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Evaluation of Inert Dusts on Surface Applications and Factors That Maximize Their Insecticidal Efficacy

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Simple Summary: Inert dusts including diatomaceous earths and zeolites have been proved very effective as grain protectants against stored product insects. However, little progress has been made towards the evaluation of their insecticidal activity when applied directly to different types of surfaces. In this study, we evaluated two diatomaceous earth formulations and one zeolite deposit from Greece when applied to concrete and steel against three major stored product insect pests. Based on the results, both dusts achieved complete control of all insect species within a week. The type of surface was found to not be a factor of significance for the effectiveness of the dusts. These data further encourage the exploitation of these natural insecticides for structural treatments in storage facilities, either as alternatives to residual insecticides and fumigants that have been used as essential components for stored-grain protection systems, or for integrated pest management (IPM) approaches.

Abstract: We evaluated formulations of diatomaceous earth and zeolite originated from natural deposits from Greece as insecticides in concrete and steel surfaces for the control of three major beetle species of stored products: *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae) and *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae). The formulations were tested as dusts at 0.5 or 1 g/m^2. Our results indicated that, in most of the cases tested, the inert materials caused 100% adult mortality for all three species, even at the lowest dose, after 7 days of exposure. At the same time, there were no considerable differences in the insecticidal effect of the formulations between concrete and steel surfaces. Among the species tested, *R. dominica* was the most susceptible, followed by *S. oryzae* and *T. confusum*. Our results indicate that natural resource-based inert silicaceous deposits could be used with success in stored product protection against insects at dose rates that are comparable with other commercially available inert material-based formulations.

Keywords: diatomaceous earth; zeolite; natural insecticides; stored product insects; structural treatments

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1. Introduction

Inert dusts, such as diatomaceous earths (DEs) and zeolites are promising alternatives to the use of traditional chemical methods in stored product protection [1]. A high number of studies have documented their efficacy against a wide range of insect species [2–6], while some studies have shown that these dusts are effective against fungi and bacteria that may co-infest the commodity along with insects [7]. In a recent review, Zeni et al. [8] illustrated the insecticidal potentials of DEs and related materials against a variety of insect taxa (Blattodea, Coleoptera, Diptera, Hemiptera, Hymenoptera and Lepidoptera), including household or agricultural pest species but also non-target insects. Similarly, Eroglou et al. [9] reviewed the potentials of utilizing zeolites in food and agriculture including their important role in stored-pest management as inert dust applications.

One of the incontestable advantages of inert materials and their use in stored product protection is the fact that, in most cases, they are of natural origin [10,11]. The utilization of natural resource-based agents has been long regarded as an important parameter in modern pest control, and natural insecticides are now prioritized in terms of Integrated Pest Management (IPM) protocols [1]. At the same time, the exploitation of locally produced mineral materials can also be economically beneficial for the local economy of a given country [12–14]. Athanassiou et al. [12], in a thorough screening of DEs that were sampled from different countries of southeastern Europe, found certain deposits that had a high insecticidal efficacy, and could be further used in stored product protection. There are some studies indicating that this geographical area is rich in zeolites with good insecticidal value as well, with some of which to be evaluated successfully in stored product protection [15–18]. Korunić [16] introduced a series of methods that can be used for the rapid evaluation of the insecticidal value of DEs, without the need to conduct bioassays, which could be adopted in the case of handling a large number of samples. This series of tests is mostly based on the particle size, but also to other parameters, such as pH and flowability [16]. Vayias et al. [19] reported that particle size is a crucial parameter and that the smaller the DE particles the higher the insecticidal efficacy against certain stored product beetle species. In a similar way, Rumbos et al. [6] found that particle size of zeolites was important in terms of efficacy against insects, while particles with different sizes had a different degree of attachment in the insects' cuticle. Most of the above studies have been carried out with locally-produced deposits of amorphous silicacesous materials, such as DEs and zeolites, which have been proved equally effective, if not more effective, than silicates, which are artificially made [9,11–14,19].

A plethora of data are available in the literature regarding the efficacy of inert materials for the control of major stored product insects. However, most of the published studies are focused on the utilization of inert materials as grain protectants, i.e., admixture with the bulked grains, despite the fact that there are numerous obstacles and disadvantages over this practice [15,20,21]. For instance, Athanassiou et al. [20] found that the commercially available DE formulation SilicoSec® (Biofa GmbH, Munsingen, Germany), was effective against the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), but this efficacy varied according to the commodity on which the DE was applied. In the same context, Eroglou et al. [21] reported that different formulations of zeolites could be effective for the control of *S. oryzae* and the saw-toothed grain beetle, *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) on grains, but their efficacy varied according to certain abiotic conditions, such as temperature and relative humidity. Additionally, the grain industry is reluctant to use inert dusts as grain protectants over the fact that the recommended dosages of DEs and zeolites can sufficiently reduce the grain bulk density, an extensively used grading factor that affects the grain prices [15].

