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Silica nanoparticle effect on an internal feeder, American serpentine leafminer

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Abstract

Insects quickly develop resistance to conventional chemical insecticides. The use of silica nanoparticles (SiO_2NPs) is a promising new approach for managing insect pests. The insects that have been studied are, in most cases, external feeders and internal leaf feeders have not been well studied. Here, we investigate the efficiency of SiO_2NPs in controlling the American serpentine leafminer *Liriomyza trifolii* (Diptera: Agromyzidae), a devastating insect pest of a wide range of crops. SiO_2NPs at concentrations of 50, 100, 200 and 400 mg/L compared to a control of distilled water were applied to intact *Phaseolus vulgaris* leaves by spraying on two-week-old intact seedlings to evaluate their effects, via the plant, on the feeding, survival and body mass of *L. trifolii* released after spraying. A qRT-PCR analysis was conducted to assess oxidative stress in *L. trifolii* based on the gene expression level of the two major antioxidant enzymes; catalase and superoxide dismutase. The survival rate of larvae was lower than the control at the highest concentration of SiO_2NPs, and both larval feeding velocity and pupal weight decreased at the high SiO_2NPs concentrations. Gene expression levels of the antioxidant enzymes at the pupal stage were not significantly affected by SiO_2NPs at any concentration but some individuals showed up-regulated gene expression level at low SiO_2NP concentrations, indicating development of resistance. This study suggests that a high concentration of SiO_2NPs should be applied to reduce a leafminer population while avoiding development of resistance.

Keywords Silica nanoparticles • *Liriomyza trifolii* • *Phaseolus vulgaris* • qRT-PCR • Oxidative stress • Dose dependency

Key message

- Silica nanoparticles effect on internal leaf feeders has not been well studied.
- A high concentration (400 mg/L) of silica nanoparticles reduced the larval survival, feeding velocity and weight of the leafminer *Liriomyza trifolii* when applied to intact *Phaseolus vulgaris* leaves before leafminer release
- Gene expression levels of two antioxidant enzymes were high in some individuals at low concentrations, indicating development of resistance
- The high concentration of silica nanoparticles can be used for reducing leafminer populations

Introduction

To date, synthetic insecticides are the main strategy in the management of insect pests. Despite their efficacy, these chemical substances raise concerns of increasing insect resistance over successive generations. Moreover, the widespread and haphazard use of synthetic pesticides has negative effects on non-target organisms and poses a threat to human health. The use of nanoparticles (less than 100 nm in size in all three dimensions) in pest management can overcome the emerging problems of insecticide resistance by reducing the high-volume application of traditional chemical insecticides (Rani et al. 2014).

Silicon dioxide nanoparticles (SiO_2NPs) are the most popular among numerous types of nanomaterials (materials less than 100 nm in size at least in one dimension) and ensure definite interactions between

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molecules through a mesoporous structure carrying specific functional groups (Kaziem et al. 2017). SiO_2NPs are a stable and environmentally safe approach to pest management not only because of their versatility as an insecticide but also for improving the activity of other chemical insecticides. Neither the World Health Organization (WHO) nor the US Department of Agriculture have indicated any significant negative effects of SiO_2NPs on human health (El-Naggar et al. 2020). Depending on their application, size, and concentration, SiO_2NPs might be a viable alternative to conventional pesticides. SiO_2NPs are believed to interrupt insect physiological functions through respiratory blockage, cuticular damage, desiccation and injuries to the midgut epithelium (Caceres et al. 2019) in addition to altering enzymatic protection from oxidative stress. Studies that investigate the mechanisms by which SiO_2NPs affect the behavior, life history traits and gene expression of target organisms are critical in furthering our understanding of the entomotoxic effects of these nanoparticles and their applications in pest control.

