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Insecticidal Effect of Zinc Oxide and Titanium Dioxide Nanoparticles against *Bactericera cockerelli* Sulc. (Hemiptera: Triozidae) on Tomato *Solanum lycopersicum*

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Abstract: The use of nanoparticles (NPs) has generated an alternative pest control. The objective was to evaluate the insecticidal effect of zinc oxide nanoparticles (ZnO NPs), titanium dioxide nanoparticles (TiO_2 NPs), and their combination on *Bactericera cockerelli* (Hemiptera: Triozidae) second-stage nymphs under laboratory and greenhouse conditions in tomato. The laboratory research was carried out with the leaf immersion bioassay method under a complete randomized design, and in the greenhouse by direct plant spraying under a randomized block design; in both designs, a control without NPs was added. Mortality was recorded every 24 h for 4 days. Both NPs in the laboratory and greenhouse showed toxicity to *B. cockerelli* nymphs. Results in the laboratory showed that NPs significantly caused increased mortality of 88, 99, and 100% 96 h after treatment of ZnO NPs, TiO_2 NPs, and their combinations, at 1000, 100, and 250 ppm, respectively. Direct spray of plants in the greenhouse showed low mortality with 27, 32, and 23% after 96 h of ZnO NPs, TiO_2 NPs, and their combinations, at 3000, 500, and 250 ppm, respectively. These results on *B. cockerelli* control seem promising. Nanoparticles as insecticides are a novel strategy, however, further investigation is required in field tests to obtain suitable efficacy for use in a pest management system.

Keywords: nanoinsecticide; nanoparticles; nanotechnology; pest control; tomato psyllid; TiO_2; ZnO

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1. Introduction

Mexico is one of the main tomato (*Solanum lycopersicum* L., Solanaceae) growers, with a production of 3,249,186 tons, with a market share of 25% of world exports, ranking second in terms of agricultural products with the highest export [1]. Tomato is an important vegetable, not only economically, but also with a high nutritional value considered an important source of vitamins, minerals, proteins, fiber, and the main source of lycopene, a carotenoid which has antioxidant, anticancer, and anti-inflammatory effects [2].

Tomato psyllid, *Bactericera cockerelli* Sulc. (Hemiptera: Triozidae), is one of the most destructive pests in the western hemisphere. It is a serious and economically important

pest of potatoes, tomatoes, and other crops within the family Solanaceae [3]. It is native of North America and occurs mainly in the United States, southern Canada, and Mexico; it also grows in Central America; was recently reported in South America, Ecuador; and it is also widespread in New Zealand, with a few occurrences in Australia. It has been placed on the list of quarantine pest in EPPO region [4].

This psyllid causes direct damage through feeding and extraction of sap and indirect damage by transmission of *Candidatus* Liberibacter solanacearum [5,6], which is a causal agent of the tomato permanent disease (TP) [5]. The main symptoms of TP are leaf curling, with a brittle structure with an intense green color; the apical leaflets become chlorotic with purple margins, causing flower abortion, growth reduction, and generally plants become weak, which increases their susceptibility to other diseases [5]; disease losses in tomato are up to 80% [7].

In Mexico, *B. cockerelli* control is carried out mainly through the application of chemical insecticides. In the states of Coahuila and San Luis Potosí, up to 12 applications are applied during the tomato and potato-growing season with thiacloprid, imidacloprid, and other insecticides [8]. Control is ineffective, not necessarily due to insect resistance, but to poor insecticide use [8]. Other control strategies have been suggested, such as plant resistance improvement [9] and the implementation of biological control using predatory insects and parasitoids [4], as well as entomopathogenic fungi [10].

Nanotechnology is currently considered of great importance for different sectors: industrial, cosmetics, medicine, pharmaceutical, electronics, and agricultural [11–13], and particularly in the elaboration, characterization, and application of nanometric dimensions materials, with sizes between 1–100 nanometers (nm) [14]. Different compounds are used in nanomaterial elaboration, such as metallic nanoparticles (NPs), gold (Au), silver (Ag), copper (Cu), iron (Fe), aluminum (Al), cobalt (Co), titanium (Ti), and zinc (Zn) [15]. The application of NPs in agriculture and the environment are important for their potential use in solving problems that with normal scale products are very expensive and/or are not always efficiently solved. In agricultural and food production, the basic premise is to minimize losses and reduce adverse effects on the environment due to the excessive use of agricultural inputs such as insecticides for pest control.

Nanotechnology is on the rise in the agricultural and food sector, because its potential benefits are focused on improving the quality and safety of agricultural inputs by being used in less volume and promoting improvements in nutrition [16]. NPs with unique chemical properties influence plant growth, cell structure, and physiological and biochemical functions [17]. Benefits in agriculture include reduced fertilizer loss; enhancement of agricultural productivity [13]; increased crops quality and yield [18]; and the potential for pest control [19,20], becoming an alternative to chemical insecticides, because they are considered relatively safe for humans compared to synthetic insecticides [21,22].

Zinc oxide nanoparticles (ZnO NPs) in agriculture have potential in promoting seed germination rate [23,24], plant growth and development [25–28], as well as fungicide and bactericide properties [28–31]. However, phytotoxic effects have also been attributed to it, particularly inhibiting root growth [24,25]. Research has reported the insecticidal effects of ZnO NPs, ie, Hamza [32] suggests that ZnO NPs have the capacity to be used to protect rice grain, *Oryzae sativa* L. (Poaceae), from the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) feeding; in another study, Keratum et al. [21], with the same pest, found 46.8% mortality and a reduction of the progeny of 85.80% at a concentration of 0.8% w/w after 15 days of evaluation. This effect was also recorded on the pink bollworm, *Pectinophora gossypiella* Saunders (Lepidoptera: Gelechiidae), with a mortality of 100% at LC_{50} of 11.29 ppm [33] as well as 96% mortality of second-stage larvae of *Culex quinquefasciatus* Say (Diptera: Culicidae) at 30 mg/L [34].

Titanium dioxide nanoparticles (TiO_2 NPs) in agricultural areas have shown an increase in the activity of several enzymes and promote nitrate absorption, accelerating the transformation of inorganic to organic nitrogen, making it more assimilable, hence increasing vegetative growth [35] in addition to having fungicidal and bactericidal activity

against various important fungi and phytopathogenic bacteria [36,37]. The TiO_2-NPs have affected quantitative and nutritional parameters such as oil content and changed sunflower (*Helianthus annuus* L., Asteraceae) physiology to early maturation [38]. Insecticidal activity of TiO_2 was proved against the red palm weevil, *Rhynchophorus ferrugineus* Olivier (Coleoptera: Curculionidae), with 100% mortality at 75 mg/L 10 days after treatment [39], and also showed insecticidal activity against *Spodoptera littoralis* Boisd., larvae (Lepidoptera: Noctuidae) with a mortality of 80 and 100% from 250 ppm on fourth and second instar larvae after 15 days of evaluation [12].

The titanium dioxide is a plant-growth enhancer, of which low-level concentrations improve plant physiology and stress response [38,40]. TiO_2 NPs induce plant growth; it can also delay senescence and accelerate cell division via changes in phytohormones levels, stimulate chlorophyll biosynthesis, and strengthen the photosynthetic machinery of plants, increasing the total chlorophyll content and maximum photochemical efficiency photosystem II (PSII) [20,41] and increasing yields after foliar TiO_2-NP application [42].

The mineral nutrition of plants is of primordial importance, and in these areas, nanofertilizers, based on elements such as zinc, are among the metals most used for the synthesis of NPs [31].

Zinc is an essential microelement for the general growth and development of plants [43]. It acts as a precursor of phytohormones such as auxins, which promote cell elongation and division and have an influence on the reactivity of indoleacetic acid, and as a hormonal phytostimulant, associated with the biosynthesis of cytokinins and gibberellins. In addition, it is essential for photosynthesis and facilitates carbohydrate metabolism in plants because Zn stabilizes or activates the proteins involved in these processes [30,44–46], increases biological fitness, and helps plants to cope with stress conditions [20].

Currently, environmentally friendly pest control alternatives are being sought that are economical and efficient for use in pest management. Thus, the objective of this research was to evaluate the insecticidal effect of zinc oxide and titanium dioxide nanoparticles on *B. cockerelli* nymphs, under controlled laboratory and greenhouse conditions in tomato.

2. Materials and Methods

2.1. Experiment Location

Research was conducted at the Entomology Molecular Laboratory and greenhouse of Universidad Autonoma Agraria Antonio Narro (UAAAN), in Saltillo, Coahuila, Mexico.

2.2. Insect Rearing

Bactericera cockerelli insects were reared on tomatoes Floradade variety plants, in 60 × 60 × 60 cm, pathogen- and parasitoid-free cages, under greenhouse conditions at 25 ± 2 °C, RH of 60 ± 10%, 14:10 h L:D photoperiod and automated ventilation to reduce heat and renewal of carbon dioxide.

2.3. Vegetative Material

Tomato plants of Floradade variety were used (Fax de Occidente S.A. de C.V., Guadalajara, Jalisco, Mexico) and determined growth plant, which was selected due its high susceptibility to *B. cockerelli* [47]. Seeds were planted in expanded polystyrene germination trays of 200 cavities in peat moss-perlite substrate in a 2:1 ratio. Transplant was made 30 days after planting in 2 L polyethylene containers for the laboratory tests (25 ± 2 °C, 70% RH and 14:10 h L:D photoperiod) and in 10 L polyethylene containers for greenhouse tests, under conditions at 25 ± 2 °C, RH of 60 ± 10%, 14:10 h L:D photoperiod and automated ventilation to reduce heat and renewal of carbon dioxide.

2.4. Synthesis and Characterization of ZnO Nanoparticles

ZnO-NPs were synthesized at Centro de Investigación en Química Aplicada (CIQA), in Saltillo, Coahuila, through controlled precipitation according to [48] technique, by the chemical hydrolysis method as follows: 13.7 g of $Zn(O_2CCH_3)_2$ and 600 mL of ethanol

were placed in a ball flask with three necks. This solution was constantly stirred at 75 °C under reflux for 2 h. Then an aqueous solution of 0.22 M NaOH and an additional 100 mL of distilled H_2O were added to complete the reaction mixture. Constant stirring was continued for 24 h. Subsequently, the ZnO-NPs obtained immersed in ethanol was recovered by centrifugation at 15,000 rpm during 5 min. The precipitate was washed two times with ethanol and dried in an oven at 60 °C for 24 h. The dried ZnO-NPs was crushed in an agate mortar to obtain a fine powder and stored at room temperature until use. Size and morphology nanoparticles were measured by means of a high-resolution transmission electronic microscope (HRTEM) Titan 80–300 kV (FEI Company, Hillsboro, OR, USA).

2.5. Obtaining TiO_2 Nanoparticles

Powdered TiO_2-NPs were obtained commercially from Universal Selector™ (Paris, France).

2.6. Bioassays

2.6.1. Evaluation of ZnO and TiO_2 NPs on *B. cockerelli* under Laboratory Conditions

Tomato plants were placed in a 60 × 60 × 60 cm cage, and *B. cockerelli* adults were released during 48 h to oviposit; after hatching, nymphs were followed until reaching the second instar.

