

Copper nanoparticles: Insecticidal action, resistance management and effect on endosymbiont abundance in olive fruit fly *Bactrocera oleae*

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Technical University of Crete

EGU24- 2545
Session HS8.1.7
Board Number A.56

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Abstract

The effectiveness of copper containing nanoparticles (Cu/CuO-NPs) against insecticide-resistant olive fruit flies (*Bactrocera oleae*) and their impact on the insect's reproductive and endosymbiotic parameters were evaluated. The insecticidal activity of both nano and bulk copper [Cu(OH)₂] was comparable or greater than that of the reference insecticide deltamethrin at recommended doses as revealed by feeding experiments. A significant synergistic effect between Cu-NPs or CuO-NPs and deltamethrin was observed in terms of adult mortality. Furthermore, the deltamethrin+Cu-NPs combination decreased the total number of offspring as compared with the untreated control. The above combination also significantly decreased the mean number of stings, pupae, female and total number offspring of the surviving female, compared to deltamethrin applied alone. The abundance of the *Candidatus Erwinia dadicola* - a *B. oleae* larvae bacterial gut endosymbiont- was adversely affected by bulk and nanosized copper. Concluding, Cu-NPs have a great potential to control insecticide-resistant *B. oleae* populations by reducing adult and larval survival and fecundity and provide the means for reducing the environmental footprint of pesticides by minimizing their required doses.

