

SiO₂ Applications as an Alternative to Insect Control in Greenhouses [†]

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Abstract: Silicon dioxide (SiO₂) has been extensively studied as an alternative method to pest management chemical applications in agriculture. The present work aimed to evaluate the insecticidal efficacy of different SiO₂ nanoparticle formulations and their effectiveness when integrated into the textiles of an insect-proof net. For this reason, *Sitophilus oryzae* and *Tribolium confusum* were exposed to three inert dust formulations, namely Sylobloc[®] S200, S200-OH, and S200MEC to investigate their effect on the mortality of the aforementioned stored-product insects. The results of a series of bioassays showed that Sylobloc[®] S200 was the most effective nanoparticle among all the formulations tested. Thus, five samples of the same 50 mesh size insect-proof net were coated with the S200 nanoparticle followed either by different coating repetitions or by the addition of paraffin. *T. confusum* was indicated as the most tolerant species, as the recorded mortality rate was significantly low when exposed to all samples tested. However, the mortality rate of *S. oryzae* was strongly related to the coating repetition as well as the exposure intervals. The highest mortality (70%) was detected after 7 days of exposure to the net and it consisted of three coating repetitions while no paraffin was added to its surface. The results of the present study underline the insecticidal efficacy of SiO₂ treated nets against storage insects and their subsequent application in greenhouses for the control of more susceptible insects.

Keywords: silicon dioxide; insecticidal efficacy; stored-product insects; insect-proof net

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1. Introduction

The use of inert dust formulations such as silicon dioxide (SiO₂), as a non-toxic and environmentally friendly substance, has been well established in agriculture. The mode of action of SiO₂ lies in the adherence and friction of its nanoparticles on the insect's exoskeleton increasing water loss and mortality [1,2]. Nanoparticles present significant differences in comparison to their bulk formation [3]. *Sitophilus oryzae*, an important stored-product insect, was significantly affected by the use of nanoparticles in contrast with bulk silica which did not have any significant effect on the mortality of the species [4]. Unlike other nanoparticles, SiO₂ is a very promising substance in stored-product pest management as its insecticidal efficacy was indicated to be greater than that of ZnO and TiO₂ nanoparticles on *S. oryzae* and *Tribolium castaneum* due to its spherical morphology [5].

Moreover, silica aerogels and mixtures consisting of enhanced SiO₂ nanoparticles are expected to lead to higher and faster levels of pest mortality [6]. The efficacy of inert dust formulations in the control of insect population density depends to a great extent on the time intervals of exposure. Contrary to synthetic insecticides, inert dust's time of action is not direct which means that recovery is likely to happen when insects are exposed for short periods to the dust [6,7]. In a recent study, it was demonstrated that a significant mortality of *S. oryzae* and *T. confusum* is likely to occur at a slower rate when exposed to siliceous nanoparticles, as they are considerably hard-bodied and tolerant insects [8]. Following the previous study, the insecticidal efficacy of a 50 mesh size screen coated with SiO₂ nanoparticles was evaluated under laboratory conditions against *S. oryzae* and *T. confusum* [9]. The results indicated the efficiency of SiO₂ nanoparticles as an alternative biological insect control agent, as delayed mortality was promoted after 7 days of post-exposure to the treated screens. Considering the above, this study aimed to determine the mortality of two stored-product insects, *T. confusum* and *S. oryzae* when exposed to three silica nanoparticle types and five insect-proof screen formulations of different properties coated with SiO₂ nanoparticles for different time intervals in laboratory experiments.

2. Materials and Methods

Sitophilus oryzae (Coleoptera: Curculionidae) adults and *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) larvae were reared at the Laboratory of Entomology and Agricultural Zoology (LEAZ), Department of Agriculture, Crop Production and Rural Environment at the University of Thessaly in a growth chamber at 25 °C and 65% relative humidity, in continuous darkness, on whole wheat kernels and wheat flour, respectively.