The available literature on the entomotoxic effects of SiO_2NPs focuses on insect pests of stored grains, such as *Sitophilus oryzae* (Goswami et al. 2010; Debnath et al. 2011), *Callosobruchus maculatus* (Rouhani et al. 2012; Arumugam et al. 2016), and *Corcyra cephalonica* (Vani and Brindhaa 2013). More recently, entomotoxic effects have also been tested on insect pests in field settings, including the lepidopterans *Spodoptera littoralis* (El-Samahy et al. 2015; Ayoub et al. 2017), *Spodoptera litura*, *Achaea janata* (Rani et al. 2014), *Mythimna separata* (Mousa et al. 2014) and *Plutella xylostella* (Shoaib et al. 2018; Bilal et al. 2020), as well as the aphids *Lipaphis pseudobrassicae* (Debnath et al. 2010), *Aphis gossypii* (Pavitra et al. 2018), and the mealybug *Phenacoccus solenopsis* (Pavitra et al. 2018). These insects are external feeders and internal leaf feeders such as leafminers have not been well studied of SiO_2NP effects. As an intraspecific variation, internal feeding developmental stage (the larva) of seed feeders is less affected than external feeding stage (the adult) as was observed in *C. maculatus* (Rouhani et al. 2012). The American serpentine leafminer *Liriomyza trifolii* (Diptera: Agromyzidae) is a highly polyphagous species and one of the most damaging leafminers found in tropical and subtropical regions. *Liriomyza trifolii* larvae consume leaf mesophyll tissue, which interrupts the photosynthetic process in and allows for the spread of diseases. This leads to a decrease in crop production, including production of the common bean *Phaseolus vulgaris* (Ibrahim 2008; Yıldırım et al. 2010).

To determine the usefulness of SiO_2NPs in controlling the leafminer *L. trifolii*, we investigated (1) the effect of SiO_2NPs on the feeding velocity, survival and body mass of developing *L. trifolii* on plant treated with SiO_2NPs and (2) the expected genotoxicity induced by SiO_2NPs on the expression of oxidative stress genes in *L. trifolii*. This study may be the first report on the entomotoxic effect of SiO_2NPs on internally feeding insects, dipteran leafminers.

Materials and methods

Silicon dioxide nanoparticles

A white powder of silicon dioxide (SiO_2) nanoparticles (NPs) [99.5% purity, 19.6 ± 5.8 nm (mean ± SD) in size and spherical in shape] were obtained from US Research Nanomaterials, Inc. (Houston, Texas, USA). A stock solution (1000 mg/L) of SiO_2NPs was prepared by dissolving the powder of SiO_2NPs in distilled water.

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This solution was sonicated for 30 min and centrifuged (2000 rpm, 25°C) for another 30 min to precipitate the non-dispersed agglomerated particles, which were filtered out of the solution using filter paper No. 2 (90 mm, Advantec, Japan). Then, four different concentrations of SiO_2NPs—50, 100, 200 and 400 mg/L—were prepared for investigating the toxic and genotoxic effects against the insect pest *L. trifolii*. The size and shape of SiO_2NPs were inspected using a high-resolution transmission electron microscope (TEM) (JEM-2100, JEOL Ltd.) at an accelerating voltage of 200 kV.

Toxicity to the pest insect *L. trifolii*

Stock culture

A culture of *L. trifolii* was maintained under constant laboratory conditions (25 ± 3 °C, 50 ± 10% RH and a 12L:12D photoperiod). Insects were reared inside transparent cuboid cages (70 × 50 × 50 cm) with a window (20 × 20 cm) on both sides and two windows (50 × 20 cm) on the back; all windows were covered with mesh to ensure sufficient ventilation. Two trays containing about 40 plants of the common bean *Phaseolus vulgaris* (a preferred host) [seeds obtained from Hokkaido, Japan by Nakahara Seed Co., Ltd. (Fukuoka, Japan)] were introduced daily. This allowed for continuous oviposition by *L. trifolii*, providing pupae to be used subsequently. *Phaseolus vulgaris* was planted weekly under long-day conditions (21–27°C and a 16L:8D photoperiod).

Biological parameters

Two-week-old healthy *P. vulgaris* plants were sprayed with one of the prepared concentrations of SiO_2NPs (5 ml/plant) or with distilled water alone as a control treatment. Plants were sprayed using an ordinary drizzle (drop size of 1.05 ± 0.13 mm) at a distance of 15 cm to ensure a full coverage of both the upper and lower surfaces of the leaves (two leaves/plant). Plants were then left to dry at room temperature inside a transparent cylindrical cage (70 cm height × 25 cm diameter) covered with a fine mesh at the top in a randomized complete block design (RCBD) with six replications. After one hour, 48 h old male and female *L. trifolii* were released into the previously described cages (six for each treatment) to feed and lay eggs. After 24 h, the adults were removed and the eggs were allowed to complete their life cycle. To evaluate the toxic effect of SiO_2NPs, three biological parameters were recorded; larval survival rate, larval feeding velocity and pupal weight.

Larval survival rate

The total number of mines during the early larval stage on each day was counted for four days from the release of the adults. The number of pupae was counted every day for three days after the first pupa emerged. The larval survival rate was derived by dividing the number of pupae by the number of mines during the early larval stage.