In contrast, there are limited published data for the use of inert materials as surface treatments, i.e., in walls, floor etc., despite the fact that most of the insecticidal formulations that are now registered in stored product protection are for this use and not as admixture with grains. More than two decades ago, Dowdy and Fields [22] combined DEs with heat treatment in a flour mill and found that the occurrence of DEs could drastically accelerate insect mortality of the confused flour beetle, *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) that is caused by elevated temperatures. Similar results have been reported by Dowdy [23] in the case of this combination for the red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). Bohinc et al. [24] found that zeolites can be effective as a surface treatment against the maize weevil, *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). As the cases of resistance to conventional insecticides not only by the aforementioned species but also by other major stored product pests such as the lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae), are constantly increasing [25–27], the utilization of inert dust applications on stored product protection has gained distinct attention for their vital importance in resistant pest management strategies. Thus, the evaluation of these materials as surface or crack and crevice treatments in storage and processing facilities becomes even more important, in order to illustrate if eventually DEs and zeolites can play a viable role as insecticides that can be used for this purpose.

Recently, Baliota and Athanassiou [28] found that certain DE deposits from Greece were very effective as grain protectants against several beetle pests at concentrations that were comparable with those of commercial DE formulations. In addition to these DEs, the same surveillance indicated that there is a zeolite formulation that could be effective for this purpose. Based on the above, and taking into account the scarcity of data for the application of DEs and zeolites in surfaces, the aim of the present study was to evaluate these specific Greek deposits for this purpose under a range of factors, such as the target species, the concentration, the exposure interval and the type of surface.

2. Materials and Methods

2.1. Inert Dusts Used

The DE deposit was obtained from a single mine located in the area of Elassona (39.894524, 22.185065), in the Prefecture of Thessaly, Greece. The zeolite was obtained from a single mine located in the area of Orestiada (41.506950, 26.531530), in the Prefecture of Thrace, Greece. Regarding the DE formulations used in these bioassays, two enriched in diatoms and with different particle size DE powders (namely DE5 and DE6) were created and evaluated. More specifically, DE5 had 70% semi fractured diatoms content and 80% of the particles were smaller than 45 μm, while DE6 contained 68% of totally fractured diatoms with 99% of the particles being smaller than 45 μm. These powders were created by separating the diatoms from other elements from a given DE formulation with high clay content, containing only 40–60% discoid and cylindrical structure diatoms, using air separation methods. Supplementary data about the geographical origin of this DE deposit, the processing to create these new, enriched in diatoms, DE formulations and their insecticidal activity against several stored product insects are available in the work of Baliota and Athanassiou [28]. Data about the insecticidal activity of DE deposits of the same wider area are available in the literature with various results [12,19,29]. The geochemical analysis measurements (X-ray Powder Diffraction, XRPD) of zeolite sediments were carried out by The Mineral Lab Inc., Golden, CO, USA. The zeolite sentiments were found to contain 85% clinoptilolite, 9% plagioclase feldspar, and <3% quartz and mica/illite, while <5% of the minerals could not be identified.

2.2. Insect Species Tested

Mixed-sex adults (7–21 days old) of *S. oryzae*, *T. confusum* and *R. dominica* were used in the bioassays. All insects were taken from standard laboratory cultures maintained at the Laboratory of Entomology and Agricultural Zoology, Department of Agriculture, Crop Production and Rural Environment, University of Thessaly, N. Ionia, Magnesia, Greece, for more than 15 years, at 25 ± 1 °C and 56 ± 5% relative humidity (RH), in continuous darkness. *Tribolium confusum* was reared in whole wheat flour, and *S. oryzae* and *R. dominica* on whole soft wheat kernels.

2.3. Bioassays

The trials were carried out in standard plastic 90-mm-diameter Petri dishes (90 mm diameter, 15mm high, 63.6 cm^2 bottom surface, Sarstedt Ag & Co. Kg, Nümbrecht, Germany) with bottoms covered with cement (concrete) or steel. For the preparation of the concrete surfaces, 400 g of concrete (Grey cement 42.5, Isomat S.A., Thessaloniki, Greece) was mixed thoroughly with 100 mL of tap water. Approximately 15 mL of this mixture was poured into each dish in order to cover the dish bottom with an even concrete surface. Afterwards, the dishes were left to dry for 24 h at room temperature. Steel surfaces were cut in a local factory and adjusted in the bottom of each dish with silicone glue. A polytetrafluoroethylene preparation (60 wt% dispersion in water, Sigma-Aldrich Chemie GmbH, Steinheim, Germany) was applied to the internal dish walls to avoid insects' escape. Quantities of DE5, DE6 and zeolite were added uniformly in each dish at two different application rates: 0.5 and 1 g/m^2, equal to 31.8 mg and 63.6 mg per dish, respectively. The whole process was repeated nine times, creating new dishes for each combination of species–surface-insecticide–dose, whereas nine dishes were left untreated and served as controls (thus, we used one set of control for all insecticides). Then, 20 adults of *S. oryzae*, *R. dominica* or *T. confusum* were added into the dish, without any trace of food and with separate dishes for each dose and species. After that, all the dishes were placed at 25 °C and 55% RH. Mortality of the exposed adults was measured after 1, 3, 7 and 14 days (d) of exposure to the insecticides.