Two bioassay series were performed under laboratory conditions to investigate the insecticidal efficacy of dust and insect-proof screen formulations against the aforementioned pests. During the 1st bioassay, three inert nanoparticle formulations, namely Sylobloc® S200, S200–OH, and S200MEC were tested as wheat protectants to monitor their effect on the mortality of *S. oryzae* adults and *T. confusum* larvae. A dosage of 1000 ppm of each silica dust was added in a glass jar containing soft wheat and the mixture was shaken manually for 5 min to achieve equal distribution of the silica nanoparticles in the entire wheat mass. 20 individuals of each species were carefully transferred in a plastic cylindrical vial (3 × 8 cm) in which 20 g of the aforementioned mixture were added, with separate vials for each species. The mortality rate of each species was measured 1 and 7 days of post-exposure to the dust formulations. Each bioassay was replicated three times per treatment.

In the 2nd bioassay, *S. oryzae* and *T. confusum* individuals were exposed to a 50 mesh insect-proof screen provided by Thrace Nonwovens & Geosynthetics S.A. without being a commercial product of the company, using High-Density Polyethylene (HDPE) monofilament woven yarns. The textiles of the screen were coated with the Sylobloc® S200 formulation. Table 1 shows the physical properties of each tested screen.

Table 1. Properties of the coated insect-proof screens.

SiO ₂ -Screens	Organic Primer	SiO ₂ Particle Diameter (µm)	Coating Repetition	SiO ₂ Mass on the Screen's Surface (g m ⁻²)
S200-1	-	2–4	1	1.2
S200-3	-	2–4	3	1.7
S200-0-P	Paraffin	2–4	0	15.4
S200-1-P	Paraffin	2–4	1	2.1
S200-2-P	Paraffin	2–4	2	2.5

The mass of deposited silica particles on the screen’s surface reached the maximum deposition rate after two coating repetitions and differs between samples due to the coating repetition as well as, to the addition of paraffin. In this series of bioassays, screens were cut and glued at the bottom of plastic Petri dishes (59.4 cm² on the surface). Ten insects of each species were transferred on each Petri dish and exposed to each treated screen for 72 h. Then, the insects were carefully transferred to a Petri dish consisting of a “clear” screen without any dust added. The response of all insects exposed to the treated screens was monitored at different time intervals, specifically after 1, 3, 5, and 7 days. For each of the five samples, the bioassay was replicated three times per exposure interval followed by three sub replicates. Cracked wheat (0.5 ± 0.1 g) and wheat flour (1.0 ± 0.1 g) were added in all Petri dishes to supply food to *S. oryzae* and *T. confusum* respectively.

Statistical Analysis

Multiple Comparison of means was performed by applying one-way ANOVA using the Tukey-Kramer HSD test at the 5% level ($p \leq 0.05$). In case data did not follow a normal distribution, a non-parametric Kruskal–Wallis test ($p \leq 0.05$) was performed. Significance of values have been adjusted by the Bonferroni correction for multiple tests. All comparisons were performed through SPSS (Statistical Package for the Social Sciences, IBM, Armonk, NY, USA).

3. Results

The recorded mortality of *T. confusum* larvae was significantly low. None of the tested stored-product insects were affected after 24 h of continuous exposure to the three silica nanoparticle formulations. However, as shown in Figure 1a, the highest mortality rate was recorded 7 days after the initial exposure of the larvae to the S200 formulation. In particular, Sylobloc® S200 was the most effective since the mortality rate was two- and four times higher in comparison to S200–OH and S200MEC, respectively. Notwithstanding, these results do not differ significantly among treatments ($p > 0.05$).