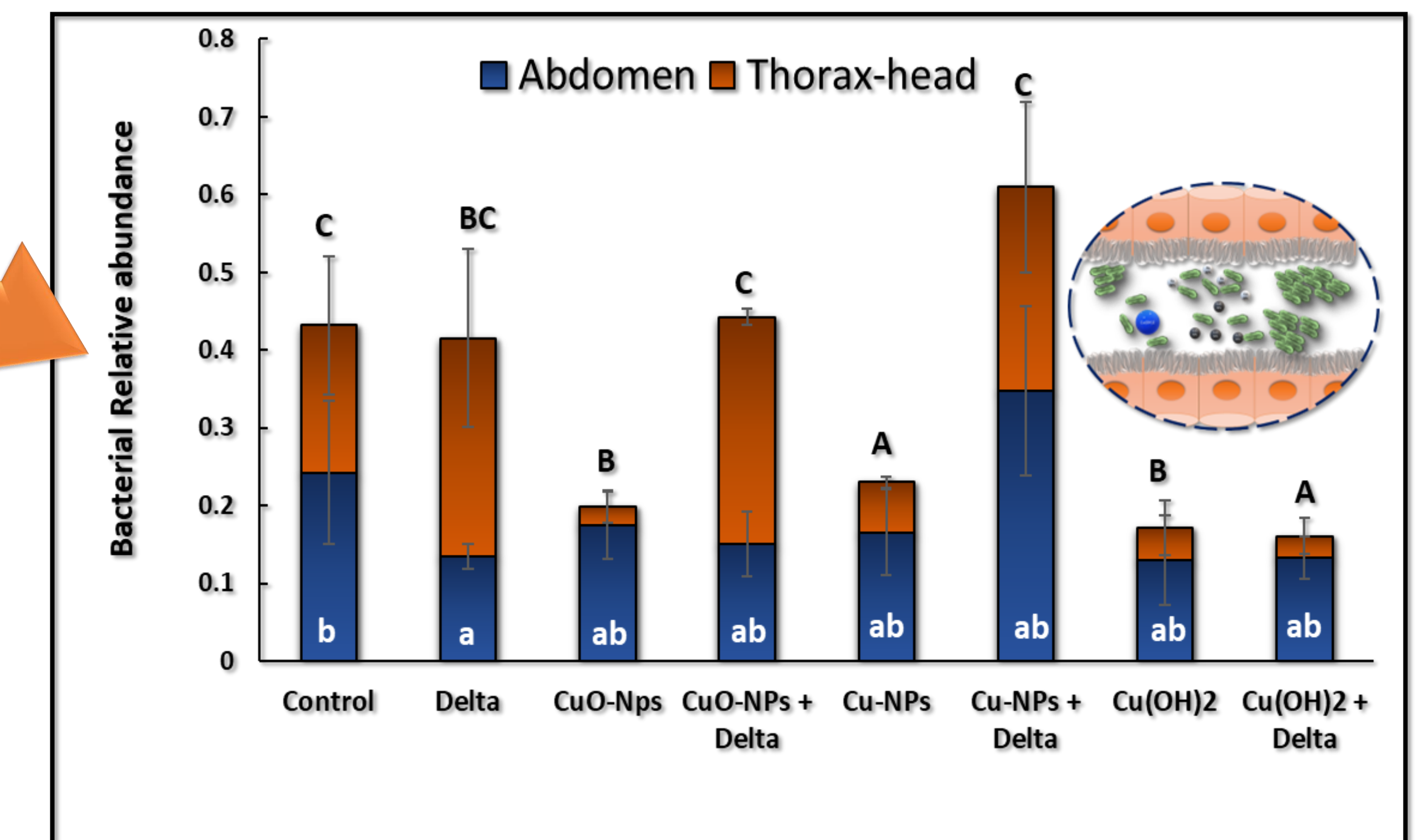