Larval feeding velocity

One mine was randomly chosen for each replicate and digital pictures of the mine on the 2nd and 3rd days were captured. The length of the mines in both images were measured by dividing each mine (winding feeding tunnel, Ayabe et al. 2008) into short, straight lines (and by calibrating with the reference scale of each picture). Then, the larval feeding velocity was calculated as a measure of larval activity as

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$$\text{Larval feeding velocity (LFV)} = \frac{L2-L1}{\Delta T},$$

where L1 and L2 refer to the lengths of the mine on the 2nd and 3rd days, respectively, and ΔT is the time between the two images. One mine per replicate and six replicates per treatment were measured.

Pupal weight

Five pupae (24 h old) per replicate were randomly selected and weighed with a microbalance (AT-20, Mettler Toledo, Switzerland). Afterwards, the pupae were stored in a freezer at −20 °C for future genetic analysis.

Quantitative RT-PCR analysis of gene expression

The expression levels of two major antioxidant enzymes, catalase (*CAT*) and superoxide dismutase (*SOD*), in *L. trifolii* pupae (48 h old) as a response to the SiO_2NPs treatments were examined using qRT-PCR. Pupae was individually homogenized in the homogenizer (BHA-6, As One) with Isogen II (Nippon Gene) and zirconia beads (3,000 rpm, 1 min). Total RNA was extracted from the homogenate and used as a template for RT-PCR. The cDNAs were synthesized using SuperScript IV VILO Master Mix (Thermo Fisher Scientific) according to the manufacturer's instruction. The target genes were amplified using recombinant Taq DNA polymerase and the tracer EvaGreen (Biotium) with gene-specific primer sets obtained from FlyPrimerBank (Hu et al. 2013); catalase (CG6871): forward (F): 5′-GATGCGGCTTCCAATCAGTTG-3′ and reverse (R): 5′-GCAGCAGGATAGGTCCTCG-3′; superoxide dismutase 2 (CG8905): F: 5′-AAGTCGGGCAAACTGCAACT-3′ and R: 5′-GGACGCACGTTCTTGTACTG-3′. As a reference gene, β-actin was chosen and amplified with the following primer set: F: 5′- TTGTATTGGACTCTGGTGACGG-3′ and R: 5′- GATAGCGTGAGGCAAAGCATAA-3′ (Chang et al. 2017). Negative controls containing water instead of cDNA template were included for each primer set. The amplification plots were analyzed in the StepOnePlus real-time PCR system (Applied Biosystems) with the following cycling conditions: 94 °C for 3 min followed by 40 cycles consisting of 94 °C for 10 s, 60 °C for 15 s, and 72 °C for 15 s. Fluorescence readings were taken at the end of each cycle. The melting curve protocol contained 1 cycle at 95 °C for 15 s and 60 °C for 1 min. The temperature was increased from 60 °C to 95 °C at a rate of 0.3 °C s^{-1}. The gene expression level was normalized by dividing by the mean gene expression level of the control. Three pupae were analyzed per treatment.

Statistical analysis

The following methods were applied to test the effect of different concentrations of SiO_2NPs on *L. trifolii*. Data of insect biological parameters (larval feeding velocity and pupal weight) were analyzed with ANOVAs after confirming the normality of data distributions. A posthoc Dunnett test was performed for multiple comparisons with the control. Larval survival rate was analyzed with a logistic regression model with a logit link function and followed by a posthoc pairwise comparison with a Bonferroni correction. Kruskal-Wallis tests were used to compare gene expression levels. All statistical tests were performed using JMP 13.2.1.

Results

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Toxicity to the insect pest *L. trifolii*

Larval survival rate

The larval survival rate was affected by SiO_2NP treatments (likelihood ratio $\chi^2_4 = 25.25$, $P < 0.0001$). Survival was lower than the control at a concentration of 400 mg/L (Fig. 1).

Larval feeding velocity (LFV)

The larval feeding velocity was affected by SiO_2NP treatments ($F_{4,25} = 8.60$, $P = 0.0002$). Feeding velocity was significantly lower than the control at concentrations of 200 and 400 mg/L (Fig. 2).

Pupal weight

The pupal weight (as a mean of five late-stage pupae) showed significant differences among SiO_2NPs concentrations ($F_{4,25} = 8.38$, $P = 0.0002$, Fig. 3) and was lower than the control at concentrations of 50, 200 and 400 mg/L. This pattern partially reflects the LFV results.