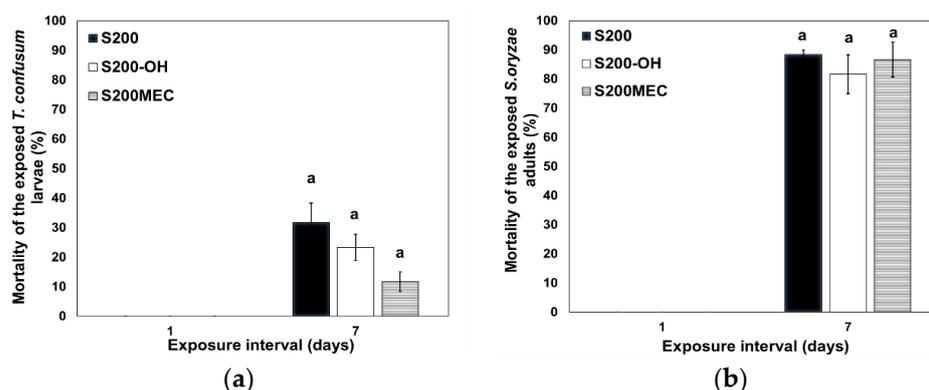


Figure 1. (a) Mortality rate of *T. confusum* larvae; (b) Mortality rate of *S. oryzae* adults after 1 and 7 days of post-exposure to the S200, S200–OH, S200MEC formulation.

On the other hand, the results depicted in Figure 1b indicated that the tested nanoparticle formulations were more lethal for *S. oryzae* adults as compared to *T. confusum* larvae. No mortality was detected one day after the exposure of *S. oryzae* to the siliceous nanoparticles. However, on the 7th day of exposure, all formulations led to an increase in mortality rate up to 86%. Taking into consideration the results of the three siliceous dust trials, S200 was selected as the most efficient formulation against both insects. Thus, the following experiments with the silica-treated screens were conducted on samples that were coated only with the Sylobloc® S200 nanoparticles.

As shown in Table 2, *T. confusum* larvae were indicated as the most tolerant species when exposed to all SiO₂-treated screens. In all bioassays, it was shown that none of the

five tested samples affected significantly the population density of *T. confusum* since the highest recorded mortality did not exceed 8.3% (S200-0-P) even 7 days after the exposure of the individuals to the treated screens.

In contrast to the results obtained from the *T. confusum* bioassay, the mortality rate of *S. oryzae* followed an upward trend for all tested screens as the exposure interval increased. In particular, the highest mortality was achieved 7 days of post-exposure to all treated screens except for the S200-1-P screen and ranged from 56.7 to 70.0% (Table 3).

Table 2. Mortality rate of *T. confusum* larvae after 72 h of continuous exposure to the treated screens and 1, 3, 5, and 7 days of post-exposure to treated SiO₂-treated screens.

SiO ₂ -Screens	72 h	1 d	3 d	5 d	7 d
S200-1	1.7 ± 1.7 Aab	1.7 ± 1.7 Ab	1.7 ± 1.7 Aab	1.7 ± 1.7 Aa	1.7 ± 1.7 Aa
S200-3	0.0 ± 0.0 Ab	5.0 ± 3.4 Aab	5.0 ± 3.4 Aab	5.0 ± 3.4 Aa	5.0 ± 3.4 Aa
S200-0-P	6.7 ± 2.1 Aa	8.3 ± 3.1 Aa	8.3 ± 3.1 Aa	8.3 ± 3.1 Aa	8.3 ± 3.1 Aa
S200-1-P	0.0 ± 0.0 Ab	0.0 ± 0.0 Ab	0.0 ± 0.0 Ab	1.7 ± 1.7 Aa	3.3 ± 3.3 Aa
S200-2-P	0.0 ± 0.0 Ab	0.0 ± 0.0 Ab	0.0 ± 0.0 Ab	1.7 ± 1.7 Aa	1.7 ± 1.7 Aa

Within each column, means followed by the same lowercase letters do not differ significantly ($p < 0.05$) across the insect-proof treatments for each of the tested exposure intervals. Within each row, means followed by the same uppercase letters do not differ significantly ($p < 0.05$) across the exposure intervals for each of the tested screens.