The susceptibility test method 002 based on *Psylla* spp. Geoffroy (Hemiptera: Triozidae) was used to evaluate mortality of the Insecticide Resistance Action Committee [49]. Bioassay consisted in examining detached tomato leaflets aided by a Carl Zeiss Stemi DV4 binocular Stereomicroscope (Carl Zeiss Microscopy GmbH, Jena, Thuringia, Germany). The number of *B. cockerelli* live second instar nymphs per leaflet were counted and recorded.

The bioassay technique used was leaflet immersion, in which infested leaflets with nymphs were immersed in each treatment for 5 seconds. Treated leaflets were kept in plastic trays with cotton saturated with distilled water under controlled laboratory conditions (25 ± 2 °C, 70% RH and 14:10 h L:D photoperiod). The different concentrations of NPs were prepared using distilled water for dilution and polysorbate 20 (Tween 20) as an emulsifying agent at a ratio of 1 mL: 1 L of water. Six concentrations and replicates were established for each nanoparticle alone and in combination (both nanoparticles mixed in equal parts) in addition to a check treatment without NPs, applying distilled water only (Table 1). Each infested leaflet was considered an experimental unit and replicated three times.

Table 1. Nanoparticles concentrations evaluated for *Bactericera cockerelli* control in the laboratory and greenhouse.

Treatment	Laboratory	Greenhouse
	Concentration (ppm)	
Control	0	0
ZnO NPs	100	—
	300	300
	500	—
	1000	1000
	2000	—
	3000	3000
TiO_2 NPs	40	40
	60	—
	80	—
	100	100
	300	—
	500	500

Table 1. *Cont.*

Treatment	Laboratory	Greenhouse
	Concentration (ppm)	
ZnO NPs-TiO_2 NPs (combination *)	20	20
	30	—
	40	—
	50	50
	150	—
	250	250

* Both nanoparticles mixed in equal parts.

2.6.2. Evaluation of ZnO and TiO_2 NPs on *B. cockerelli* under Greenhouse Conditions

Tomato plants 50 days after planting, grown in polyethylene containers with approximately 10 L of peat moss-perlite substrate in a 2:1 ratio were used. Tomato plants were placed in a 60 × 60 × 60 cm cage, releasing 15 adults of *B. cockerelli* on each plant during 48 h to oviposit; adults were then removed and eggs were followed from hatching until second nymphal instar, with 50 nymphs on average per plant treated.

Testing consisted of applying the treatments directly to the plants with a 500 mL manual sprinkler, at a rate of 25 mL per plant. Based on the laboratory tests, three concentrations were used per NPs alone and in combination (low, medium, and high concentration), in addition to a control treatment without nanoparticles applying distilled water only, with six replicates per concentration (Table 1). The different concentrations of NPs were prepared following the same methodology of the laboratory bioassay.

2.7. Mortality Evaluation

In both bioassays, mortality evaluation was carried out 24, 48, 72, and 96 h after treatment. The laboratory bioassay was evaluated with a binocular stereoscope microscope and in the greenhouse with a magnifying glass. The number of surviving and dead nymphs was recorded in which a nymph was considered dead, when not responding to a stimulus, appendages detached from to the body, and/or was dehydrated.

2.8. Data Analysis

Mortality data were analyzed using probit analysis to estimate LC_{50}, and LC_{95} values and fiducial limits at 95% significance. Before probit analysis, mortality was corrected by Abbott formula [50] with an accepted mortality in the control of 17%. To determine the effect of treatments on *B. cockerelli*, data were transformed by arcsine square root, and analysis of variance under a completely randomized design (laboratory), complete randomized block design (greenhouse), and mean comparison with a Tukey's multiple range test ($p < 0.05$) were performed. For both, the statistical package SAS/STAT [51] was used.

3. Results

3.1. Size and Morphology Nanoparticles of ZnO and TiO_2

To obtain the size distribution of the particles by transmission electronic microscope technique, 300 and 250 ZnO and TiO_2 NPs were analyzed, respectively.

The transmission electronic microscope micrograph of ZnO nanoparticles is shown in Figure 1, which displays a morphology crystalline semi-sphere with an average size of 23.44 nm and even distribution size (a). The larger particles are distributed to particle agglomeration. The particle size distribution histogram indicates that it ranges from 7.3 to 42.7 nm (b).

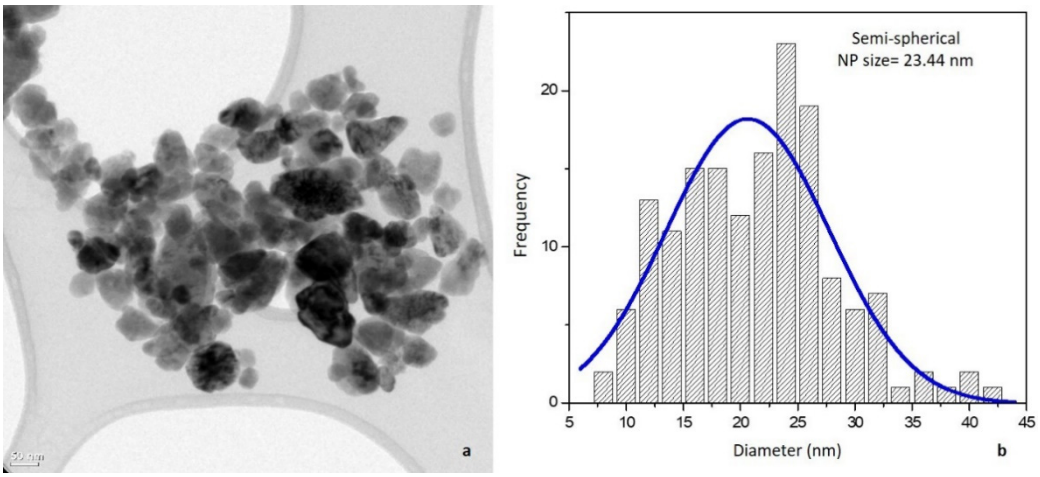

Figure 1. Transmission electron microscopy micrograph showing the semi-spherical morphology of ZnO-NPs (**a**) and histogram of particle size distribution (**b**).

The transmission electronic microscope micrograph of TiO_2 nanoparticles is shown in Figure 2, which displays a needle-shaped morphology with a diameter of 76.15 nm long and 8.52 nm wide.

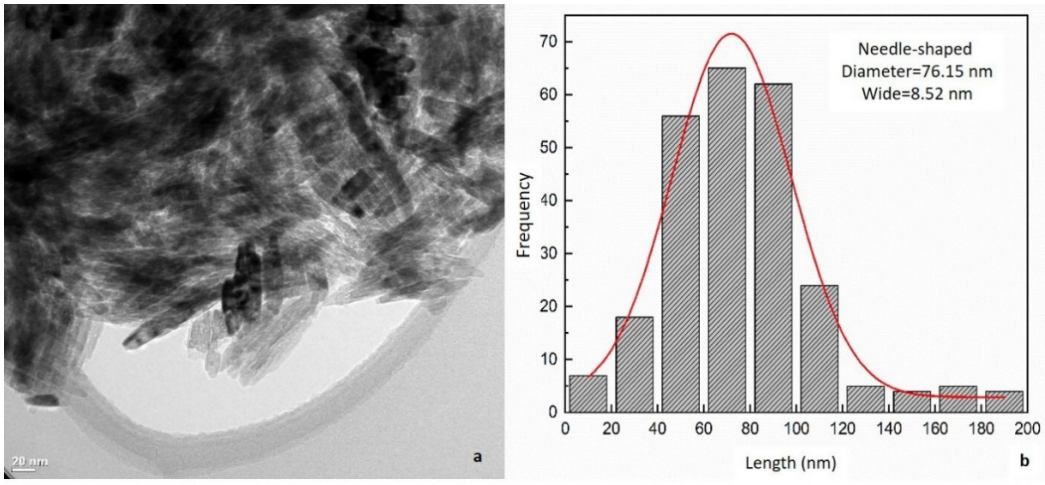

Figure 2. Transmission electron microscopy micrograph showing the needle-shaped morphology of TiO_2 NPs (**a**) and histogram of particle size distribution (**b**).

3.2. Mortality of Tomato Psyllid B. cockerelli by NPs on Tomato Leaves in the Laboratory

Nanoparticles of ZnO, TiO_2, and their combination showed significant insecticidal action ($p < 0.05$) against *B. cockerelli* second stage nymphs under laboratory and greenhouse conditions.

The ZnO NPS at 24 and 48 h after application, in general, did not show differences among concentrations, but were different with respect to the control, with the exception of 1000 ppm. After 72 and 96 h again, no differences were observed among concentrations, however, significant differences were found when compared to the control. The NPs showed high insecticide activity, particularly in concentrations ranging from 500 ppm to 3000 ppm, with mortality above 80% and an LC_{50} of 14.14 ppm 96 h after evaluation (Table 2).

Table 2. Mortality (±SD) of tomato psyllid *Bactericera cockerelli*, at different concentrations of ZnO NPs on tomato leaves in the laboratory.

Concentration (ppm)	Mortality (%) [1]			
	24 h	48 h	72 h	96 h
0	2.16 ± 1.07 c	7.29 ± 2.88 c	12.04 ± 2.33 b	16.86 ± 3.70 b
100	18.82 ± 5.33 a	38.74 ± 3.80 ab	54.91 ± 2.92 a	75.04 ± 5.16 a
300	16.22 ± 7.68 a	45.33 ± 8.75 a	58.83 ± 8.37 a	79.00 ± 7.24 a
500	12.32 ± 7.66 ab	41.65 ± 20.76 ab	61.68 ± 18.22 a	80.48 ± 20.24 a
1000	4.86 ± 4.87 bc	27.33 ± 4.07 b	62.56 ± 10.96 a	88.02 ± 13.07 a
2000	17.84 ± 6.37 a	42.41 ± 3.90 ab	64.87 ± 6.24 a	87.80 ± 3.54 a
3000	17.74 ± 3.33 a	40.12 ± 11.22 a	64.18 ± 14.04 a	88.58 ± 9.97 a
df	6.41	6.41	6.41	6.41
F	14.06	15.21	24.39	21.84
Pr > F	<0.0001 ***	<0.0001 ***	<0.0001 ***	<0.0001 ***
R2	0.76	0.77	0.84	0.82
[&]LC_{50} ([$]FL, 95%)	4.999×10^{-18} (NC)	1.12909×10^{-8} (NC)	160.99 (0.00146–468.04)	14.14 (0.64269–47.91883)
[&]LC_{95} ([$]FL, 95%)	1.439×10^{-46} (NC)	1.1502×10^{-49} (NC)	2,780,761,440 (2,051,410–9.57389 $\times 10^{-61}$)	22,832 (6655–519,178)

[1] Data transformed by arcsine square root. Means with the same letter in same column are not significantly different (Tukey; $p < 0.05$). *** Indicate significant contrast value F to $p < 0.001$. [&] Letal concentration. [$] Fiducial limits. N.C. = Not calculated by statistical software.