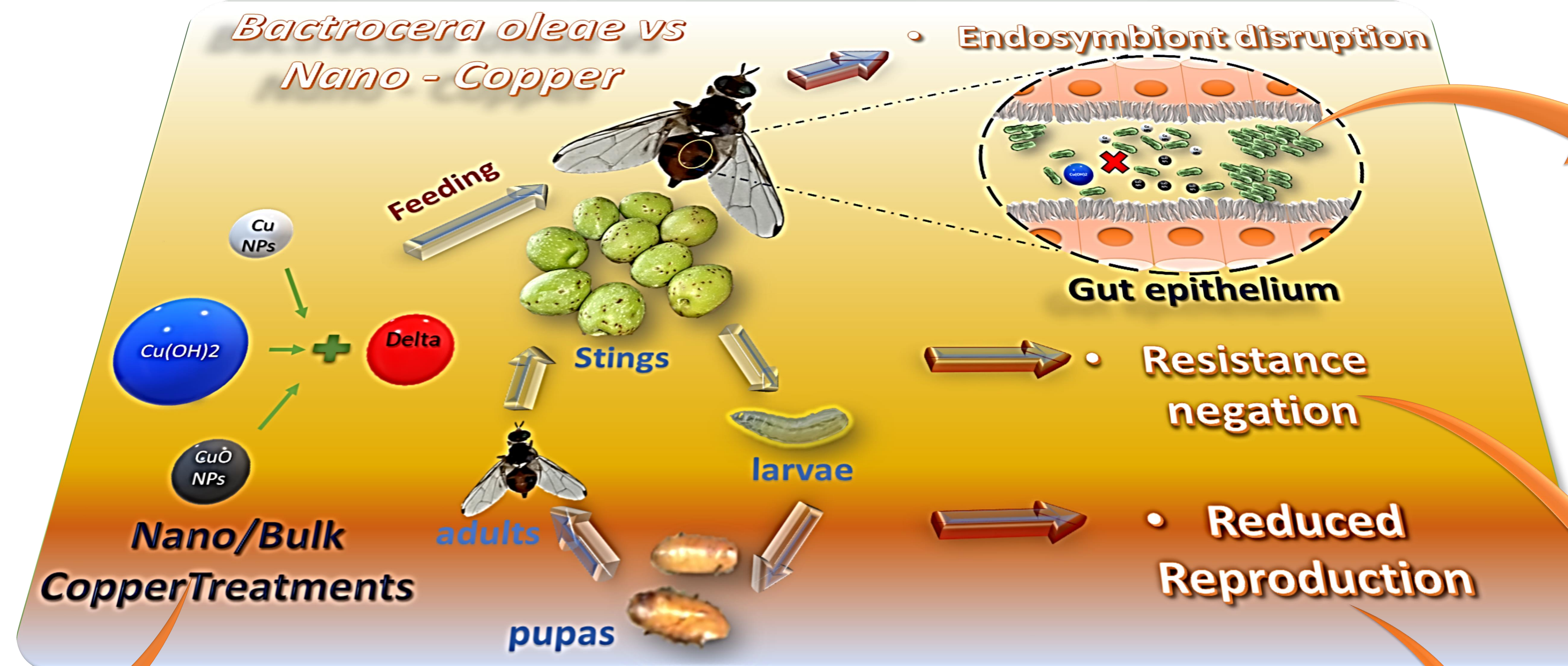


