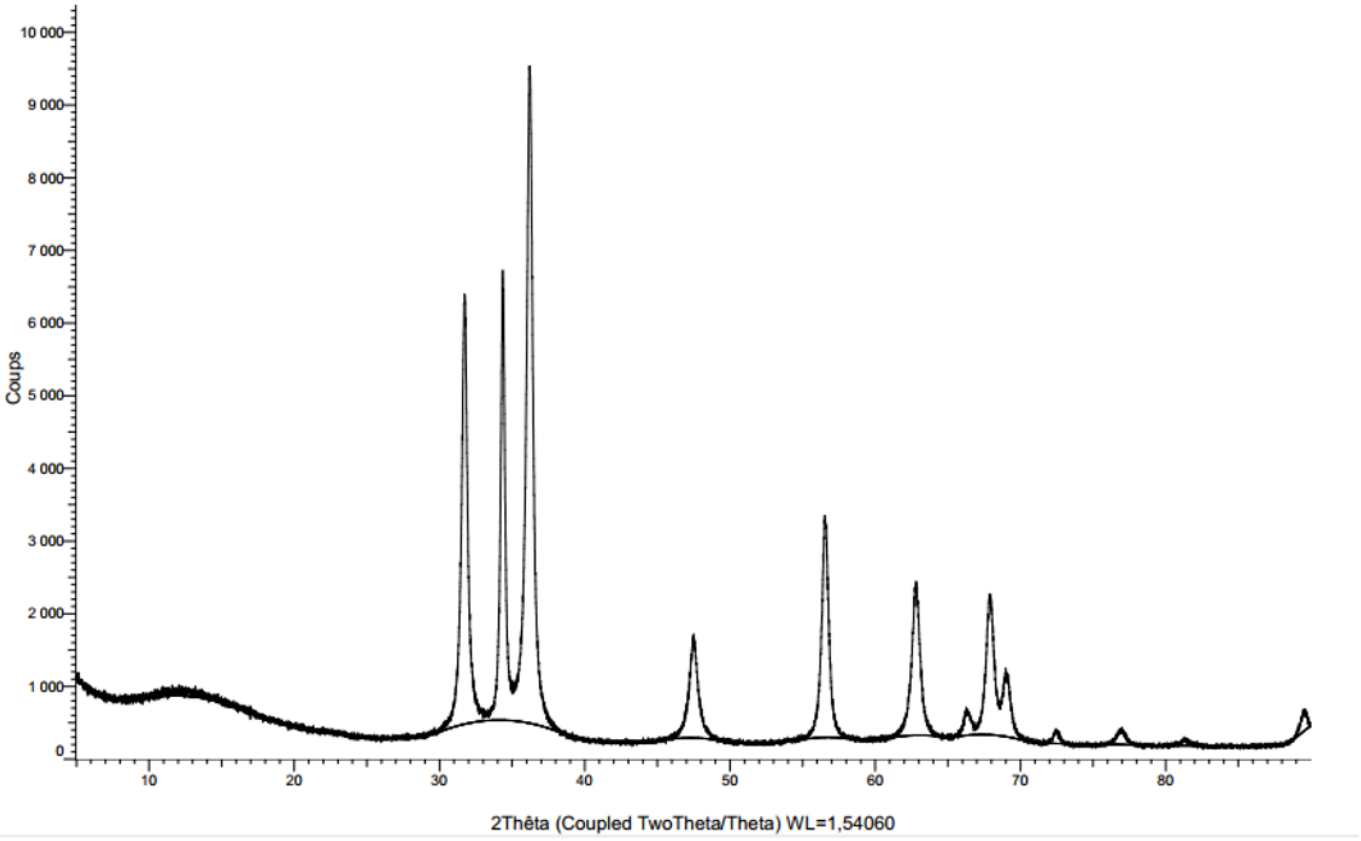


Figure 5

XRD pattern of PG-ZnONPs

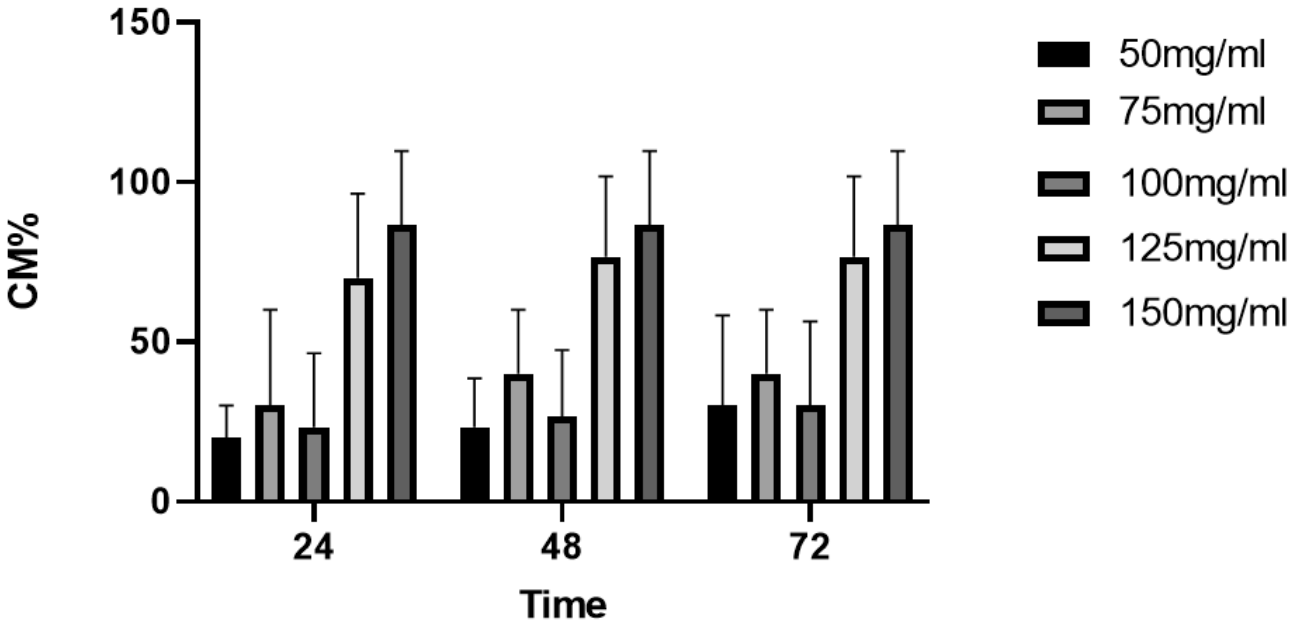


Figure 6

corrected mortality rate of Tribolium confusum DuVal adults exposed to PG-ZnONPs