2.4. Statistical Analysis

Before the analysis, all data were assessed for assumptions of normality using the Shapiro–Wilk Test and for homoscedasticity using Levene's Test. The assumptions were met for non-parametric analysis for all data. Thus, the Kruskal–Wallis Test was performed at $p < 0.05$ to compare the mortality between the three insecticides and the control (0 g/m^2) within each insect species, dose, surface and exposure interval. Nonparametric comparisons for all pairs using Dunn method for joint ranking with Bonferroni adjustments were followed at $p < 0.05$, for post-hoc testing the differences between the insecticides and the control within each insect species, dose, surface and exposure interval. All analyses were performed using the JMP®Software (version 17.0) (SAS Institute Inc., Cary, IL, USA) [30].

3. Results

3.1. *Sitophilus oryzae*

Mortality of *S. oryzae* adults on steel surfaces differed significantly among the treatments after 1 d ($x^2 = 27.7$, $p < 0.01$, df = 3), after 3 d ($x^2 = 29.5$, $p < 0.01$, df = 3) and after 7 d ($x^2 = 9.5$, $p = 0.02$, df = 3) of exposure to the lower dose (0.5 g/m^2), and after 1 d ($x^2 = 20.6$, $p < 0.01$, df = 3), after 3 d ($x^2 = 29.5$, $p < 0.01$, df = 3) and after 7 d ($x^2 = 9.5$, $p = 0.02$, df = 3) of exposure to the higher dose (1 g/m^2) (Figure 1). Zeolite was observed to be the most effective of the tested DEs after the first d of exposure to the lower dose and especially in contrast with DE5, which was comparable with the control treatments. By increasing the dose, the effectiveness of DE5 increased and was comparable to that of zeolite at the same interval (1 d). After 3 d, all insecticides caused similar mortality rates in *S. oryzae* adults, which were significantly higher than that of the controls, regardless of the dose tested. All adults were dead after 7 d of exposure to insecticides but not to the control treatments (Figure 1).

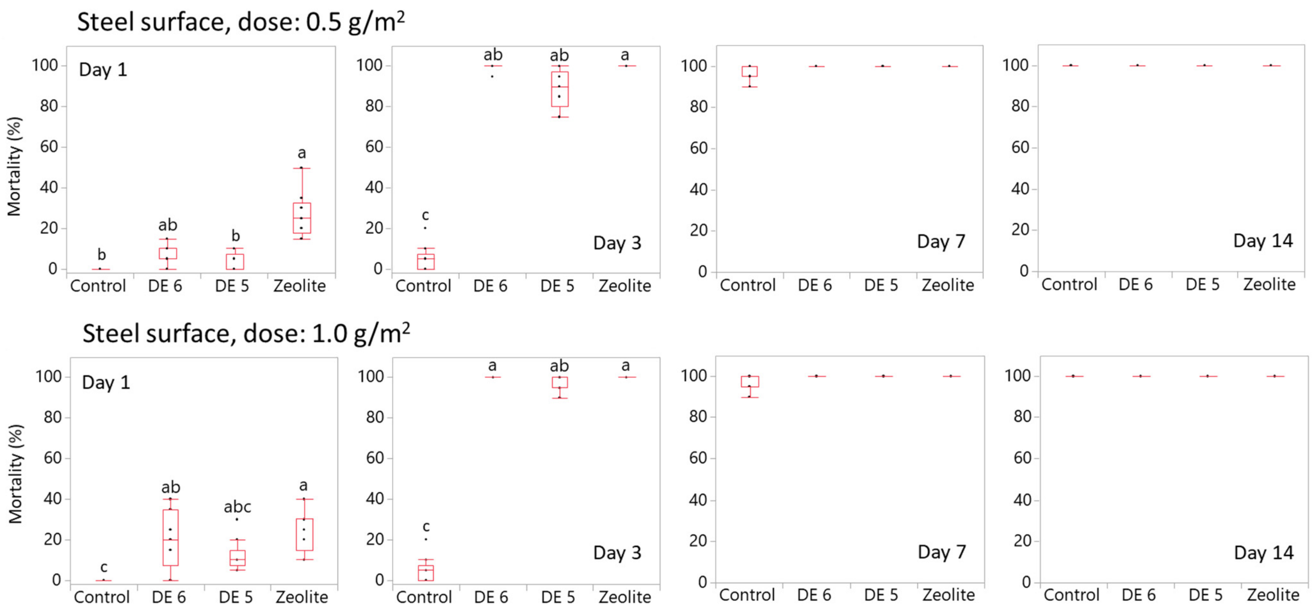

Figure 1. Mortality (%) of *S. oryzae* adults after exposure for 1, 3, 7 and 14 d at three different doses [0 (control), 0.5 and 1 g/m^2] of DEs and zeolite applied on steel surfaces (boxplots followed by the same letter are not significantly different, according to the Dunn test at $p < 0.05$. Where no letters exist, no significant differences were noted).