The insecticidal effect of TiO_2 NPs after 24 h showed over 90% mortality in concentrations above 100 ppm. After 48 h of the application of treatments, the insecticide activity had a significant increased mortality with 80% at the lowest concentration, a trend observed in all evaluations and concentrations, with 93% in the lowest concentration at 72 h and 100% mortality in highest concentration at 72 and 96 h with an LC_{50} of 11.18 ppm of NPs-TiO_2 to control *B. cockerelli* second instar nymphs (Table 3).

Table 3. Mortality (±SD) of tomato psyllid *Bactericera cockerelli*, at different concentrations of TiO_2 NPs on tomato leaves in the laboratory.

Concentration (ppm)	Mortality (%) [1]			
	24 h	48 h	72 h	96 h
0	2.16 ± 1.07 c	7.29 ± 2.88 c	12.04 ± 2.33 d	16.86 ± 3.70 b
40	39.02 ± 12.47 b	80.65 ± 10.80 b	93.91 ± 11.44 c	95.36 ± 9.82 a
60	41.95 ± 14.71 b	80.52 ± 16.31 b	92.59 ± 8.89 c	94.84 ± 7.71 a
80	54.57 ± 6.94 b	90.59 ± 4.07 b	97.36 ± 3.97 abc	98.39 ± 3.34 a
100	93.01 ± 2.80 a	98.42 ± 1.57 a	99.90 ± 0.25 ab	100.00 ± 0.00 a
300	94.09 ± 5.53 a	99.83 ± 0.42 a	100.00 ± 0.00 a	100.00 ± 0.00 a
500	97.78 ± 2.18 a	99.70 ± 0.38 a	100.00 ± 0.00 a	100.00 ± 0.00 a
df	6.41	6.41	6.41	6.41
F	153.97	125.60	119.94	107.12
Pr > F	<0.0001 ***	<0.0001 ***	<0.0001 ***	<0.0001 ***
R2	0.97	0.96	0.96	0.96
[&]LC_{50} ([$]FL, 95%)	60.05 (13.97–102.9)	21.99 (0.006–39.13)	12.67 (NC)	11.18 (0.96–21.12)
[&]LC_{95} ([$]FL, 95%)	297.51 (148.51–36,371)	113.12 (74.34–11,845)	66.95 (NC)	58.92 (45.48–78.71)

[1] Data transformed by arcsine square root. Means with same letter in same column are not significantly different (Tukey; $p < 0.05$). *** indicate significance contrast value F to $p < 0.001$. [&] Letal concentration. [$] Fiducial limits. N.C. = Not calculated by statistical software.

Combined action of ZnO and TiO_2 NPs was observed at 24 h with significant differences in *B. cockerelli* mortality with concentrations of 150 and 250 ppm. From 48–96 h after application, mortality increased significantly at the different concentrations, reaching 99% mortality at the highest evaluated concentration. The different concentrations showed to be efficient to control *B. cockerelli* second instar nymphs, with an LC_{50} of 5.59 ppm of nanoparticles in combination, after 96 h post application (Table 4).

Table 4. Mortality (±SD) of tomato psyllid *Bactericera cockerelli*, with different concentrations of a combination of ZnO and TiO_2 NPs on tomato leaves in the laboratory.

Concentration (ppm) [1]	Mortality (%) [2]			
	24 h	48 h	72 h	96 h
0	2.16 ± 1.07 c	7.29 ± 2.88 d	12.04 ± 2.33 e	16.86 ± 3.70 c
20	6.65 ± 4.77 c	35.77 ± 5.86 c	74.33 ± 9.23 d	85.12 ± 8.58 b
30	17.95 ± 2.72 b	59.63 ± 9.68 b	80.45 ± 10.85 cd	89.27 ± 5.35 b
40	28.81 ± 6.55 b	78.10 ± 10.33 a	95.90 ± 5.01 ab	98.40 ± 2.84 a
50	26.84 ± 5.89 b	64.26 ± 6.76 b	91.07 ± 2.08 bc	97.96 ± 1.16 a
150	58.59 ± 11.30 a	82.12 ± 6.59 a	96.16 ± 2.33 ab	98.96 ± 1.22 a
250	45.77 ± 7.33 a	82.96 ± 3.05 a	98.22 ± 1.41 a	99.72 ± 0.69 a
df	6.41	6.41	6.41	6.41
F	61.41	85.19	113.30	124.32
Pr > F	<0.0001 ***	<0.0001 ***	<0.0001 ***	<0.0001 ***
R2	0.92	0.94	0.96	0.96
[&]LC_{50} ([$]FL, 95%)	180.60 (97.13–1387)	26.81 (0.0057–58.59)	6.72 (0.0029–16.45)	5.59 (2.457–13.93)
[&]LC_{95} ([$]FL, 95%)	3405 (686.75–617,363)	805.77 (194.007–6.125)	101.43 (54.81–7482)	45.55 (27.78–4088)

[1] Both nanoparticles mixed in equal parts. [2] Data transformed by arcsine square root. Means with the same letter in same column are not significantly different (Tukey; $p < 0.05$). *** indicate significance contrast value F to p <0.001. [&] Letal concentration. [$] Fiducial limits.

3.3. Mortality of Tomato Psyllid B. cockerelli by NPs in the Greenhouse

Significant results ($p < 0.05$) were found in the mortality of *B. cockerelli* due to the action of ZnO and TiO_2 NPs and their combination under greenhouse conditions; although insecticide activity was obtained, these results were not as expected, given the results found under laboratory conditions. *B. cockerelli* mortality is considered low for both NPs and their combination, with mortality observed 48 h after treatment in all concentrations. The ZnO NPs had 5.79% mortality at the highest concentration after 48 h, increasing to 27.02%, 96 h after treatment, at 3000 ppm concentration, with an LC_{50} of 15,862 ppm (Table 5).

Table 5. Mortality (±SD) of tomato psyllid *Bactericera cockerelli*, at different concentrations of ZnO NPs on tomato in the greenhouse.

Concentration (ppm)	Mortality (%) [1]			
	24 h	48 h	72 h	96 h
0	0.00 ± 0.00 a	0.00 ± 0.00 a	4.14 ± 1.87 b	9.09 ± 6.30 b
300	0.00 ± 0.00 a	1.39 ± 3.40 b	4.63 ± 3.52 b	12.00 ± 9.58 b
1000	0.00 ± 0.00 a	5.11 ± 6.03 ab	9.21 ± 4.08 ab	17.00 ± 7.83 ab
3000	0.00 ± 0.00 a	5.79 ± 1.64 a	13.40 ± 4.30 a	27.02 ± 6.92 a
df	3.23	3.23	3.23	3.23
F	N.C.	6.21	5.98	4.34
Pr > F	N.C.[ns]	0.0059 **	0.0068 **	0.0217 *
R2	0.00	0.62	0.57	0.50
[&]LC_{50} ([$]FL, 95%)	N.C.	203,872 (17,195–2.06583 × 10^{-90})	69,511 (N.C.-N.C.)	15,862 (5894–150,031,026)
[&]LC_{95} ([$]FL, 95%)	N.C.	18,147,270 (172,073–3.1171 × 10^{-170})	4,503,170 (N.C.-N.C.)	402,640 (36,256–5.01396 × 10^{-15})

[1] Data transformed by arcsine square root. Means with the same letter in same columns are not significantly different (Tukey; $p < 0.05$). *, ** indicate significance contrast value F to $p < 0.05$, <0.01 respectively. ns = not difference. [&] Lethal concentration, [$] Fiducial limits. N.C. = Not calculated by statistical software.

The TiO_2 NPs showed results similar to the ZnO NPs under this same condition, with a low insecticide effect on all concentrations tested, with 11.81% after 48 h at the highest concentration up to 32.71% mortality 96 h after treatment, with 9.09% mortality in control treatment and an LC_{50} of 1657 ppm of TiO_2 NPs (Table 6).

Table 6. Mortality (±SD) of tomato psyllid *Bactericera cockerelli*, at different concentrations of TiO_2 NPs on the greenhouse.

Concentration (ppm)	Mortality (%) [1]			
	24 h	48 h	72 h	96 h
0	0.00 ± 0.00 a	0.00 ± 0.00 c	4.14 ± 1.87 b	9.09 ± 6.30 b
40	0.00 ± 0.00 a	1.64 ± 1.92 bc	9.04 ± 5.28 b	14.49 ± 7.75 b
100	0.00 ± 0.00 a	6.38 ± 6.29 ab	10.02 ± 7.09 b	14.76 ± 11.55 b
500	0.00 ± 0.00 a	11.81 ± 5.25 a	22.26 ± 5.60 a	32.71 ± 6.00 a
df	3.23	3.23	3.23	3.23
F	N.C.	15.58	13.43	9.27
Pr>F	N.C. [ns]	<0.0001 ***	0.0002 ***	0.0010 **
R2	0.00	0.76	0.75	0.69
[&]LC_{50} ([$]FL, 95%)	N.C.	19,319 (2723–1,187,447,001)	3776 (1384–57,527)	1657 (887.33849–5952)
[&]LC_{95} ([$]FL, 95%)	N.C.	2,479,473 (48,757–1.77645 × 10^{-16})	138,470 (16,440–58,180,024)	31,049 (7826–631,186)

[1] Data transformed by arcsine square root. Means with the same letter in the same columns are not significantly different (Tukey; $p < 0.05$). **, *** indicate significance contrast value F to $p < 0.01$, <0.001 respectively. ns = not difference. [&] Letal concentration, [$] Fiducial limits. N.C. = Not calculated by statistical software.

In the combined NPs experiment, the same pattern was observed as in the individual evaluation, with low mortality in all concentrations used. Mortality was observed at 48 h without significant differences among concentrations, and with 20% more mortality at 96 h at a concentration of 250 ppm, which had 23.89% mortality and an LC_{50} of 2.15679 × 10^{10} ppm (Table 7).

Table 7. Mortality (±SD) of tomato psyllid *Bactericera cockerelli*, at different concentrations of combined ZnO-TiO_2 NPs on tomato greenhouse.

Concentration (ppm) [1]	Mortality (%) [2]			
	24 h	48 h	72 h	96 h
0	0 ± 0.00 a	0.00 ± 0.00 a	4.14 ± 1.87 b	9.09 ± 6.30 a
20	0 ± 0.00 a	0.19 ± 0.47 a	4.56 ± 2.92 b	9.08 ± 6.02 a
50	0 ± 0.00 a	1.70 ± 2.31 a	7.34 ± 5.21 ab	18.26 ± 9.53 a
250	0 ± 0.00 a	2.37 ± 2.04 a	11.51 ± 6.37 a	23.89 ± 15.50 a
df	3.23	3.23	3.23	3.23
F	N.C.	3.50	4.12	3.95
Pr>F	N.C. [ns]	0.0418 *	0.0257 *	0.0292 *
R2	0.00	0.50	0.57	0.57
[&]LC_{50} ([$]FL, 95%)	N.C.	368,554 (N.C.-N.C.)	281,036 (N.C.-N.C.)	2.15679 × 10^{-10} (N.C.-N.C.)
[&]LC_{95} ([$]FL, 95%)	N.C.	231,517,521 (N.C.-N.C.)	226,344,843 (N.C.-N.C.)	3.99698 × 10^{-19} (N.C.-N.C.)