Gene expression

No concentration-dependent impacts of SiO_2NPs were observed on the expression levels of catalase or superoxide dismutase genes (catalase, $H_{3,3,3,3,3} = 3.33$, $P > 0.1$; superoxide dismutase, $H_{3,3,3,3,3} = 4.77$, $P > 0.1$). This is due to the large variation in gene expression levels among individuals (surviving pupae) in each treatment (Fig. 4). At concentrations of 50 and 100 mg/L, some pupae showed up-regulated gene expression for catalase compared to the control, whereas the gene expression levels of superoxide dismutase were relatively constant across the different SiO_2NP concentrations.

Discussion

The larval survival rate, feeding velocity and pupal weight were all correlated and lower at higher SiO_2NP concentrations compared to the control; since feeding affects the development of later stages in the life cycle of an insect, a higher feeding velocity contributes to an increase in pupal weight. Gene expression levels were not significantly different among SiO_2NP concentrations but some individuals showed higher expression levels than the control. The amount of time since the initial exposure to SiO_2NPs may have allowed for the development of resistance in surviving immature *L. trifolii*. Up-regulated individuals might have had a genetic background that allowed for higher resistance against the toxic effects of feeding on SiO_2NP-treated plant tissues as larvae.

When applied externally, SiO_2NPs kill insects by dehydration (Ayoub et al. 2017; Shoaib et al. 2018). Other possible mechanisms of SiO_2NP-induced insect mortality include the blockage of spiracles and tracheae (part of the respiratory system) or damage to the surface wax on the outer cuticle of the insect by sorption (physisorption) and abrasion (Rastogi et al. 2019). In this study, however, SiO_2NPs did not contact the outer cuticle of the insect *L. trifolii* since the nanoparticle solution was applied to the plant prior to infestation by *L. trifolii*. Silicon absorbed and deposited in plant epidermal tissues (e.g. cell walls, lumen, intracellular space and trichomes, Cooke and Leishman 2011) can increase the rigidity of the tissue, thereby increasing its

mechanical resistance to herbivory and causing abnormal level of wear on mandibles of the insects that feed on it (Painter 1951; Sasamoto 1955; Takahashi 1995; Keeping and Meyer 2002; Reynolds et al. 2009; Massey and Hartley 2009). Silica may also cause impairment of the digestive tract in insect herbivores (Smith 1969). Additionally, the application of silicon can enhance natural defense system of the host plant by producing increased quantities of flavonoids and phenolic acids (Fawe et al. 1998) and by promoting jasmonate-mediated defenses (Ye et al. 2013) in response to herbivory (Coscun et al. 2019). Similar effects and possibly faster action and response are expected for nano-size silica.

Based on our results (Figs. 1, 2, and 3), we can recommend applying a moderately high concentration (400 mg/L) of SiO_2NPs to reduce the leafminer *L. trifolii* population while avoiding development of resistance and without damaging *P. vulgaris* (promoted seedling growth at 400 mg/L but negative effects on growth at 2000 and 4000 mg/L, Sharifi-Rad et al. 2016). For a more accurate estimation of the overall effects of SiO_2NPs on *L. trifolii*, future studies should investigate the M_2 generation for adult fecundity, offspring survival and the effects of directly applying SiO_2NPs to different developmental stages.

Author Contribution Statement

Conceptualization, A.F.T., O.A.G. and M.T.; investigation, A.F.T. and M.H.; methodology, A.F.T., M.T., R.F. and M.H.; statistical analysis, M.T.; writing—original draft preparation, A.F.T., M.T. and R.F.; writing—review and editing, M.T. and O.A.G.; visualization, A.F.T., M.T. and R.F.; supervision, M.T., O.A.G. and M.F.M.E. All authors have read and agreed to the published version of the manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This article does not contain any studies with animals performed by any of the authors.

Informed consent Informed consent was obtained from all co-authors included in the study.

