Suez Canal University Faculty of Science Ismailia



Biochemical and Physiological Impacts of Some Nanoparticles For Using As Alternative Insecticides

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IV-SUMMARY Summary

The current study aimed at preparing CuO and ZnO nanoparticles that can be used as alternative effective, ecofriendly and economic insecticides against the 4th instar of *Schistocereca gregaria* as model insect and monitoring physiological, biochemical and ultra structural changes resulted after treatments with the prepared CuO and ZnO nanoparticles.

Standard procedures were followed to synthesize CuO NPs and ZnO NPs via chemical precipitation method and calcination method respectively. The formation of ZnO and CuO nanoparticles were investigated and confirmed by instrumental analysis and physical measurements such as Ultra violet-visible Spectrophotometer (UV-Vis), Fourier-Transform Infrared (FTIR), X-Ray diffraction analysis, Energy Dispersive X-ray (EDX),Scanning Electron Microscope (SEM), Dynamic light scattering (DLS) and Transmission Electron Microscope Imaging (TEM).

The size distribution of CuO nanoparticles was around 35 nm the actual size of nanoparticles was estimated from the TEM micrograph. Most of the nanoparticles have an avarege size of 20 nm and are nearly uniform in size.

The surface area measured by BET showed that the surface area of CuO NPs is $23m^2/g$.

The TEM data of ZnO NPs showed that more than 50% of particles were in range (7 nm up to 35 nm) and the largest percentage was at a size of 35nm. Also the particles are found to be spherical. The surface area measured by BET showed that the surface area of ZnO NPs is $28 \text{ m}^2/\text{g}$.

Toxicological investigations

The insecticidal efficacy of synthesized ZnO NPs and CuO NPs against the 4th instar of *Schistocerca gregaria* was demonstrated after 7 days of exposure to different concentrations. The two used compounds stated satisfying toxicity levels. CuO NPs achieved highly toxicity compared with ZnO NPs throughout the study and examination days. Where the results were recorded LC₅₀ for CuO NPs (0.487%) and ZnO NPs LC₅₀ was (9.822%). These results stated that more toxicity for CuO NPs than ZnO NPs.

The LT_{50} of sub-lethal concentration (0.25%) of CuO NPs and ZnO NPS against 4th nymphs recorded after 10.994, 21.148 days, respectively. Furthermore, that clued the CuO NPs considered an acute toxicant while ZnO NPs maybe has chronic toxicity. These results lead to those ZnO nanoparticles may result in a residual effect more than CuO nanoparticles.

Biological investigations

The effect of sub-lethal concentration of NPs on nymphal mortality was initially observed during 4th instar where both ZnO NPs and CuO NPs resulted in considerable mortality compared to untreated controls. Mortality rates attained 10.34%, 16.67% respectively, whereas the residual mortality recorded during the 5th instar didn't exceed 12% for CuO NPs and 4.4% for ZnO NPs compared to untreated controls

Furthermore, the effect of NPs on Nymphal duration was verified also. At ZnO NPs treatment the duration of nymphal instars was longer as insects developed into a more advanced stage. While exposure to CuO NPs resulted in reversed findings on the duration of nymphal instars

Additional findings were reported that nymphal development was affected by treatments with ZnO NPs which revealed a notable deficient molting during development from 4th instar to 5th instar reached 12.4% of survival nymphs likewise, 18.5% deficient molting recorded in development from 5th to adult stage. No deficient development was recorded with CuO NPs treatment.

Results reveal the effects of the two NPs on the nymphal metamorphosis into the adult stage after treatment of 4th instar nymphs whereas the adult emergence decreased significantly with nanoZnO treatment recording 82.20% when the nanoCuO didn't show any effects.

In connection with the impaired adult morphogenesis of *S. gregaria* treated by the two tested nanoparticles only 16.66 % adult deformities were recorded at the treatment of ZnO NPs but it failed to affect the morphogenesis at other (CuO NPs) as well as the two controls.

The treatment of 4th instar nymphs with ZnO NPs an important solitary effect was exhibited because 13.33% of the completely emerged nymphes appeared with some symptoms of the solitary phase as well as the albino phase. Neither solitary nor albino effects were recorded after the treatment of 4th instar nymphs by CuO NPs and controls.

Data exiguously revealed that ZnO NPs prohibited the maturation of *S*. *gregaria* thought remarkably prolonged duration (27.305 day), On the other hand, CuO NPs treatment slightly prohibited with non-significant prolonged duration (25.882 days) against positive control with tween 0.1%. (25.603 days).

Biochemical effect

To understand the mechanism of the toxic effect of tested nanoparticles Production of specific ROS and enzymes involved in the occurrence of oxidative stress were evaluated.

The activity of AChE. enzyme showed a marked decrease in both tested nanoparticle where the enzyme activity with ZnO NPs reached (4.781 U/gm), and (9.41 U/gm) with CuO NPs treatment. Comparing to the negative and positive control that recorded (22.48, 15.06 U/gm.)

Phenol oxidase enzyme activity has increased significantly in both treatments as well as the positive control with tween that recorded (2048U/g). Where both ZnO NPs recorded result (2352.2U/g) and CuO NPs (3734.02U/g) respectively compared with negative control (484.76U/g)

GST activity for ZnO NPs treatment was mildly increased compared to Control-tween (0.01%) as shown in the result table (447.35, 341.57U/g) respectively, where CuO NPs effect on the same enzyme was more than four-fold compared to a positive control (1430.90, 341.57U/g).

Moreover, the increasing of lipid peroxidation were stated for the two compounds CuO NPs (228.41nmol/g) and ZnO NPs (124.39nmol/g)

Also, the results of the activity of the CAT enzyme decreased in all the groups. ZnO NPs (248.35U/g) compared to the negative and positive control (774.45, 443U/g) respectively, on the contrary CuO NPs.

On the other hand, SOD activity was affected positively by CuO NPs more than ZnO NPs as a clear difference between the two nanocompounds compared to control samples especially negative control

Genotoxic effects and DNA damages

The results of the comet assay for DNA strand breaks in the nuclei of the brain cells of *schistocerica gregaria* indicated a significant increase in DNA damage in the two treated samples by two prepared nanocompounds as compared to the reference positive either negative samples

Data revealed that the numbers of DNA damaged cells categorized in class (3, 4, and 5) were also greater in the case of the sample treated with CuO NPs compared to the reference ones.

The second nanocompounds (ZnO NPs) showed significant damage for nuclei and so apoptosis for cells but less than copper oxide nanoparticles.

Finally, the author likes to confirm that the findings will throw some light on the effect of using nanoparticles as alternatives to conventional insecticides, besides its usage as fertilizers.