[1] Both nanoparticles mixed in equal parts. [2] Data transformed by arcsine square root. Means with the same letter in the same column are not significantly different (Tukey; $p < 0.05$). * indicate significance contrast value F to $p < 0.05$. ns = not difference. [&] Letal concentration, [$] Fiducial limits. N.C. = Not calculated by statistical software.

4. Discussion

4.1. Effect of ZnO and TiO_2 NPs against Tomato Psyllid B. cockerelli in Laboratory and Greenhouse

The ZnO and TiO_2 NPs and their combination showed significant effects against *B. cockerelli* second instar nymphs under laboratory and greenhouse conditions; however, toxicity was significantly higher in laboratory tests. The results obtained in this study, at the laboratory level, evidenced the insecticidal activity of ZnO and TiO_2 Nanoparticles, given the direct contact of the NPs with the insect and under totally controlled conditions (25 ± 2 °C, 70% RH and 14:10 h L:D photoperiod), without any other factor that interacts with the result. Although, in the greenhouse test, the conditions are semi-controlled (25 ± 2 °C, RH of 60 ± 10%, 14:10 h L:D photoperiod and automated ventilation to reduce

heat and renewal of carbon dioxide), meaning more similar conditions to open field when other factors can interact (light, night, temperature, water, wind), it is not considered that these factors will spread the nanoparticles outside of the plant. The main factor that could determine the lack of efficiency in greenhouse conditions is the size of the plants, which were already large (40–45 cm approximately) and with a large amount of foliage, and also the application of a low volume (25 mL per plant), which did not cover the foliage sufficiently. For the greenhouse test, more studies are needed to establish the specific dose for insect control, as well as to increase the volume of nanoinsecticide application, which is correlated also with foliage surface, at efficient insecticides doses that will not cause phytotoxicity.

4.2. Nanoparticles Effects on Plants

The use of NPs in the field for plant protection, chemical pesticides reduction, and environment care, is an increasing need, particularly due to their size that ranges from 120–250 nm and an efficiently water-soluble size range compared to existing pesticides [12].

ZnO NPs have attracted special attention over time, due to their stability and biosafety use; recently, TiO_2 NPs have gained more attention for different applications; however, to a greater degree in the medical area [52].

In the agricultural sector, NPs are studied for their potential as nanofertilizers, correcting zinc deficiencies in plants, and as promoters of plant growth and development [53–55]. The growth promotion in plants is related to the concentration, size, and inherent properties of the element involved, as well as the physiological and biochemical function it plays in plant, and whether it acts as a micronutrient, as is the case with zinc [56].

Several investigations confirm the role of ZnO NPs as promoters of germination, seedling growth, dry biomass production, root and stem elongation, and significant increase in chlorophyll and protein in seedlings [57–59].

TiO_2 NPs positive effect on plant growth, and especially in plant tolerance against abiotic stress, is known. The application of TiO_2 NPs application improved all agronomic traits, increased antioxidant enzymes activities, photosynthetic rate, chlorophyll formation, soluble sugars, amino acids, and proline content, in addition to significantly reducing the hydrogen peroxide (H_2O_2) concentration and lipid peroxidation (MDA) contents in plants under saline conditions, which subsequently caused enhanced crop yield [60–63]. Servin et al. [64] in their study with cucumber *Cucumis sativus* L. (Cucurbitaceae) showed that TiO_2 NPs increase catalase and chlorophyll in leaves, decrease ascorbate peroxidase, and significantly increase potassium and phosphorus.

4.3. Translocation of ZnO and TiO_2 Nanoparticles

The absorption effectiveness of conventional fertilizers and pesticides by foliar application is low because they are highly soluble and leach rapidly. Highly water-soluble ions may have difficulty penetrating the lipophilic cuticle, limiting the availability of the active ingredient [57].

In contrast, NPs, when applied to foliage, enter cells through the vascular or stomatal system, depending on the size range of the metallic particles [58,65,66], and its basipetal translocation or transport towards the base of the stem is by the phloem. Once NPs enter the phloem, further translocation to various plant organs and developing sinks are mediated by short- and long-distance pathways [67]. When NPs are applied to the soil or in the irrigation water, they penetrate through the epidermis of the root and the bark, later they pass to the endodermis, and finally they enter the conductive tissue of the xylem to be translocated by a long distance to the branches and the foliage of plants [66]. In addition, it presents a slow and gradual availability of the active ingredient, which results in greater efficiency [68].

It has been pointed out that cell walls and membranes act as an effective barrier to the entry of any type of NP and that the effectiveness of their entry and transport is determined by the size of the cell wall pores, which are in the range of 2–20 nm [69,70]. The stoma

appears as the most feasible route for the penetration of NPs [70]. The mobility of Zn is higher in phloem than xylem due to increased concentration of chelating solutes (peptides, organic acids etc.) in phloem sap. Zn is transported in ionic form or as complexes of Zn–nicotianamine, Zn–malate, and Zn–histidine in phloem tissues [67].

Servin et al. [64], in cucumber, demonstrated the ability of TiO_2 NPs to translocate from root to fruit, while Vittori et al. [71] in tomato grown in soil and watered with TiO_2 NPs showed that these nanoparticles do not accumulate in the crop, observing the presence of TiO_2 NPs only distributed in the longitudinal section of the roots.

Kolenčík et al. [38], in foliar application, did not detect differences in titanium translocation or accumulation in fully ripe sunflower seeds *Helianthus annuus* L. (Asteraceae) compared to the control, which encourages further research. However, the TiO_2-NPs have affected quantitative and nutritional parameters such as oil content and changed sunflower physiology to early maturation.

4.4. Nanoparticles as Nano-Insecticides

In recent years, the use of nanotechnology has become a promising tool for pest control [72], for example, in pest control for crop protection, different NPs have been tested against various pests, mainly under laboratory conditions.

ZnO NPs were tested on a poisoned diet, using the waxworm, *Galleria mellonella* L. (Lepidoptera: Pyralidae), reporting its efficacy 48 h after application, causing death and subsequent abnormalities in the life cycle of the insect in surviving larvae at low concentrations [73].

The Ag and Ag-Zn NPs were recorded as an alternative in controlling *Aphis nerii* Boyer de Fonscolombe (Hemiptera: Aphididae), with an LC_{50} of 424.67 and 539.46 mg/mL, respectively [74].

In wheat grains *Triticum* sp. L. (Poaceae) on the flour beetle, *Tribolium castaneum* (Herbest) (Coleoptera: Tenebrionidae), Salem et al. [22] reported 69% mortality with ZnO NPs and 68% with aluminium oxide NPs (Al_2O_3), in 15 days of evaluation, compared with malathion with a 98% control.

These findings were corroborated, when observing that pirimiphos-methyl insecticide was more effective than the Al_2O_3 and ZnO NPs for *S. oryzae* control with 100% mortality 3 days after application, whereas the Al_2O_3 nanoparticles were effective after 15 days with 53.35% mortality, and the ZnO NPs had a moderate effect with 46.8% control; both nanoparticles significantly inhibited pest progeny [21].

On the other hand, ZnO NPs was the most toxic compound, with a greater effect (100% mortality) with LC_{50} value of 11.29 ppm on the pink bollworm, *P. gossypiella*, compared to silica nanoparticles and spinosad and pyriproxyfen insecticides [33].

Al-Bartya and Hamza [39] studied the larvicida activity of TiO_2 NPs combined with the aqueous extract of *Moringa oleifera* Lam. (Moringaceae) over the red weevil, *R. ferrugineus*, finding 100% mortality at a concentration of 75 mg/L 10 days after treatment.

Copper nanoparticles (CuO NPs) showed an insecticide effect on *S. littoralis* larvae after 15 days of evaluation, with 95 and 75% mortality at a concentration of 1000 mg/L on the second and fourth instar larvae, respectively [75].

Khooshe-Bast et al. [76], under laboratory conditions with greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) (Hemiptera: Aleyrodidae), obtained a mortality rate of 91.6% with ZnO NPs at a concentration of 20 mg L^{-1} and an LC_{50} of 7.35 mg L^{-1}.

In ingestion evaluations, Shaker et al. [12] demonstrated that the application of TiO_2 nanoparticles was effective after 15 days at 62.5 and 125 ppm on *S. littoralis* second and fourth instar larvae, affecting biological aspects such as larval period, pupation, adult emergence, fertility, sex ratio, longevity, and egg hatching, in addition to causing malformations in larvae, pupae, and adults.

Jovanović et al. [77] demonstrated that titanium dioxide nanoparticles have little insecticidal activity on the fruit fly, *Drosophila melanogaster* Meigen (Diptera: Drosophilidae) at concentrations relevant to oral exposure of humans (0.002 mg mL^{-1}, 0.02 mg mL^{-1},

0.2 mg mL^{-1}, and 2 mg mL^{-1} of TiO_2 in feeding medium). The TiO_2 did not affect survival and fecundity, but significantly increased time to pupation. Expression of the gene for catalase was markedly down regulated by the treatment TiO_2, while the effect on the down regulation of superoxide dismutase 2 was less pronounced.

Mostafa et al. [34] examined the efficacy of two nanoparticles and their combination on *C. quinquefasciatus* larvae, finding that ZnO NPs, Al_2O_3 NPs, and aluminum-doped zinc oxide (AZO) are effective, with mortality rates of 96, 74, and 86% respectively at the highest concentration of 30 mg L^{-1}, confirming their larvicidal effect.

Green zinc oxide nanoparticles were synthesized from spinach leaves *Spinacia oleracea* L. (Amaranthaceae) and tested on *Callosobruchus analis* F. (Coleoptera: Chrysomelidae) with 100% mortality at 1500 ppm after 10 days treatment and used as chickpea seed protectors *Cicer arietinum* L. (Poaceae) for 1 month of storage, with less oviposition and damage to seeds and less weight loss of them [72].

Ishwarya et al. [78] examined the larvicidal activity of ZnO NPs fabricated with *Ulva lactuca* L. (Ulvaceae) seaweed extract on *Aedes aegypti* (L.) (Diptera: Culicidae) fourth instar larvae. They observed that the highest mortality (100%) of larvae of *A. aegypti* within 24 h was obtained testing 50 µg/mL of ZnO NPs, and LC_{50} was 22.38 while LC_{90} was 41.94 µg/mL.

Green nanoparticles of *Scadoxus multiflorus* (Martyn) Raf. (Amaryllidaceae) were synthesized with leaf powder aqueous extract as a capping and stabilizing agent for the synthesis of ZnO NPs and were tested against *A. aegypti* larvae and eggs, giving a significant LC_{50} value of 34.04 ppm, and ovicidal activity resulted in a mortality rate of 96.4% at 120 ppm with an LC_{50} value of 32.73 ppm [79].