References

Arumugam G, Velayutham V, Shanmugavel S, Sundaram J (2016) Efficacy of nanostructured silica as a stored pulse protector against the infestation of bruchid beetle, *Callosobruchus maculatus* (Coleoptera: Bruchidae). *Appl Nanosci* 6:445–450

Ayabe Y, Tuda M, Mochizuki A (2008) Benefits of repeated mine trackings by a parasitoid of tortuous feeding pattern of a host leafminer. *Anim Behav* 76:1795–1803

Ayoub H A, Khairy M, Rashwan FA, Abdel-Hafez HF (2017) Synthesis and characterization of silica nanostructures for cotton leaf worm control. *J Nanostruct Chem* 7:91–100

Azimi R, Borzelabad MJ, Feizi H, Azimi A (2014) Interaction of SiO_2 nanoparticles with seed prechilling on germination and early seedling growth of tall wheatgrass (*Agropyron elongatum* L.). *Pol J Chem Tech* 16:25–29

Bao-shan L, Chun-hui L, Li-jun F, Shu-chun Q, Min Y (2004) Effect of TMS (nanostructured silicon dioxide) on growth of Changbai larch seedlings. *J For Res (Harbin)* 15:138–140

Bilal M, Xu C, Cao L, Zhao P, Cao C, Li F, Huang Q (2020) Indoxacarb-loaded fluorescent mesoporous silica nanoparticles for effective control of *Plutella xylostella* L. with decreased detoxification enzymes activities. *Pest Manag Sci* 76:3749–3758

Caceres M, Vassena CV, Garcera MD, Santo-Orihuela PL (2019) Silica nanoparticles for insect pest control. *Curr Pharm Design* 25:4030–4038

Chang YW, Chen JY, Lu MX, Gao Y, Tian ZH, Gong WR, Dong CS, Du YZ (2017) Cloning and expression of genes encoding heat shock proteins in *Liriomyza trifolii* and comparison with two congener leafminer species. *PLoS ONE* 12:e0181355

Cooke J, Leishman MR (2011) Silicon concentration and leaf longevity: is silicon a player in the leaf dry mass spectrum. *Funct Ecol* 25:1181–1188

Coscun D, Deshmukh R, Sonah H, Menzies JZ, Reynolds O, Ma JF, Kronzucker HJ, Bélanger RR (2019) The controversies of silicon's role in plant biology. New Phytol 221:67–85

Dahindwal AS, Lather BPS, Singh J (1991) Efficacy of seed treatment on germination, seedling emergence and vigor of cotton (*Gossypium hirsutum*) genotypes. *Seed Res*19:59–61

Debnath N, Das S, Brahmachary RL, Chandra R, Sudan S, Goswami A (2010) Entomotoxicity assay of silica, zinc oxide, titanium dioxide, aluminium oxide nanoparticles on *Lipaphis pseudobrassicae*. *AIP Conference Proceedings* 1276:307–310

Debnath N, Das S, Seth D, Chandra R, Bhattacharya SC, Goswami A (2011) Entomotoxic effect of silica nanoparticles against *Sitophilus oryzae* (L.). *J Pest Sci* 84:99–105

El-Naggar ME, Abdelsalam NR, Fouda MMG, Mackled MI, Al-Jaddadi MAM, Ali HM, Siddiqui MH, Kandil EE (2020) Soil application of nano silica on maize yield and its insecticidal activity against some stored insects after the post-harvest. *Nanomaterials* 10:739

El-Samahy MFM, Khafagy IF, El-Ghobary AMA (2015) Efficiency of silica nanoparticles, two bioinsecticides, peppermint extract and insecticide in controlling cotton leafworm, *Spodoptera littoralis* Boisd. and their effects on some associated natural enemies in sugar beet fields. *J Plant Prot Path Mansoura Univ* 6:1221–1230

Fawe A, Abou-Zaid M, Menzies J, Bélanger R (1998) Silicon-mediated accumulation of flavonoid phytoalexins in cucumber. *Phytopathology* 88:396–401

Galal OA, Thabet AF, Tuda M, El-Samahy MFM (2020) RAPD analysis of genotoxic effects of nano-scale SiO_2 and TiO_2 on broad bean (*Vicia faba* L.). *J Fac Agr Kyushu Univ* 65:57–63

Goswami A, Roy I, Sengupta S, Debnath N (2010) Novel applications of solid and liquid formulations of nanoparticles against insect pests and pathogens. *Thin Solid Films* 519:1252–1257

Hu Y, Sopko R, Foos M, Kelley C, Flockhart I, Ammeux N, Wang X, Perkins L, Perrimon N, Mohr SE (2013) FlyPrimerBank: An online database for *Drosophila melanogaster* gene expression analysis and knockdown evaluation of RNAi reagents. G3: Genes Genomes Genetics 3:1607–1616

Ibrahim EG, Abd El-Latif ME, Barakat AST (2008) Efficacy of certain insecticides on leaf-miner *Liriomyza trifolii* (Burgess) (Diptera: Agromyzidae) infesting green bean (*Phaseolus vulgaris* L.) and its parasitoid *Diglyphus isaea* (Hymenoptera: Eulophidae) at Alexandria, Egypt. *Alexandria Science Exchange Journal* 29:64–70