In concrete surfaces, mortality of *S. oryzae* adults differed significantly among the three insecticides and the control after 1 d (x^2 = 16.1, $p < 0.01$, df = 3), after 3 d (x^2 = 27.2, $p < 0.01$, df = 3), after 7 d (x^2 = 13.1, $p < 0.01$, df = 3) of exposure to the lower dose, and after 1 d (x^2 = 13.8, $p < 0.01$, df = 3), after 3 d (x^2 = 29.6, $p < 0.01$, df = 3) and after 7 d (x^2 = 13.1, $p < 0.01$, df = 3) of exposure to the higher dose (Figure 2). Although the overall mortality rates of all insecticides were rather low in concrete after 1 d of exposure to both doses, zeolite and DE6 observed to be significantly more effective than DE5, with the latter one to have no effect on the insects. The high survival that was observed in the control treatments at the same interval (3 d) indicates that the absence of food had no effect on insects until that point (Figure 2). All adults were dead after 7 d of exposure to insecticides but also in the control.

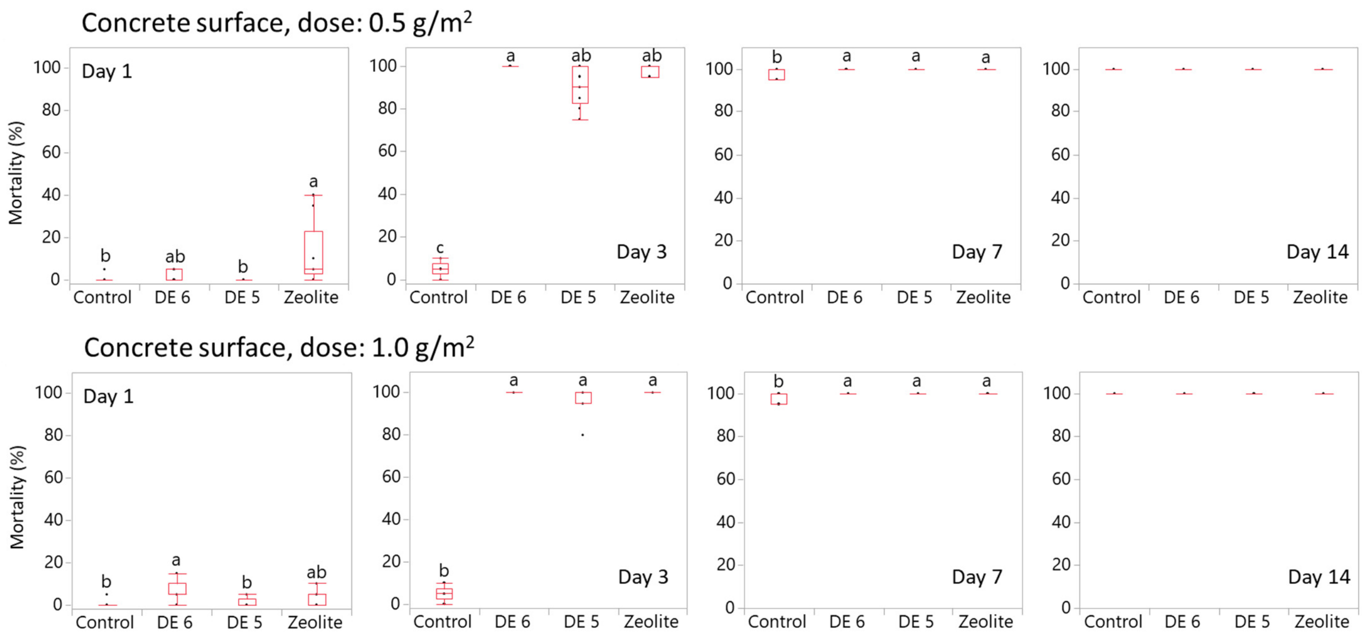

Figure 2. Mortality (%) of *S. oryzae* adults after exposure for 1, 3, 7 and 14 d at three different doses [0 (control), 0.5 and 1 g/m^2] of DEs and zeolite applied on concrete surfaces (boxplots followed by the same letter are not significantly different, according to the Dunn test at $p < 0.05$, where no letters exist, no significant differences were noted).

3.2. Rhyzopertha dominica

Mortality of *R. dominica* adults observed to be affected by the insecticides applied in steel surfaces after 1 d (x^2 = 31.1, $p < 0.01$, df = 3), after 3 d (x^2 = 32.4, $p < 0.01$, df = 3) and after 7 d (x^2 = 34.1, $p < 0.01$, df = 3) to the lower dose, and after 1 d (x^2 = 29.0, $p < 0.01$, df = 3), after 3 d (x^2 = 34.3, $p < 0.01$, df = 3) and after 7 d (x^2 = 34.1, $p < 0.01$, df = 3) in the higher dose (Figure 3). Zeolite was the most effective against *R. dominica,* causing over 60% mean mortality after only 1 d of exposure to both doses tested, followed by DE6 and DE5 respectively. Adult mortality was 100% or close after 3 d of exposure, regardless of the insecticide and dose. Still, significantly lower was the control's mortality compared to the treatments with the insecticides, until the 14th d of exposure (Figure 3).