The insecticide efficacy of Ag NPs with malathion on *T. castaneum* was compared, and it was found that the Ag NPs combined with malathion at 150 ppm showed high control, repellency, and oviposition efficiency reduction [80].

Different concentrations of TiO_2 NPs (100, 500, 1000, 3000, and 5000 ppm) in poisoned diets were evaluated on *G. mellonella*. The authors reported that larval and pupal developmental times significantly increased at 100, 500, 1000, and 3000 ppm when compared with the control and highest dose of TiO_2 NPs. They also reported that adult longevity time was shortened at low concentrations of TiO_2 NPs (100, 500, and 1000 ppm). Exposure to TiO_2 NPs caused a significant increase in the total protein amount and content of malondialdehyde and glutathione S-transferase activity in the hemolymph at 100, 500, and 1000 ppm. While the activity of catalase increased by 1000, 3000, and 5000 ppm and superoxide dismutase activity increased at all doses of TiO_2 NPs [81].

In a similar study, Lopez Muñoz et al. [82] synthesized TiO_2 and Al_2O_3 NPs for testing its toxicity against *Oncopeltus fasciatus* (Dallas) (Hemiptera: Lygaeidae), and the effects were monitored in the filial generation to know the effect on the insect composition (protein and lipid content and lipid peroxidation). The results indicated that the ectopic exposure to nanoparticles at 1 mg/cm^2 (TiO_2) and 0.5 mg/cm^2 (Al_2O_3) did not induce lethal toxicity in *O. fasciatus*, nor did it modify reproductive parameters. However, both NPs produced an increase in nymphal life span. In the parental generation, TiO_2 NPs increased protein content whereas Al_2O_3 NPs decreased it. Al_2O_3 NPs decreased protein content, and TiO_2 NPs decreased lipid content. Responses observed in the individuals of the filial generation demonstrated the existence of trans-generational effects of Al_2O_3 and TiO_2 NPs.

Eskin et al. [83] performed a toxicity test with ZnO NPs to determine the lethal concentrations of ZnO NPs on *G. mellonella* larvae by force-feeding method. After 24 h of the treatment, 100% larval death rate was observed at 30 and 100 µg/10 µL ZnO NPs doses; LC_{50} was 6.03 µg/10 µL; and LC_{99} was 12.86 µg/10 µL.

ZnO nanoparticles are alternatives in the protection of stored seeds of *Vigna sinensis* L. (Fabaceae) and *Triticum sativum* Lam. (Poaceae) and effective in controlling adults of weevils *S. oryzae* and *Callosobruchus maculatus* F. (Coleoptera: Chrysomelidae) with 88.3 and 100% mortality, in addition to causing large reductions in the progeny (F1) of these

insects; whereas with the flour beetle *T. castaneum*, lower mortality of 38.3% was shown at the highest concentration (8 g/kg) [84].

Two nanoparticles, silica oxide (SiO_2) and Al_2O_3 with the insecticide malathion, against were compared *T. castaneum* [85]. Their results showed that malathion had the greatest effect on the insect; although to a lesser degree, NPs also inhibited the *T. castaneum* progeny with a higher effect of Al_2O_3 NPs than SiO_2 NPs, concluding that NPs are adequate to protect stored grains as an alternate method to chemical insecticides because they are safer for humans.

The efficacy of ZnO NPs was tested against first instar larvae of white grubs *Holotrichia* (Coleoptera: Scarabaeidae) and 20% mortality was found in the highest concentration (30 ppm) and LD_{50} of 12.63 ppm [86].

Only a few studies were done to evaluate the efficacy of ZnO/TiO_2 NPS or any other nanoparticles against plant pests in greenhouse and field.

The greenhouse evaluation results showed low mortality (32.71%) at the highest concentrations with TiO_2, 27.02% with ZnO NPs, and their combination with 23.89%.

These results are different from those by [87], under semi-field conditions in broad bean (*Vicia faba* L.) (Fabaceae), who report 100% mortality with foliar application of hydrophilic nanosilice and silice to control *Myzus persicae* Sulzer, *Acyrthosiphon pisum* Harris, and *Aphis craccivora* C.L. Koch (Hemiptera: Aphididae), surpassing the chemical insecticide lambda-cyhalothrin control of 78% in its highest mortality.

4.5. Nanoparticles Action Mode

Nanoparticles can affect various physiological parameters in treated organisms. Volker et al. [88] indicated with in vitro assays that the dose-dependent cell death-inducing oxidative stress was the most likely toxicity pathway.

The high efficiency of NPs also is generally attributed to their capacity to destroy the protective waxy layer of the insect's cuticle, through the absorbance of lipids, which induces death by desiccation [21,33]. In addition to reducing lipid content, they also reduce the amount of total protein, as occurs with chemical and botanical growth-regulating insecticides [33].

The mode of action of NPs also attributed to desiccation strengthens the use of these materials [89], since it is very unlikely that insect pests treated with nanoparticles genetically or physiologically generate resistance to this mechanism of action [21]. With the low-risk of developing long-term insect resistance and relative safety for humans and the environment as compared to chemical insecticides, nanoparticles are a viable option as an alternative to insecticides [21,33,80].

Precise information on the mechanisms of action of nanoparticles against insects is limited. Benelli [90] summarized the mechanism of action of some nanoparticles against insects. The author informed that silver and graphene oxide nanoparticles have a significant impact on insect antioxidant and detoxifying enzymes, leading to oxidative stress and cell death.

Ag nanoparticles also reduced acetylcholinesterase activity, up- and down-regulated key insect genes, and reducing protein synthesis and gonadotrophin release, leading to developmental damages and reproductive failure. While polystyrene nanoparticles inhibited CYP450 isoenzymes.

Au nanoparticles can act as trypsin inhibitors and disrupt development and reproduction. Metal nanoparticles can bind to S and P in proteins and nucleic acids, respectively, leading to a decrease in membrane permeability and, therefore, to organelle and enzyme denaturation, followed by cell death. The toxicity of SiO_2 and Al_2O_3 nanoparticles is due to their binding to the insect cuticle, followed by physico-sorption of waxes and lipids, leading to insect dehydration.

Ishwarya et al. [78] demonstrated that ZnO NPs-treated *A. aegypti* larvae showed the disintegration of the epithelial layer and outer cuticle, compared to Zn acetate-treated larvae, which showed the deposition of zinc inside the larval body. The histopathological

images showed the effect of exposure to ZnO NPs against *A. aegypti* larvae. The histology of *A. aegypti* larval tissues experienced different histological modifications; the epithelium was spoiled and cells were vacuolated, enclosing the nuclei in the NP-treated larvae.

Under scanning electron microscopy, Mostafa et al. [34] detected ZnO NPs on the surface of the dead larvae of *C. quinquefasciatus* and demonstrated adhesion of ZnO NPs particles on the head region, practically on the hair of the head, as well as particles adhering to the abdominal cuticle and to the gills and respiratory siphon.

4.6. Nano-Formulation of NPs for Future Nano-Insecticides

The low toxicity under greenhouse conditions in this research and based on the laboratory results suggest that more tests should be carried out with an increase in the concentrations, or if necessary, adding surfactants and adjuvants in order to improve their efficacy; increase its dispersion capacity, wetting, adherence, penetration, and droplet deposition; and improve the wetting and persistence of the active ingredient, to achieve higher effectiveness and thus confirm their insecticide effect on *B. cockerelli*.

Other insecticidal nano-systems could be designed as nano-formulations, nano-composites, and nano-encapsulation, with the objective that the insecticides are more effective and that the insecticide presents a gradual release that is synergistic and prevents indiscriminate applications.

An example of nano-encapsulation was shown in the research of Khoshraftar et al. [91] on the insecticidal activity of nano-encapsulated plant extract *Eucalyptus globulus* Labill. (Myrtaceae) investigated against *Myzus persicae* Sulzer (Hemiptera: Aphididae). They found 100% mortality with an increased solution concentration and exposure time of *Eucalyptus* extract nanocapsule. The emergent nano-technology by nano-encapsulation for the controlled release of active ingredients overcame the restrictions of plant extract usage in laboratory conditions, with a toxicity durability of *Eucalyptus* extracts reported of 8% mortality, and 72% mortality for nanocapsules for 30 days.

ZnO nanoparticles with a thiamethoxam nanocomposite were synthesized and their synergistic effect was further investigated on fourth instar larvae of *Spodoptera litura* (F.) (Lepidoptera: Noctuidae), with observations showing an increased larval mortality (27% increased mortality), a malformation in pupae and adults, overdue emergence, and reduced fecundity and fertility [92].

The growing use of NPs has led to their release into the environment, and the toxicity of metal oxide NPs on organisms has become a concern. There are still widespread controversies and ambiguities with respect to the toxic effects and mechanisms of metal oxide NPs [29]. Raliya et al. [93] indicated that there is a critical concentration of TiO_2 and ZnO nanoparticles up to which the plant's growth and development are promoted, with no improvement beyond that.

4.7. Phytotoxicity in the Plant by Nanoparticles

Regarding phytotoxicity in the plant, during the development of the present study, no physiological changes were observed or any negative impact, both in plants with NPs and in the control. Nevertheless, it is known that nanomaterials are considered a stress factor in plants since there is the possibility that they can remodel and modify the structure and constitution of membranes and cell walls in plants [63,94]. However, the phytotoxicity of NPs as a plant fertilizer and/or pesticide is determined by the applied concentration, the dissolution of its ionic forms, and the absorption and transport of the active element and its accumulation in plant tissues [68]. Once the NPs have penetrated the plant tissue, they present a slow and gradual availability of the active ingredient at the cellular level, which results in a greater accumulation of the ++ ion and, therefore, the generation of oxidative stress [95,96].

The high concentration of metallic NPs in plant tissues can influence the production of reactive oxygen species (ROS) and lipid peroxidation (MDA) [63,97–100]. ROS are reduced molecules of atmospheric oxygen like the superoxide radical $O_2\bullet$, the hydrogen peroxide

H_2O_2, the singlet oxygen O_2, and the hydroxyl radical OH•, which are highly reactive and can cause oxidative stress in organisms [101], and can affect proteins, lipids, carbohydrates, and DNA. NPs can also alter photosynthetic efficiency, photochemical fluorescence, and performance in plants, due to their interactions with photosystems I and II, since studies have shown that chlorophylls transfer energy to NPs [17,102].

According to Raliya et al. [93], aerosol- and soil-mediated exposure of TiO_2 and ZnO nanoparticles lead to varying effects on plant phenology, fruit and biomass yield, nutritional quality, and chlorophyll contents of tomato. However, they found that aerosol-mediated application was more effective than soil-mediated application, although a higher concentration (4250 mg kg^{-1}) of nanoparticles delivered by foliar application could have reached toxic levels that reduce plant height.

The main objective of this research was to evaluate the insecticidal activity of TiO_2 and ZnO nanoparticles in the short term, while long-term effects, such as phytotoxicity, were not evaluated.