Kaziem AE, Gao Y, He S, Li J (2017) Synthesis and insecticidal activity of enzyme-triggered functionalized hollow mesoporous silica for controlled release. *J Agric Food Chem* 65:7854–7864

Keeping MG, Meyer JH (2002) Calcium silicate enhances resistance of sugarcane to the African stalk borer *Eldana saccharina* Walker (Lepidoptera: Pyralidae). *Agric Forest Entomol* 4:265–274

Khan Z, Ansari MYK (2018) Impact of engineered Si nanoparticles on seed germination, vigour index and genotoxicity assessment via DNA damage of root tip cells in *Lens culinaris. J Plant Biochem Physiol* 6:5243–5246

Le VN, Rui Y, Gui X, Li X, Liu S, Han Y (2014) Uptake, transport, distribution and bio-effects of SiO_2 nanoparticles in Bt-transgenic cotton. *J Nanobiotechnol* 12:50

Massey FP, Hartley SE (2009) Physical defences wear you down: progressive and irreversible impacts of silica on insect herbivores. *J Anim Ecol* 78:281–291

Mousa KM, Elsharkawy MM, Khodeir IA, El-Dakhakhni TN, Youssef AE (2014) Growth perturbation, abnormalities and mortality of oriental armyworm *Mythimna separata* (Walker) (Lepidoptera: Noctuidae) caused by silica nanoparticles and *Bacillus thuringiensis* Toxin. *Egypt J Biol Pest Control* 24:283–287

Painter RH (1951) Insect resistance in crop plants. MacMillan, New York, 520 pp

Pavitra G, Sushila N, Sreenivas AG, Ashok J, Sharanagouda H (2018) Biosynthesis of green silica nanoparticles and its effect on cotton aphid, *Aphis gossypii* Glover and mealybug, *Phenacoccus solenopsis* Tinsley. *Int J Curr Microbiol App Sci* 7:1450–1460

Ranal MA, Santana DG (2006) How and why to measure the germination process? *Braz J Bot* 29:1–11

Ranal MA, Santana DG, Ferreira WR, Mendes-Rodrigues C (2009) Calculating germination measurements and organizing spreadsheets. *Braz J Bot* 32:849–855

Rani PU, Madhusudhanamurthy J, Sreedhar B (2014) Dynamic adsorption of a-pinene and linalool on silica nanoparticles for enhanced antifeedant activity against agricultural pests. *J Pest Sci* 87191–200

Rastogi A, Tripathi DK, Yadav S, Chauhan DK, Živčák M, Ghorbanpour M, El-Sheery NI, Brestic M (2019) Application of silicon nanoparticles in agriculture. *3 Biotech* 9:90

Reynolds OL, Keeping MG, Meyer JH (2009) Silicon-augmented resistance of plants to herbivorous insects: a review. *Ann Appl Biol* 155:171–186

Rouhani M, Samih MA, Kalantari S (2012) "Insecticidal effect of silica and silver nanoparticles on the cowpea seed beetle, *Callosobruchus maculatus* F. (Col.: Bruchidae). *J Entomol Res* 4:297–305

Sasamoto K (1955) Studies on the relation between insect pests and silica content in rice plant (III). On the relation between some physical properties of silicified rice plant and injuries by rice stem borer, rice plant skipper and rice stem maggot. *Oyo Kontyu* 11:66–69

Sharifi-Rad J, Sharifi-Rad M, Teixeira da Silva JA (2016) Morphological, physiological and biochemical responses of crops (*Zea mays* L., *Phaseolus vulgaris* L.), medicinal plants (*Hyssopus officinalis* L., *Nigella sativa* L.), and weeds (*Amaranthus retroflexus* L., *Taraxacum officinale* F. H. Wigg) exposed to SiO_2 nanoparticles. *J Agric Sci Technol* 18:1027–1040

Shoaib A, Elabasy A, Waqas M, Lin L, Cheng X, Zhang Q, Shi Z (2018) Entomotoxic effect of silicon dioxide nanoparticles on *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) under laboratory conditions. *Toxicol Environ Chem* 100:80–91, DOI: 10.1080/02772248.2017.1387786

Slomberg DL, Schoenfisch MH (2012) Silica nanoparticle phytotoxicity to *Arabidopsis thaliana*. *Environ Sci Technol* 46:10247–10254