In concrete surfaces, differences were observed among the treatments after 1 d (x^2 = 30.1, $p < 0.01$, df = 3), 3 d (x^2 = 34.3, $p < 0.01$, df = 3) or 7 d (x^2 = 34.0, $p < 0.01$, df = 3) of exposure to the lower dose, and after 1 d (x^2 = 30.9, $p < 0.01$, df = 3), 3 d (x^2 = 34.3, $p < 0.01$, df = 3) and 7 d (x^2 = 34.0, $p < 0.01$, df = 3) to the higher dose (Figure 4). Zeolite and DE6 were equally more effective than DE5 or control in both doses, with over 60% mortality after 1 d of exposure. Nevertheless, complete control was achieved after 3 d of exposure to all insecticides and doses tested, while adults of the control treatments were dead after 14 d (Figure 4).

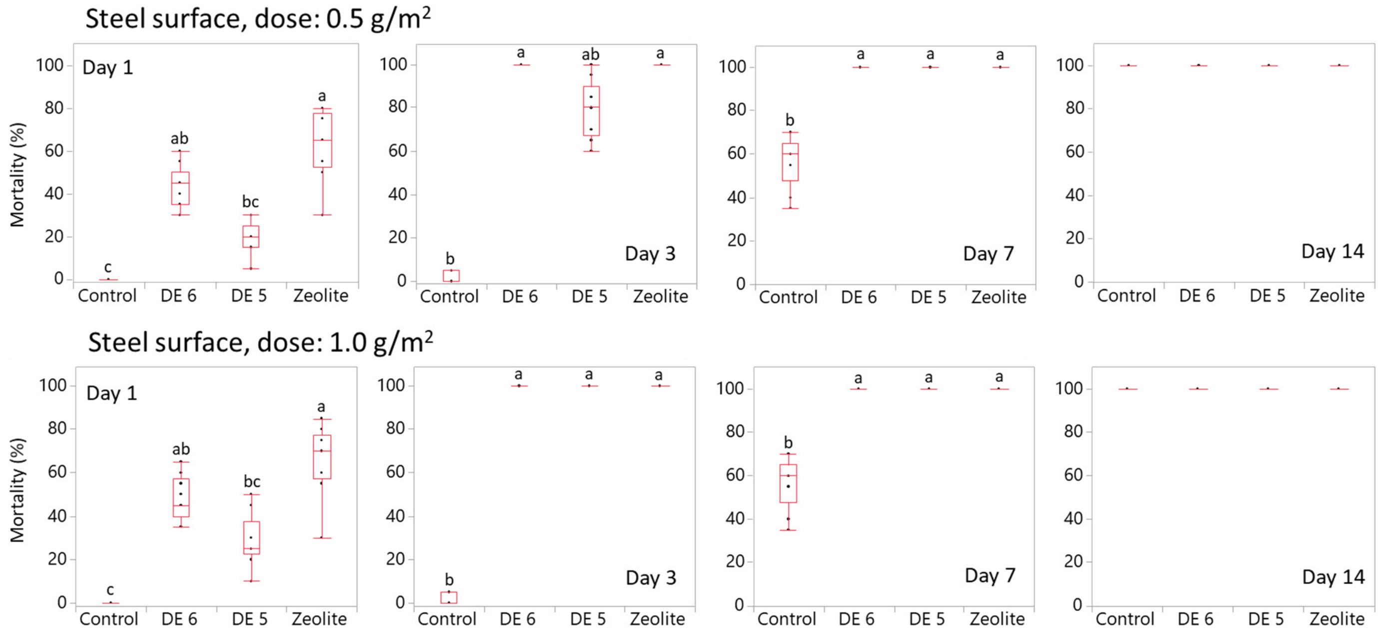

Figure 3. Mortality (%) of *R. dominica* adults after exposure for 1, 3, 7 and 14 d at three different doses [0 (control), 0.5 and 1 g/m^2] of DEs and zeolite applied on steel surfaces (boxplots followed by the same letter are not significantly different, according to the Dunn test at $p < 0.05$, where no letters exist, no significant differences were noted).