Nanoparticles as insecticides are a novel and promising strategy that can be useful in pest management, in which conventional management has ceased to be effective, due to resistance problems; however, more research is required to evaluate their insecticide effects on other pests and crops. It is necessary to evaluate effective concentrations for the control that are not phytotoxic, the lethal time of NPs, their effect on non-target and beneficial insects, the possible risk of resistance development, their safety in human health, and their impact on environment.

5. Conclusions

The ZnO NPs showed insecticide activity over *B. cockerelli* second instar nymphs under laboratory conditions, particularly in concentrations ranging from 500 ppm to 3000 ppm, with mortality above 80% at 96 h.

The TiO_2 NPs under laboratory conditions showed a high insecticidal effect after 24 h, with over 93% mortality in concentrations above 100 ppm to control *B. cockerelli* second instar nymphs.

The combined action of ZnO and TiO_2 NPs over *B. cockerelli* mortality was observed at 48 h with concentrations of 150 and 250 ppm, reaching 82% mortality, and at 72 h, reaching 80% mortality from 30 ppm.

The ZnO and TiO_2 NPs, as well as the combination of these under greenhouse conditions, showed low toxicity for *B. cockerelli* second instar nymphs. The ZnO and TiO_2 NPs had 27 and 32% mortality rate after 96 h at the highest concentrations of 3000 ppm ZnO and 500 ppm TiO_2. In the combined experiment, NPs showed 23% mortality at 96 h at a concentration of 250 ppm.

The ZnO and TiO_2 NPs insecticide activity, as well as their combination, indicate their potential as control agents and could be a control alternative for their development and integration into a pest management system; however, more studies are needed in the field.

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References

1. Servicio de Información Agroalimentaria y Pesquera (SIAP). Anuario Estadístico de la Producción agrícola 2021. Available online: https://nube.siap.gob.mx/cierreagricola/ (accessed on 10 March 2021).
2. Castagna, A.; Chiavaro, E.; Dall'Asta, C.; Rinaldi, M.; Galaverna, G.; Ranieri, A. Effect of postharvest UV-B irradiation on nutraceutical quality and physical properties of tomato fruits. *Food Chem.* **2013**, *137*, 151–158. [CrossRef] [PubMed]
3. Munyaneza, J.E. Zebra Chip Disease of Potato: Biology, Epidemiology, and Management. *Am. J. Pot. Res.* **2012**, *89*, 329–350. [CrossRef]
4. CABI. *Bactericera cockerelli* (Tomato/Potato Psyllid). Invasive Species Compendium. Available online: https://www.cabi.org/isc/datasheet/45643 (accessed on 10 February 2021).
5. Garzón-Tiznado, J.A.; Cárdenas-Valenzuela, O.G.; Bujanos-Muñiz, R.; Marín-Jarillo, A.; Becerra-Flora, A.; Velarde-Félix, S.; Reyes-Moreno, C.; González-Chavira, M.; Martínez-Carrillo, J.L. Asociación de Hemiptera: Triozidae con la enfermedad permanente del tomate en México. *Agric. Técnica México* **2009**, *35*, 61–72.
6. Munyaneza, J.E.; Sengoda, V.G.; Crosslin, J.M.; Garzón-Tiznado, J.A.; Cárdenas-Valenzuela, O.G. First report of "*Candidatus* liberibacter solanacearum" in tomato plants in México. *Plant Dis.* **2009**, *93*, 1076. [CrossRef] [PubMed]
7. Liu, D.; Trumble, J.T.; Stouthamer, R. Genetic differentiation between eastern populations and recent introductions of potato psyllid (*Bactericera cockerelli*) into western North America. *Entomol. Exp. Appl.* **2006**, *118*, 177–183. [CrossRef]
8. Vega-Gutiérrez, M.T.; Rodríguez-Maciel, J.C.; Díaz-Gómez, O.; Bujanos-Muñiz, R.; Mota-Sánchez, D.; Martínez-Carrillo, J.L.; Lagunes-Tejeda, A.; Garzón-Tiznado, J.A. Susceptibilidad a insecticidas en dos poblaciones mexicanas del salerillo, *Bactericera cockerelli* (Sulc) (Hemiptera: Triozidae). *Agrociencia* **2008**, *42*, 463–471.
9. Casteel, C.L.; Walling, L.L.; Paine, T.D. Behavior and biology of the tomato psyllid, *Bactericera cockerelli*, in response to the Mi-1.2 gene. *Entomol. Exp. Appl.* **2006**, *121*, 67–72. [CrossRef]
10. Villegas-Rodríguez, F.; Marín-Sánchez, J.; Delgado-Sánchez, P.; Torres-Castillo, J.A.; Alvarado-Gómez, O.G. Management of *Bactericera cockerelli* (Sulc) (Hemiptera: Triozidae) in greenhouses with entomopathogenic fungi (Hypocreales). *Southwest. Entomol.* **2014**, *39*, 613–624.
11. Patel, N.; Desai, P.; Patel, N.; Jha, A.; Gautam, H.K. Agronanotechnology for plant fungal disease management: A Review. *Int. J. Curr. Microbiol. Appl. Sci.* **2002**, *3*, 71–84.
12. Shaker, A.M.; Zaki, A.H.; Abdel-Rahim, E.F.M.; Khedr, M.H. TiO2 nanoparticles as an effective nanopesticide for cotton leaf worm. *Agric. Eng. Int. CIGR J.* **2017**, 61–68. Available online: https://cigrjournal.org/index.php/Ejounral/article/view/4390 (accessed on 20 July 2021).
13. Rai, M.; Ingle, A. Role of nanotechnology in agriculture with special reference to management of insect pests. *Appl. Microbiol. Biotechnol.* **2012**, *94*, 287–293. [CrossRef]
14. Ali, M.A.; Rehman, I.; Iqbal, A.; Din, S.; Rao, A.Q.; Latif, A.; Samiullah, T.R.; Azam, S.; Husnain, T. Nanotechnology: A new frontier in Agriculture. *Adv. Life Sci.* **2014**, *1*, 129–138.
15. Khot, L.R.; Sankaran, S.; Maja, J.M.; Ehsani, R.; Schuster, E.W. Applications of nanomaterials in agricultural production and crop protection: A review. *Crop. Prot.* **2012**, *35*, 64–70. [CrossRef]
16. Razzaq, A.; Ammara, R.; Jhanzab, H.M.; Mahmood, T.; Hafeez, A.; Hussain, S. A Novel nanomaterial to enhance growth and yield of wheat. *J. Nanosci. Technol.* **2016**, *2*, 55–58.
17. Rico, C.M.; Peralta-Videa, J.R.; Gardea-Torresdey, J.L. Chemistry, biochemistry of nanoparticles, and their role in antioxidant defense system in plants. In *Nanotechnology and Plant Sciences: Nanoparticles and Their Impact on Plants*, 1st ed.; Siddiqui, M.H., Al-Whaibi, M.H., Mohammad, F., Eds.; Springer International: Cham, Switzerland, 2015; pp. 1–17.
18. Velasco, E.A.P.; Galindo, R.B.; Valdez-Aguilar, L.A.; González-Fuentes, J.A.; Urbina, B.A.P.; Lozano-Morales, S.A.; Sanchez-Valdes, S. Effects of the morphology, surface modification and application methods of ZnO-NPs on the growth and biomass of tomato plants. *Molecules* **2020**, *25*, 1282. [CrossRef]
19. El-Bendary, H.M.; El-Helaly, A.A. First record nanotechnology in agricultural: Silica nano-particles a potential new insecticide for pest control. *App. Sci. Rep.* **2013**, *4*, 241–246.
20. Landa, P. Positive effects of metallic nanoparticles on plants: Overview of involved mechanisms. *Plant. Physiol. Biochem.* **2021**, *161*, 12–24. [CrossRef] [PubMed]
21. Keratum, A.Y.; Abo, A.R.B.; Ismail, A.A.; George, M.N. Impact of nanoparticle zinc oxide and aluminum oxide against rice weevil *Sitophilus Oryzae* (Coleoptera: Curculionidae) under laboratory conditions. *Egy. J. Plant Pro. Res.* **2015**, *3*, 30–38.
22. Salem, A.A.; Hamzah, A.M.; El-Taweelah, N.M. Aluminum and zinc oxides nanoparticles as a new method for controlling the red flour beetles, *Tribolium castaneum* (Herbest) compared to malathion insecticide. *JPPP* **2015**, *6*, 129–137. [CrossRef]