Smith BC (1969) Effects of silica on the survival of *Coleomegilla maculata lengi* (Coleoptera: Coccinellidae) and *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae). *Can Entomol* 101:460–462

Suriyaprabha R, Karunakaran G, Yuvakkumar R, Rajendran V, Kannan N (2014) Foliar application of silica nanoparticles on the phytochemical responses of maize (*Zea mays* L.) and its toxicological behavior. *Synth React Inorg M* 44:1128–1131

Takahashi E (1995) Uptake mode and physiological functions of silica. Science of the Rice Plant, vol. 2: Physiology. Food and Agriculture Policy Resource Center, Tokyo, pp. 420–433

Thabet AF, Galal OA, El-Samahy MFM, Tuda M (2019) Higher toxicity of nano-scale TiO_2 and dose-dependent genotoxicity of nano-scale SiO_2 on the cytology and seedling development of broad bean *Vicia faba*. *SN Appl Sci* 1:956

Vani C, Brindhaa U (2013) Silica nanoparticles as nanocides against *Corcyra cephalonica* (S.), the stored grain pest. *Int J Phar Bio Sci* 4:1108–1118

Yang Z, Chen J, Dou R, Gao X, Mao C, Wang L (2015) Assessment of the phytotoxicity of metal oxide nanoparticles on two crop plants, maize (*Zea mays* L.) and rice (*Oryza sativa* L.). *Int J Environ Res Public Health* 12:15100–15109

Ye M, Song Y, Long J, Wang R, Baerson SR, Pan Z, Zhu-Salzman K, Xie J, Cai K, Luo S, et al. (2013) Priming of jasmonate-mediated antiherbivore defence responses in rice by silicon. Proc Natl Acad Sci USA 110:E3631–E3639

Yıldırım EM, Ünay A, Civelek HS (2010) The effect of *Liriomyza trifolii* (Burgess) (Diptera: Agromyzidae) on some leaf characteristics of bean (*Phaseolus vulgaris* L.). *J Food Agric Environ* 8:839–841

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Figure captions

Fig. 1 Larval survival rate (mean ± SE) of the leafminer *Liriomyza trifolii* at different concentrations of SiO_2NPs. An asterisk indicates a significant difference from the control

Fig. 2 Larval feeding velocity (mean ± SE) of the leafminer *Liriomyza trifolii* at different concentrations of SiO_2NPs. An asterisk indicates a significant difference from the control

Fig. 3 Pupal weight (mean ± SE) of the leafminer *Liriomyza trifolii* at different concentrations of SiO_2NPs. An asterisk indicates a significant difference from the control

Fig. 4 Gene expression levels of antioxidant enzymes for the leafminer *Liriomyza trifolii* at different concentrations of SiO_2NPs. Means are presented by horizontal bars

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1 Fig. 1

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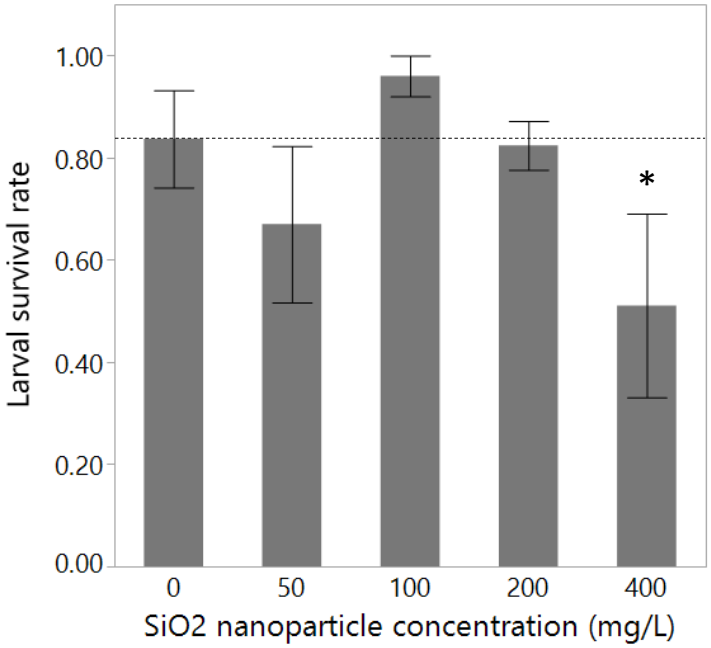