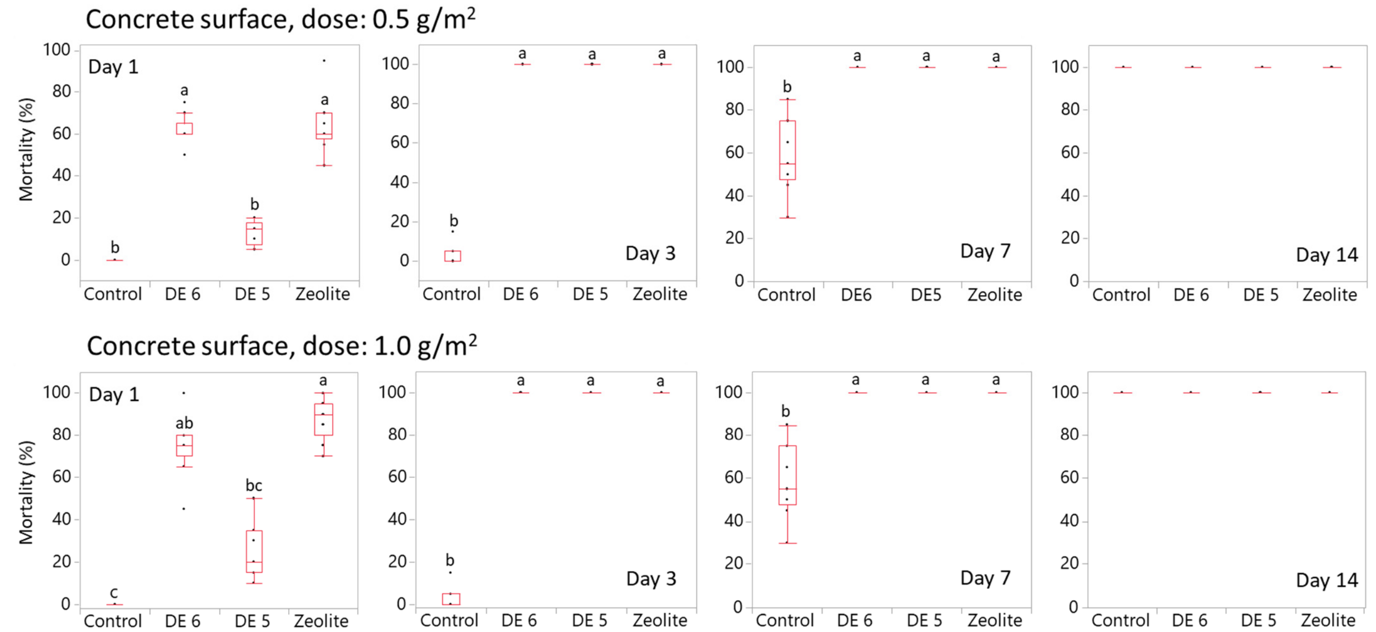

Figure 4. Mortality (%) of *R. dominica* adults after exposure for 1, 3, 7 and 14 d at three different doses [0 (control), 0.5 and 1 g/m^2] of DEs and zeolite applied on concrete surfaces (boxplots followed by the same letter are not significantly different, according to the Dunn test at $p < 0.05$, where no letters exist, no significant differences were noted).

3.3. *Tribolium confusum*

According to the data of the control treatments, *T. confusum* was found to be the most tolerant to starvation in comparison with the other species tested here, since no mortality was recorded even after 14 d without food (Figures 5 and 6). Significant differences were observed among the insecticides applied in steel surfaces after 1 d ($x^2 = 16.1$, $p < 0.01$, df = 3), 3 d ($x^2 = 24.8$, $p < 0.01$, df = 3), 7 d and 14 d ($x^2 = 35.0$, $p < 0.01$, df = 3) in the lower dose, and after 1d (x^2 = 20.3, $p < 0.01$, df = 3), 3 d (x^2 = 27.9, $p < 0.01$, df = 3), 7 d and 14 d (x^2 = 35.0, $p < 0.01$, df = 3) in the higher dose (Figure 5). Low mortality rates were observed among the treatments at the first d of exposure to both doses of the insecticides, although some differences were observed among the insecticides. After 3 d, adults observed to be more susceptible to DEs than zeolite, especially in the higher dose, while complete control was achieved after 7 d of exposure to all insecticides and doses. In some cases, the mean mortality in the lower dose was noted to be slightly higher than that of the higher dose within each time interval and insecticide, but these differences were interpreted as an indication that the increase of dose had no effect on the mortality of the species.