Figure 4. Relative abundance of 'Ca. E. dadicola' in the thorax-head and abdomen parts of *B. oleae* adults treated with deltamethrin (Delta), bulk or nanosized copper and their combinations.

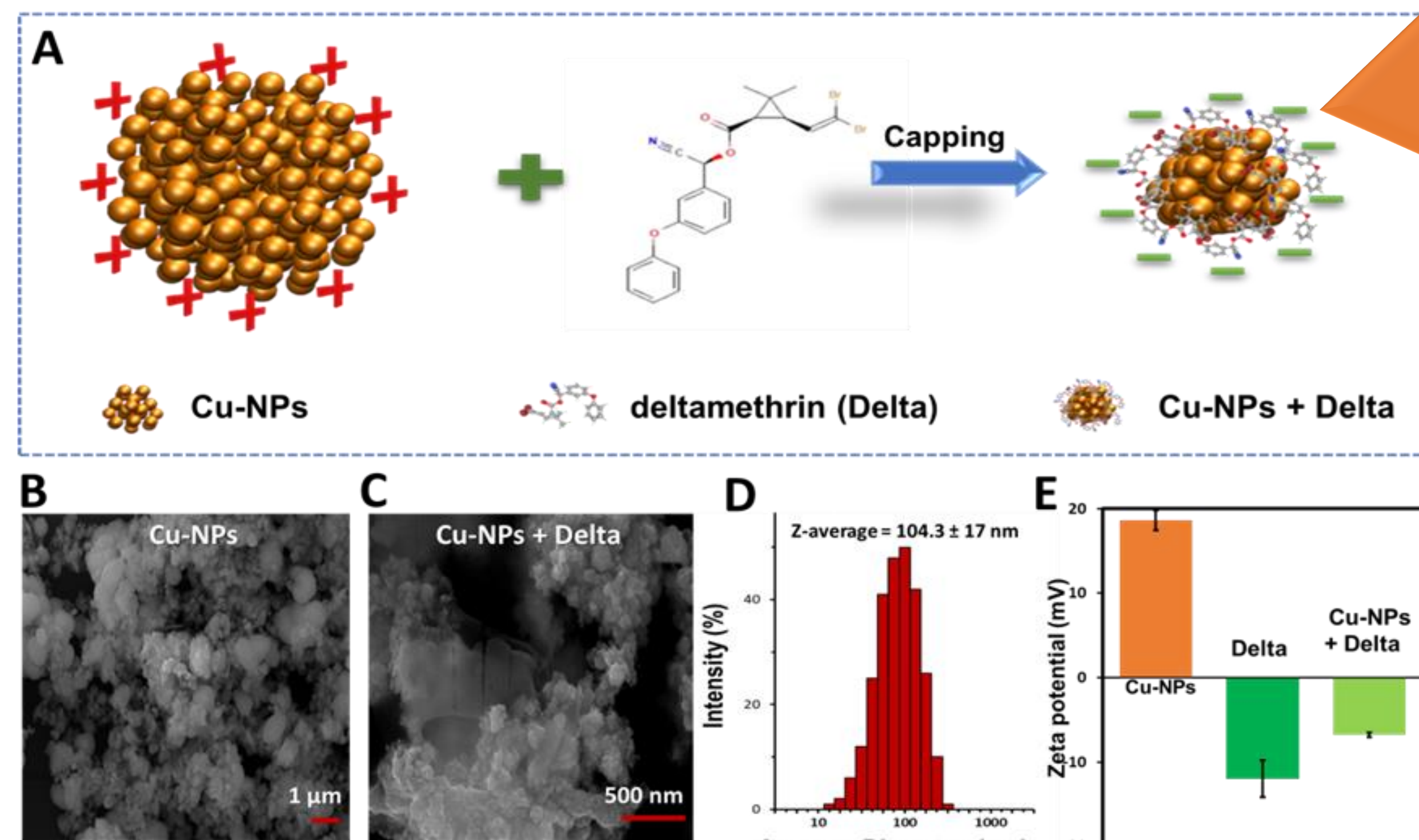


Figure 1: Characterization of copper nanoparticles and their mixture with deltamethrin (Delta). A) Capping effect and size reduction when Delta is combined with Cu-NPs. B) Representative SEM image of Cu-NPs. C) Representative SEM image of Cu-NPs+Delta. D) Particle size distribution of Cu-NPs+Delta. E) Zeta potential characterization of Cu-NPs, Delta and their combination.

Funding
The study was co-financed by the European Regional Development Fund of the European Union and Greek national funds through the Rural Development Program (RDP/IIAA) 2014 – 2020, under the call "Cooperation for environmental projects, environmental practices and actions for climate change" (project code: M16SYN2-00354).

Materials and Methods

Nanoparticles

NPs and reagents were purchased from Sigma Aldrich, MO, USA. Cu-NPs and CuO-NPs 1000 µg/mL suspensions were prepared and then combined with 100 µg/mL deltamethrin suspensions to acquire the respective samples for electron microscopy. Prepared samples were dried in a Critical Point Dryer (BAL-TEC 030) and observed in the Scanning Electron Microscope JSM-IT700HR (20 kV) (Fig.1).

B. oleae sampling and sensitivity

Infested olive fruits (cv Koroneiki) (with visible oviposition sites marks and tunnels) were collected from an unsprayed olive orchard in Nerokourou village (Crete, Kolymvari region, 2 ha, 35° 31' 45, 67" N–23° 46' 39, 32" E-71 m). Adults' sensitivity to copper compounds and deltamethrin was tested by feeding bioassays.

Endosymbiont abundance

'Ca. E. dadicola' population levels in the thorax-head or abdomen parts of *B. oleae* adults treated with the insecticide deltamethrin, copper compounds and their combinations, in comparison with water treated controls were assessed using Real-time PCR.