23. De la Rosa, G.; López-Moreno, M.L.; de Haro, D.; Botez, C.E.; Peralta-Videa, J.R.; Gardea-Torresdey, J.L. Effects of ZnO nanoparticles in alfalfa, tomato, and cucumber at the germination stage: Root development and X-ray absorption spectroscopy studies. *Pure Appl. Chem.* **2013**, *85*, 2161–2174. [CrossRef]
24. García-López, J.I.; Zavala-García, F.; Olivares-Sáenz, E.; Lira-Saldívar, R.H.; Díaz Barriga-Castro, E.; Ruiz-Torres, N.A.; Ramos-Cortez, E.; Vázquez-Alvarado, R.; Niño-Medina, G. Zinc Oxide Nanoparticles Boosts Phenolic Compounds and Antioxidant Activity of *Capsicum annuum* L. during Germination. *Agronomy* **2018**, *8*, 215. [CrossRef]
25. Burman, U.; Saini, M.; Kumar, P. Effect of zinc oxide nanoparticles on growth and antioxidant system of garckpea seedlings. *Toxicol. Environ. Chem.* **2013**, *95*, 605–612. [CrossRef]
26. Laware, S.L.; Raskar, S. Influence of zinc oxide nanoparticles on growth, flowering and seed productivity in onion. *Int. J. Curr. Microbiol. Appl. Sci.* **2014**, *3*, 874–881.
27. Tirani, M.M.; Haghjou, M.M.; Ismaili, A. Hydroponic growntobacco plants respond to zinc oxide nanoparticles and bulk exposures by morphological, physiological and anatomical adjustments. *Funct. Plant Biol.* **2019**, *46*, 360–375. [CrossRef] [PubMed]
28. González-Merino, A.M.; Hernández-Juárez, A.; Betancourt-Galindo, R.; Ochoa-Fuentes, Y.M.; Valdez-Aguilar, L.A.; Limón-Corona, M.L. Antifungal activity of zinc oxide nanoparticles in *Fusarium oxysporum-Solanum lycopersicum* pathosystem under controlled conditions. *J. Phytopathol.* **2021**, 1–12. [CrossRef]
29. Chang, Y.N.; Zhang, M.; Xia, L.; Zhang, J.; Xing, G. The toxic effects and mechanisms of CuO and ZnO Nanoparticles. *Materials* **2012**, *5*, 2850–2871. [CrossRef]
30. Méndez-Argüello, B.; Vera-Reyes, I.; Mendoza-Mendoza, E.; García-Cerda, L.A.; Puente Urbina, B.A.; Lira-Saldívar, R.H. Promoción del crecimiento en plantas de *Capsicum annuum* por nanopartículas de óxido de zinc. *Nova Sci.* **2016**, *8*, 140–156. [CrossRef]
31. Servin, A.; Elmer, W.; Mukherjee, A.; De la Torre-Roche, R.; Hamdi, H.; White, J.C.; Bindraban, P.; Dimkpa, C. A Review of the use of engineered nanomaterials to suppress plant disease and enhance crop yield. *J. Nanopart. Res.* **2015**, *17*, 92. [CrossRef]
32. Hamza, A.M. Efficacy and safety of non-traditional methods as alternatives for control of *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) in rice grains. *Egypt. J. Biol. Pest Control* **2012**, *22*, 103–108.
33. Derbalah, A.S.; Khidr, A.A.; Moustafa, H.Z.; Taman, A. Laboratory evaluation of some non-conventional pest control agents against the pink bollworm *Pectinophora gossypiella* (Saunders). *Egypt. J. Biol. Pest Control* **2014**, *24*, 363–368.
34. Mostafa, W.A.; Elsayed, E.; Beall, G.W.; Rashed, S.S.; Rashed, E.M. Insecticidal effect of zinc oxide and aluminum oxide nanoparticles synthesized by co-precipitation technique on *Culex quinquefasciatus* Larvae (Diptera: Culicidae). *Int. J. Appl. Res.* **2018**, *4*, 290–297.
35. Capaldi, A.S.C.; Diniz, S.A.L.; Moretto, G.R.; Antunes, A.R.; Zezzi, A.M.A. Nanoparticles applied to plant science: A review. *Talanta* **2015**, *131*, 693–705. [CrossRef]
36. Huang, Q.; Jiao, Z.; Li, M.; Qiu, D.; Liu, K.; Shi, H. Preparation, characterization, antifungal activity, and mechanism of chitosan/TiO_2 hybrid film against *Bipolaris maydis*. *J. Appl. Polym. Sci.* **2013**, *128*, 2623–2629. [CrossRef]
37. Boxi, S.S.; Mukherjee, K.; Paria, S. Ag doped hollow TiO_2 nanoparticles as an effective green fungicide against *Fusarium solani* and *Venturia inaequalis* phytopathogens. *Nanotechnology* **2016**, *27*, 085103. [CrossRef]
38. Kolenčík, M.; Ernst, D.; Urík, M.; Ďurišová, L.; Bujdoš, M.; Šebesta, M.; Dobročka, E.; Kšiňan, S.; Illa, R.; Qian, Y.; et al. Foliar Application of Low Concentrations of Titanium Dioxide and Zinc Oxide Nanoparticles to the Common Sunflower under Field Conditions. *Nanomaterials* **2020**, *10*, 1619. [CrossRef] [PubMed]
39. Al-Bartya, A.M.; Hamza, R.Z. Larvicidal, antioxidant activities and perturbation of Transminases activities of Titanium dioxide nanoparticles synthesized using *Moring oleifera* leaves extract against the red palm weevil (*Rhynchophorus ferrugineus*). *EJPMR* **2015**, *2*, 49–54.
40. Lyu, S.; Wei, X.; Chen, J.; Wang, C.; Wang, X.; Pan, D. Titanium as a beneficial element for crop production. *Front. Plant Sci.* **2017**, *8*, 597. [CrossRef]
41. Shabbir, A.; Khan, M.M.A.; Ahmad, B.; Sadiq, Y.; Jaleel, H.; Uddin, M. Efficacy of TiO_2 nanoparticles in enhancing the photosynthesis, essential oil and khusimol biosynthesis in *Vetiveria zizanioides* L. Nash. *Photosynthetica* **2019**, *57*, 599–606. [CrossRef]
42. Khater, M.S. Effect of titanium nanoparticles (TiO_2) on growth, yield and chemical constituents of coriander plants. *Arab. J. Nucl. Sci. Appl.* **2015**, *48*, 187–194.
43. Pandey, A.C.S.; Sanjay, S.S.; Yadav, R. Application of ZnO nanoparticles in influencing the growth rate of *Cicer arietinum*. *J. Exp. Nanosci.* **2010**, *5*, 488–497. [CrossRef]
44. Rehman, H.U.; Aziz, T.; Farooq, M.; Wakeel, A. Zinc nutrition in rice production systems: A review. *Plant Soil* **2012**, *361*, 203–226. [CrossRef]
45. Shyla, K.K.; Natarajan, N. Customizing zinc oxide, silver and titanium dioxide nanoparticles for enhancing groundnut seed quality. *Indian J Sci Technol.* **2014**, *7*, 1376–1381. [CrossRef]
46. Kriti, A.; Ghatak, A.; Mandal, N. Antimycotic efficacy of Zinc nanoparticle on dark-spore forming Phytopathogenic fungi. *J. Pharmacogn. Phytochem.* **2020**, *9*, 750–754.
47. Mayo-Hernández, J.; Flores-Olivas, A.; Valenzuela-Soto, J.; Rodríguez-Pagaza, Y.; Vega-Chávez, J.; Hernández-Castillo, F.; Aguirre-Uribe, L. *Bactericera cockerelli* Sulc oviposition preference and development on three tomato varieties. *Southwest. Entomol.* **2018**, *43*, 905–910. [CrossRef]

48. Betancourt, G.R.; Berlanga, D.M.L.; Puente, U.B.; Rodríguez, F.O.; Sanchez-Valdes, S. Surface modification of ZnO nanoparticles. *Mater. Sci. Forum* **2010**, *644*, 61–64. [CrossRef]
49. IRAC (Insecticide Resistance Action Committee). IRAC Susceptibility Test Method 002. 2021. Available online: https://irac-online.org/methods/psylla-spp-all-stages/ (accessed on 16 February 2021).
50. Abbott, W.S. A method for computing the effectiveness of an insecticide. *J. Econ. Entomol.* **1925**, *18*, 265–267. [CrossRef]
51. S.A.S. Institute. *The SAS System for Windows, Release 9.0*; SAS Institute: Cary, NC, USA, 2002.
52. Betancur, H.C.P.; Hernández, M.V.; Buitrago, S.R. Nanopartículas para materiales antibacterianos y aplicaciones del dióxido de titanio. *Rev. Cuba Investig. Biomed.* **2016**, *35*, 387–402.
53. Naderi, M.R.; Shahraki, D.A. Nanofertilizers and their roles in sustainable agriculture. *Int. J. Agric. Crop Sci.* **2013**, *5*, 2229–2232.
54. Raskar, S.V.; Laware, S.L. Effect of zinc oxide nanoparticles on cytology and seed germination in onion. *Int. J. Curr. Microbiol. Appl. Sci.* **2014**, *3*, 467–473.
55. Dimkpa, C.O.; McLean, J.E.; David, W.; Britt, D.W.; Anderson, A.J. Nano CuO and interaction with nano-ZnO or soil bacterium provide evidence for the interference of nanoparticles in metal nutrition of plants. *Ecotoxicology* **2015**, *24*, 119–129. [CrossRef]
56. Wang, S.; Liu, H.; Zhang, Y.; Xin, H. Effect of CuO nanoparticles on reactive oxygen species and cell cycle gene expression in roots of rice. *Environ. Toxicol. Chem.* **2015**, *34*, 554–561. [CrossRef]
57. Prasad, T.N.V.K.V.; Sudhakar, P.; Sreenivasulu, Y.; Latha, P.; Munaswamy, V.; Reddy, K.R.; Pradeep, T. Effect of nanoscale zinc oxide particles on the germination, growth and yield of peanut. *J. Plant Nutr.* **2012**, *35*, 905–927. [CrossRef]
58. Raliya, R.; Tarafdar, J.C. ZnO nanoparticle biosynthesis and its effect on phosphorous-mobilizing enzyme secretion and gum contents in Clusterbean (*Cyamopsis tetragonoloba* L.). *Agric. Res.* **2013**, *2*, 48–57. [CrossRef]
59. Ramesh, M.; Palanisamy, K.; Babu, K.; Sharma, N.K. Effects of bulk and nano-titanium dioxide and zinc oxide on physiomorphological changes in *Triticum aestivum* Linn. *J. Glob. Biosci.* **2014**, *3*, 415–422.
60. Lei, Z.; Mingyu, S.; Xiao, W. Antioxidant stress is promoted by nano-anatase in spinach chloroplasts under UV-B radiation. *Biol. Trace Elem. Res.* **2008**, *121*, 69–79. [CrossRef]
61. Khan, M.N. Nano-titanium Dioxide (Nano-TiO_2) mitigates NaCl stress by enhancing antioxidative enzymes and accumulation of compatible solutes in tomato (*Lycopersicon esculentum* Mill.). *J. Plant. Sci.* **2016**, *11*, 1–11. [CrossRef]
62. Latef, A.A.H.A.; Srivastava, A.; El-sadek, M.S.A.; Kordrostami, M.; Tran, L.P. Titanium dioxide nanoparticles improve growth and enhance tolerance of broad bean plants under saline soil conditions. *Land. Degrad. Dev.* **2018**, *29*, 1065–1073. [CrossRef]
63. Gohari, G.; Mohammadi, A.; Akbari, A.; Panahirad, S.; Reza, D.M.; Fotopoulos, V.; Kimura, S. Titanium dioxide nanoparticles (TiO_2 NPs) promote growth and ameliorate salinity stress effects on essential oil profile and biochemical attributes of *Dracocephalum moldavica*. *Sci. Rep.* **2020**, *10*, 912. [CrossRef]
64. Servin, A.D.; Morales, M.I.; Castillo-Michel, H.; Hernandez-Viezcas, J.A.; Muñoz, B.; Zhao, L.; Nuñez, J.E.; Peralta-Videa, J.R.; Gardea-Torresdey, J.L. Synchrotron verification of TiO_2 accumulation in cucumber fruit: A possible pathway of TiO_2 nanoparticle transfer from soil into the food chain. *Environ. Sci. Technol.* **2013**, *47*, 11592–11598. [CrossRef]
65. Reed, R.B.; Ladner, D.A.; Higgins, C.P.; Westerhoff, P.; Ranville, J.F. Solubility of nano-zinc oxide in environmentally and biologically important matrices. *Environ. Toxicol. Chem.* **2012**, *31*, 93–99. [CrossRef] [PubMed]
66. Peng, C.; Duan, D.; Xu, C.; Chen, Y.; Sun, L.; Zhang, H.; Yuan, X.; Zheng, L.; Yang, Y.; Yang, J.; et al. Translocation and biotransformation of CuO nanoparticles in rice (*Oryza sativa* L.) plants. *Environ. Pollut.* **2015**, *197*, 99–107. [CrossRef] [PubMed]
67. Gupta, N.; Ram, H.; Kumar, B. Mechanism of Zinc absorption in plants: Uptake, transport, translocation and accumulation. *Rev. Environ. Sci. Biotechnol.* **2016**, *15*, 89–109. [CrossRef]
68. Ruiz-Torres, N.; Flores-Naveda, A.; Barriga-Castro, E.D.; Camposeco-Montejo, N.; Ramírez-Barrón, S.; Borrego-Escalante, F.; Niño-Medina, G.; Hernández-Juárez, A.; Garza-Alonso, C.; Rodríguez-Salinas, P.; et al. Zinc Oxide Nanoparticles and Zinc Sulfate Impact Physiological Parameters and Boosts Lipid Peroxidation in Soil Grown Coriander Plants (*Coriandrum sativum*). *Molecules* **2021**, *26*, 1998. [CrossRef] [PubMed]
69. Fleischer, A.; O'Neill, M.A.; Ehwald, R. The pore size of non-graminaceous plant cell walls is rapidly decreased by borate ester cross-linking of the pectic polysaccharide rhamnogalacturonan II. *Plant Physiol.* **1999**, *121*, 829–838. [CrossRef]
70. Eichert, T.; Kurtz, A.; Steiner, U.; Goldbach, H.E. Size exclusion limits and lateral heterogeneity of the stomatal foliar uptake pathway for aqueaus solutes and water suspended nanoparticles. *Physiol. Plant.* **2008**, *134*, 151–160. [CrossRef]
71. Vittori, A.L.; Carbone, S.; Gatti, A.; Vianello, G.; Nannipieri, P. Uptake and translocation of metals and nutrients in tomato grown in soil polluted with metal oxide (CeO_2, Fe_3O_4, SnO_2, TiO_2) or metallic (Ag, Co, Ni) engineered nanoparticles. *Environ Sci Pollut Res.* **2014**, *22*, 1841–1853. [CrossRef]
72. Wazid; Nadagouda, S.; Prabhuraj, A.; Naik, R.H.; Shakuntala, N.M.; Sharanagouda, H. Effect of Biosynthesized Zinc Oxide Green Nanoparticles on Pulse Beetle, *Callosobruchus analis* (Coleoptera: Chrysomelidae). *Int. J. Curr. Microbiol. App. Sci.* **2018**, *7*, 503–512. [CrossRef]
73. Mekewi, M.; Shebl, A.; Imam, A.I.; Amin, M.; Albert, T. Screening the insecticidal efficacy of nano ZnO synthesized via in-situ polymerization of cress linked polycyclic acid as a template. *J. Mater Sci. Technol.* **2012**, *28*, 961–968. [CrossRef]
74. Rouhani, M.; Samih, M.A.; Kalantari, S. Insecticide effect of silver and zinc nanoparticles against *Aphis nerii* Boyer de Fonscolombe (Hemiptera: Aphididae). *Chil. J. Agric. Res.* **2012**, *72*, 590–954. [CrossRef]
75. Shaker, A.M.; Zaki, A.H.; Abdel-Rahim, E.F.; Khedr, M.H. Novel CuO nanoparticles for pest management and pesticides photodegradation. *Environ. Biol.* **2016**, *10*, 274–283.