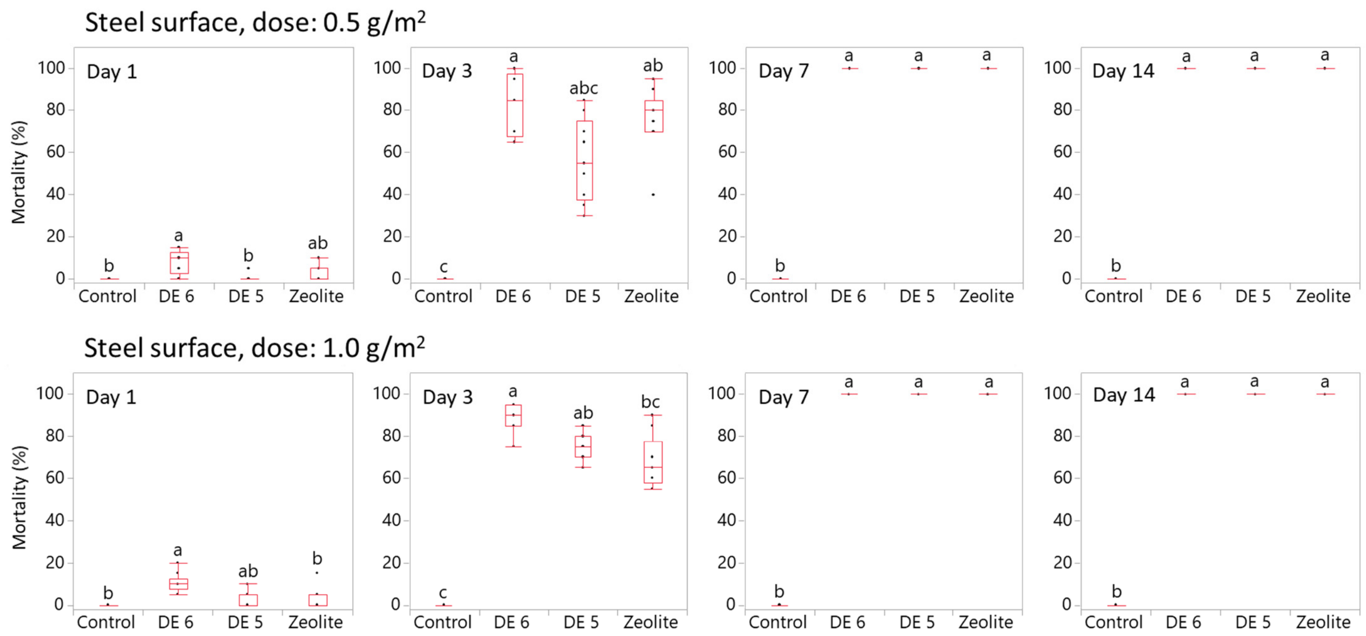

Figure 5. Mortality (%) of *T. confusum* adults after exposure for 1, 3, 7 and 14 d at three different doses [0 (control), 0.5 and 1 g/m^2] of DEs and zeolite applied on steel surfaces (boxplots followed by the same letter are not significantly different, according to the Dunn test at $p < 0.05$, where no letters exist, no significant differences were noted).

In concrete, all insecticides were found to be comparable with control, after the first d of exposure to the lower dose (x^2 = 4.8, p = 0.18, df = 3), but not after 3 d (x^2 = 31.7, $p < 0.01$, df = 3), 7 d (x^2 = 30.0, $p < 0.01$, df = 3) and 14 d (x^2 = 35.0, $p < 0.01$, df = 3) of exposure (Figure 6). Differences among the insecticides were observed with the increase of dose after 1 d (x^2 = 10.9, p = 0.01, df = 3), 3 d (x^2 = 31.3, $p < 0.01$, df = 3), 7 d (x^2 = 34.7, $p < 0.01$, df = 3) and 14 d (x^2 = 35.0, $p < 0.01$, df = 3) of exposure. All insecticides and doses caused very low adult mortality of *T. confusum* after the first d of exposure. Until the complete control of the species, DE6 and zeolite had similar mortality rates and were very effective against *T. confusum*, after only 3 d of exposure to both doses. On the other hand, DE5 was observed to have a slower effect in terms of adult mortality than the other two insecticides, especially at the 3 d-interval, even with the increase of dose.

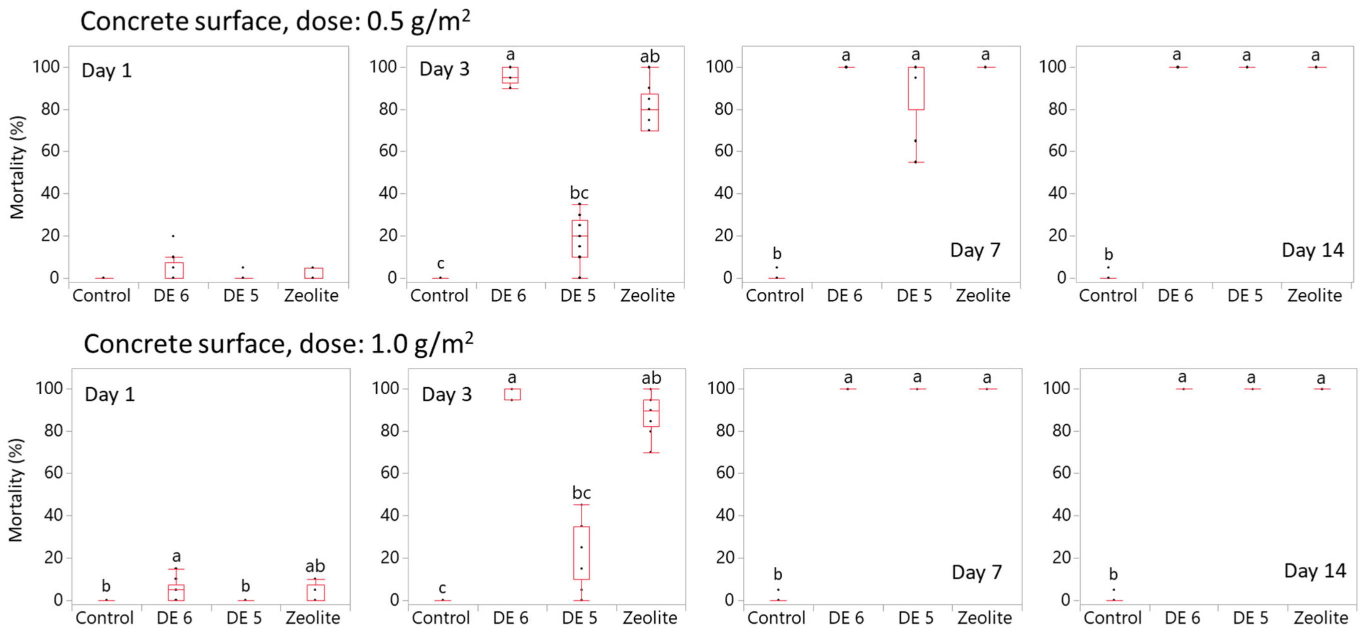

Figure 6. Mortality (%) of *T. confusum* adults after exposure for 1, 3, 7 and 14 d at three different doses [0 (control), 0.5 and 1 g/m^2] of DEs and zeolite applied on concrete surfaces (boxplots followed by the same letter are not significantly different, according to the Dunn test at $p < 0.05$. Where no letters exist, no significant differences were noted).