Results

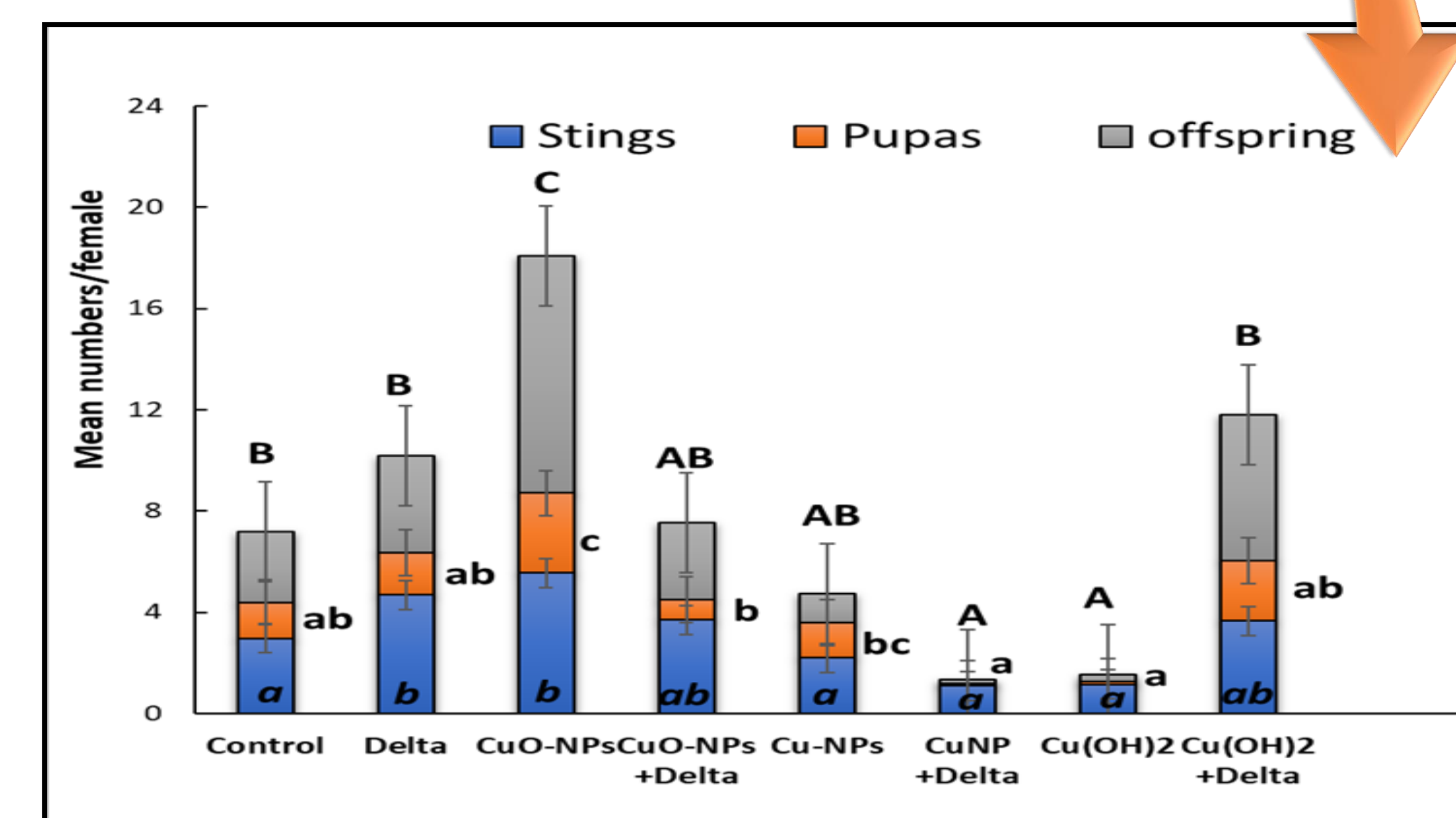


Figure 3. Effect of pesticides/NPs treatments on mean number of oviposition stings, pupas on olive fruit and offspring per female adult of *B. oleae*, 9 days post treatment.

Insecticidal action of CuNPs and synergy

Cu-NPs exhibited mortality rates similar to that of deltamethrin. The combination of copper nanoparticles with the pyrethroid insecticide deltamethrin resulted an enhanced toxic effect against adults of *B. oleae* which was more profound in the case of Cu-NPs (Fig. 2).