Table 3. Mortality rate of *S. oryzae* adults after 72 h of continuous exposure to the treated screens and 1, 3, 5, and 7 days of post-exposure to treated SiO₂-treated screens.

SiO ₂ -Screens	72 h	1 d	3 d	5 d	7 d
S200-1	5.0 ± 2.2 Cb	8.3 ± 4.0 Cb	25.0 ± 6.7 BCbc	41.7 ± 3.1 ABab	58.3 ± 4.8 Aab
S200-3	20 ± 5.2 Ca	30.0 ± 5.8 BCa	41.7 ± 6.0 BCab	48.3 ± 6.5 ABab	70.0 ± 2.6 Aa
S200-0-P	11.7 ± 3.1 Cab	23.3 ± 3.3 BCa	45.0 ± 4.3 ABa	56.7 ± 5.6 Aa	68.3 ± 4.8 Aa
S200-1-P	3.3 ± 2.1 Db	8.3 ± 3.1 CDb	23.3 ± 4.9 BCc	31.7 ± 4.0 ABb	46.7 ± 3.3 Ab
S200-2-P	8.3 ± 4.8 Cb	15.0 ± 5.6 BCab	40.0 ± 2.6 ABab	50.0 ± 5.2 Aa	56.7 ± 6.7 Aab

Within each column, means followed by the same lowercase letters do not differ significantly ($p < 0.05$) across the insect-proof treatments for each of the tested exposure intervals. Within each row, means followed by the same uppercase letters do not differ significantly ($p < 0.05$) across the exposure intervals for each of the tested screens.

4. Discussion

The results of the present study clearly show that the effect of inert dust formulations on insect mortality varies considerably among the selected species. Specifically, the mortality of *T. confusum* was relatively low as only 32% of the larvae were affected after 7 days of continuous exposure to the S200 formulation. On the other hand, *S. oryzae* adults were more susceptible to all dust formulations, particularly to S200, as the mortality rate increased at 88% after 7 days of post-exposure to the dust. In a recent study conducted to determine the efficacy of siliceous formulations against storage insects, it was indicated that after 6 h of exposure to different silica formulations, 99% of the *S. oryzae* adults were under knockdown effect (KD₉₉), i.e., they had limited movement which could not affect the commodities. Unlike *S. oryzae*, *T. confusum* larvae were more tolerant to such formulations, requiring at least 8.5 h of continuous exposure to achieve KD₉₉. Moreover, in the same study, long term exposure bioassays were assessed, suggesting that the delayed mortality of the insects was increased to 100% after 7 days of post-exposure for only 15 min to the SiO₂ nanoparticles [9]. However, the desiccation is more likely to occur for soft-bodied insects such as aphids, whilst in the case of stored-product insects, water loss becomes more difficult due to their hard exoskeleton. Thus, SiO₂ nanoparticles could be introduced in greenhouses to enhance pest management.

Moreover, a more recent survey regarding the pest management under SiO₂ treated screens against *S. oryzae* and *T. confusum* showed that the mortality of the individuals reached 100% and 34% after 10 days of post-exposure for only 15 min to the treated screen, underlying the strong tolerance of the latter species to the treated screens. In the same study, it was argued that the mortality of *S. oryzae* was independent of the addition of paraffin on the screen's surface. Our results are in agreement with those of the aforementioned study as paraffin did not play a decisive role in insect mortality. The delayed mortality recorded for both of the tested stored-product insects can also be associated with the slow-acting mode of inert dusts as the mortality is possible to occur over a period of weeks after continuous exposure to the screens [9].

The tested stored-product insects are considered more tolerant species compare to insects that usually penetrate the interior of a greenhouse structure. SiO₂ offers an innovative technological solution for sustainable agriculture, preserving pest management, whilst minimizing the environmental footprint through its non-toxic and pesticide-free basis. In this way, insect-proof screens coated with SiO₂ nanoparticles could be integrated on the greenhouses' vent openings by serving a dual role as a barrier and as a natural insecticide contributing to pest control and adequate ventilation.

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