76. Khooshe-Bast, Z.; Sahebzadeh, N.; Ghaffari-Moghaddam, M.; Mirshekar, A. Insecticidal effects of zinc oxide nanoparticles and *Beauveria bassiana* TS11 on *Trialeurodes vaporariorum* (Westwood, 1856) (Hemiptera: Aleyrodidae). *Acta Agric. Slov.* **2016**, *107*, 299–309. [CrossRef]
77. Jovanović, B.; Cvetković, V.J.; Mitrović, T.L. Effects of human food grade titanium dioxide nanoparticle dietary exposure on *Drosophila melanogaster* survival, fecundity, pupation and expression of antioxidant genes. *Chemosphere* **2016**, *144*, 43–49. [CrossRef]
78. Ishwarya, R.; Vaseeharan, B.; Kalyania, S.; Banumathia, B.; Govindarajan, M.; Alharbi, N.S.; Kadaikunnand, S.; Al-anbr, M.N.; Khaled, J.M.; Benelli, G. Facile green synthesis of zinc oxide nanoparticles using *Ulva lactuca* seaweed extract and evaluation of their photocatalytic, antibiofilm and insecticidal activity. *J. Photochem. Photobiol. B. Biol.* **2018**, *178*, 249–258. [CrossRef]
79. Al-Dhabi, N.A.; Arasu, M.V. Environmentally-Friendly Green Approach for the Production of Zinc Oxide Nanoparticles and Their Anti-Fungal, Ovicidal, and Larvicidal Properties. *Nanomaterials* **2018**, *8*, 500. [CrossRef]
80. Alisha, A.S.; Thangapandiyan, S. Comparative bioassay of silver nanoparticles and malathion on infestation of red flour beetle, *Tribolium castaneum*. *JoBAZ* **2019**, *80*, 1–10.
81. Zorlu, T.; Nurullahoğlu, Z.U.; Altuntaş, H. Influence of dietary titanium dioxide nanoparticles on the biology and antioxidant system of model insect, *Galleria mellonella* (L.) (Lepidoptera: Pyralidae). *J. Entomol. Res. Soc.* **2018**, *20*, 89–103.
82. López-Muñoz, D.; Ochoa-Zapater, M.A.; Torreblanca, A.; Garcerá, M.D. Evaluation of the effects of titanium dioxide and aluminum oxide nanoparticles through tarsal contact exposure in the model insect *Oncopeltus fasciatus*. *Sci. Total Environ.* **2019**, *666*, 759–765. [CrossRef]
83. Eskin, A.; Öztürk, Ş.; Körükçü, M. Determination of the acute toxic effects of zinc oxide nanoparticles (ZnO NPs) in total hemocytes counts of *Galleria mellonella* (Lepidoptera: Pyralidae) with two different methods. *Ecotoxicology* **2019**, *28*, 801–808. [CrossRef] [PubMed]
84. Haroun, S.A.; Elnaggar, M.E.; Zein, D.M.; Gad, R.I. Insecticidal efficiency and safety of zinc oxide and hydrophilic silica nanoparticles against some stored seed insects. *J. Plant Prot. Res.* **2020**, *60*, 77–85.
85. Zayed, G.M.M.; EL-Sagheer, S.M.; Hussain, H.B.H. Aluminum and silica oxides nanoparticles as a new approach for control the red flour beetle *Tribolium castaneum* (Coleoptera: Tenebrionidae) on wheat grains. *Egypt. J. Plant Prot. Res. Inst.* **2020**, *3*, 281–289.
86. Shukla, G.; Gaurav, S.S.; Singh, A. Synthesis of mycogenic zinc oxide nanoparticles and preliminary determination of its efficacy as a larvicide against white grubs (*Holotrichia* sp.). *Int. Nano Lett.* **2020**, *10*, 131–139. [CrossRef]
87. Abd El-Wahab, A.S.; El-Bendary, H.M.; El-Helaly, A.A. Nano silica as a promising nano pesticide to control three different aphid species under semi-field conditions in Egypt. *Egypt. Acad. J. Biol. Sci. F. Toxicol. Pest Control.* **2016**, *8*, 35–49. [CrossRef]
88. Völker, C.; Oetken, M.; Oehlmann, J. The biological effects and possible modes of action of nanosilver. *Rev. Environ. Contam. Toxicol.* **2013**, *223*, 81–106. [PubMed]
89. Debnath, N.; Sumistha, D.; Seth, D.; Chandra, R.; Bhattacharya, S.C.; Goswami, A. Entomotoxic effect of silica nanoparticles against *Sitophilus oryzae* (L.). *J. Pest Sci.* **2011**, *84*, 99–105. [CrossRef]
90. Benelli, G. Mode of action of nanoparticles against insects. *Environ Sci Pollut Res.* **2018**, *25*, 12329–12341. [CrossRef] [PubMed]
91. Khoshraftar, Z.; Safekordi, A.A.; Shamel, A.; Zaefizadeh, M. Synthesis of natural nanopesticides with the origin of *Eucalyptus globulus* extract for pest control. *Green Chem. Lett. Rev.* **2019**, *12*, 286–298. [CrossRef]
92. Jameel, M.; Shoeb, M.; Khan, M.T.; Ullah, R.; Mobin, M.; Farooqi, M.K.; Adnan, S.M. Enhanced insecticidal activity of thiamethoxam by zinc oxide nanoparticles: A novel nanotechnology approach for pest control. *ACS Omega* **2020**, *5*, 1607–1615. [CrossRef]
93. Raliya, R.; Nair, R.; Chavalmane, S.; Wang, W.N.; Biswas, P. Mechanistic evaluation of translocation and physiological impact of titanium dioxide and zinc oxide nanoparticles on the tomato (*Solanum lycopersicum* L.) plant. *Metallomics* **2015**, *7*, 1584–1594. [CrossRef] [PubMed]
94. Liu, Q.; Zhang, X.; Zhao, Y.; Lin, J.; Shu, C.; Wang, C.; Fang, X. Fullerene-induced increase of glycosyl residue on living plant cell wall. *Environ. Sci. Technol.* **2013**, *47*, 7490–7498. [CrossRef]
95. Gaschler, M.M.; Stockwell, B.R. Lipid peroxidation in cell death. Biochem. *Biophys Res.* **2017**, *482*, 419–425. [CrossRef]
96. Mosavat, N.; Golkar, P.; Yousefifard, M.; Javed, R. Modulation of callus growth and secondary metabolites in different *Thymus* species and *Zataria multiflora* micropropagated under ZnO nanoparticles stress. *Biotechnol. Appl. Biochem.* **2019**, *66*, 316–322. [CrossRef]
97. Huang, C.; Verrillo, F.; Renzone, G. Response to biotic and oxidative stress in Arabidopsis thaliana: Analysis of variably phosphorylated proteins. *J. Proteom.* **2011**, *74*, 1934–1949. [CrossRef]
98. Pullagurala, V.L.R.; Adisa, I.O.; Rawat, S.; Kalagara, S.; Hernandez-Viezcas, J.A.; Peralta-Videa, J.R.; Gardea-Torresdey, J.L. ZnO nanoparticles increase photosynthetic pigments and decrease lipid peroxidation in soil grown cilantro (*Coriandrum sativum*). *Plant Physiol. Biochem.* **2018**, *132*, 120–127. [CrossRef] [PubMed]
99. García-López, J.I.; Niño-Medina, G.; Olivares-Sáenz, E.; Lira-Saldivar, R.H.; Barriga-Castro, E.D.; Vázquez-Alvarado, R.; Rodríguez-Salinas, P.A.; Zavala-García, F. Foliar application of zinc oxide nanoparticles and zinc sulfate boosts the content of bioactive compounds in habanero peppers. *Plants* **2019**, *8*, 254. [CrossRef]
100. Kalal, P.R.; Jajoo, A. Priming with zinc oxide nanoparticles improve germination and photosynthetic performance in wheat. *Plant. Physiol. Biochem.* **2021**, *160*, 341–351. [CrossRef] [PubMed]

101. Mittler, R. ROS are good. *Trends Plant Sci.* **2017**, *22*, 11–19. [CrossRef] [PubMed]
102. Olejnik, M.; Krajnik, B.; Kowalska, D.; Twardowska, M.; Czechowski, N.; Hofmann, E.; Mackowski, S. Imaging of fluorescence enhancement in photosynthetic complexes coupled to silver nanowires. *Appl. Phys. Lett.* **2013**, *102*, 083703. [CrossRef]