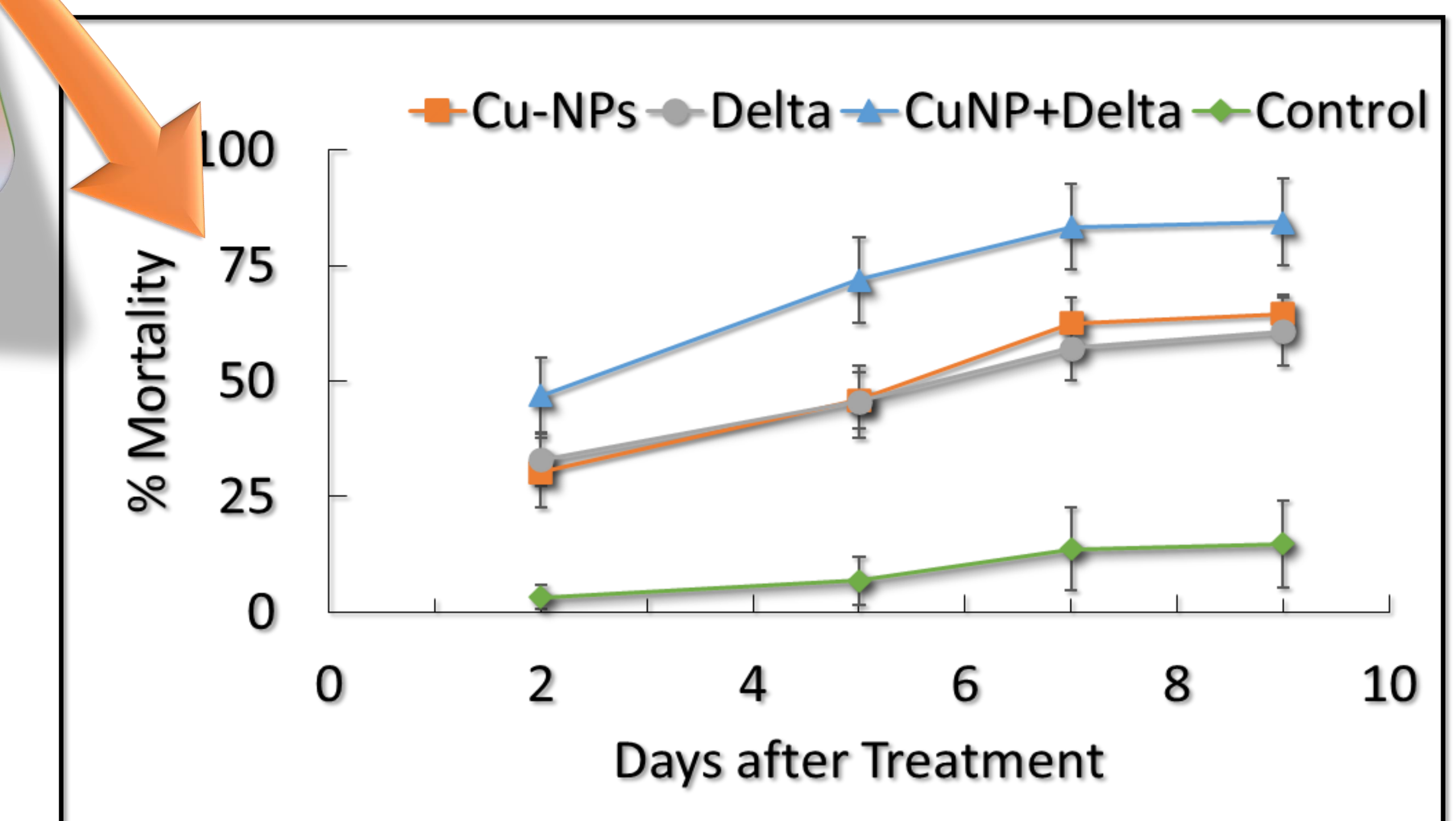


Figure 2. Effect of combination of Cu-NPs (1200 µg/mL) with deltamethrin (Delta) (125 µg/mL) on adult mortality of *B. oleae* 2, 5, 7 and 9 days after treatment.

Fecundity/ endosymbiont abundance

Both bulk and nanosized copper reduced the total number of offspring and the abundance of the gut endosymbiotic bacterium, an effect which was more profound in the head-thorax part of *B. oleae* (Figs 3 and 4).

References

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