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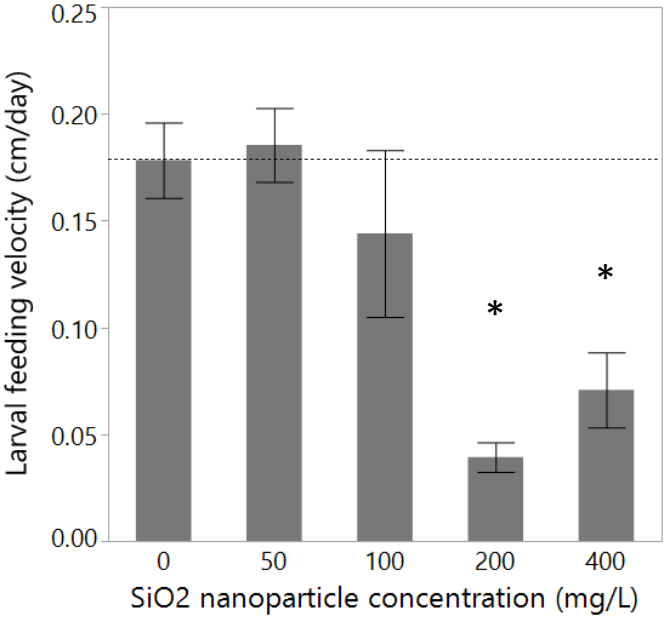
 doi:10.20944/preprints202010.0369.v1

Fig. 2



 doi:10.20944/preprints202010.0369.v1

12 Fig. 3

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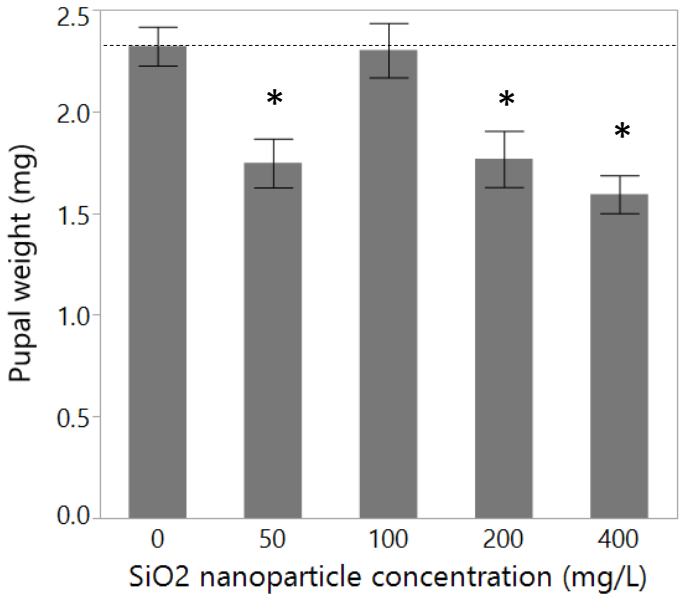

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 doi:10.20944/preprints202010.0369.v1

17 Fig. 4

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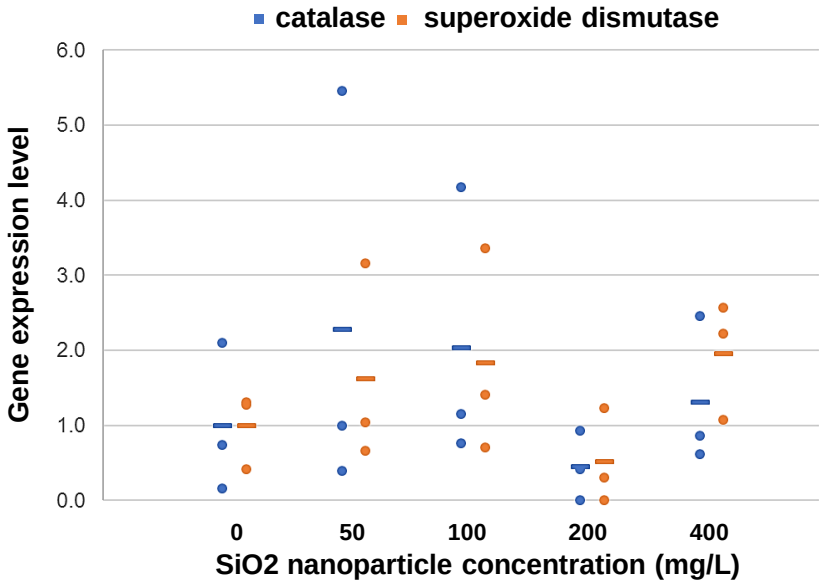

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