4. Discussion

Our data indicate that there are specific formulations that were effective at a relatively low concentration, when applied on surfaces. For the DE formulations tested, Baliota and Athanassiou [28] described specific modifications that could be performed in order to improve their insecticidal value; these modifications mainly included sieving, drying and smashing of the DE particles. In principle, smaller particles are more effective than large ones, but can negatively affect the bulk density of the grains on which they are applied [15,16]. Moreover, the shape of the diatoms can be also related with their insecticidal effect, and "flat shaped" with greater number of pores may have a higher wax adsorption from the insects' cuticle [14,16]. Still, the way that the insects are in contact with the inert dust particles that are applied in surfaces might be different than that of the particles that are applied on grains, where feeding from the treated substrate also takes place.

The comparison of the two surfaces, taking the entire mortality data into consideration, suggests that the materials used are equally effective on both concrete and steel, after a week of exposure. It is generally considered that most contact insecticides are more effective on steel than on concrete, since the latter is porous and alkaline, which are factors that contribute to the dissipation of the insecticide [31–34]. For instance, Collins et al. [35] tested four organophosphorous insecticides on concrete and galvanized steel and found that the efficacy of these active ingredients against three psocids species was higher on steel than on concrete, due to the fact that they are quickly hydrolyzed in the alkaline concrete surface. However, there are cases where the tested compounds are equally effective on different surfaces [36–38]. In fact, Toews et al. [39] reported that spinosad was more effective on concrete than galvanized steel for the control of *T. castaneum* and *T. confusum*. The fact that the inert materials evaluated here had no difference in their efficacy between the two surfaces tested can be considered as a positive characteristic, given that these dusts can be used in different application scenarios, without loss in efficacy. Moreover, for some of the species tested, the "speed of kill" of adults was comparable with other insecticides that are used in surfaces [32,37], while mortality was found to be more of a species-mediated parameter.

From the species tested here, we can conclude that both *S. oryzae* and *R. dominica* were far more susceptible than *T. confusum* to all formulations. From the most to the least susceptible, these three species can be classified as *R. dominica* > *S. oryzae* > *T. confusum*. Earlier studies have confirmed that the species of the genus *Tribolium* are probably the most tolerant stored product beetle species to inert dusts [6,8,40]. Interestingly, *R. dominica* was the most susceptible among the three species tested, despite the fact that this species is less agile than the other two [41,42]. Both *S. oryzae* and *R. dominica* are primary colonizers and can easily infest sound grain kernels, so they are most likely to be found in bulked grains, and not in surfaces, as compared with *T. confusum*, which is mostly found in processed commodities and processing facilities, such as flour mills. The application of these dusts as "barriers" to limit the spread of primary colonizers to infest additional areas with bulked grains has been found to be a viable solution, given that the presence of inert dusts has a repulsive action in some stored product insect species [1,8,43].

The concentrations tested here are generally comparable with those that have been used in similar studies testing inert materials in surfaces against insects [44–48]. We found that the formulation DE5 was usually less effective than the other two formulations for the majority of the combinations tested. However, even in the case of DE5, adult mortality reached 100% at a relatively short period of time, suggesting that the exposed individuals were largely affected even from the first days of their contact with the DE particles. Baliota and Athanassiou [28] reported that both DE5 and DE6 were effective for the control of major stored product beetle species as grain protectants, with DE6 to be slightly superior to DE5, which stands in agreement with the current observations. Nonetheless, all three formulations were able to kill all adults for all three species tested after 7 d of exposure, while, at least for *S. oryzae* and *R. dominica*, adult mortality was 100% even after 3 d of exposure. Arthur [49] used 0.5 g/m^2 of the commercial DE formulation Protect-It, and found that after 7 d, adults of *T. confusum* were still alive and not heavily affected. Extremely low "speed of kill" after exposure to DEs is not desirable, as insects can escape from the treated area and colonize uninfested grain [1]. Nevertheless, in the case of *T. confusum*, larval mortality after exposure to inert dusts may be high [50], which can gradually eliminate the population through increased immature mortality.

The current work sheds light on the application of DEs and zeolites on surfaces. It provides evidence that, in contrast with other insecticides, the type of surface may not be an important parameter here. Our study contributes further to the exploitation of the value of certain natural deposits of siliceous materials as a means to control stored product insect pests. Considering the need to adopt non-chemical solutions in stored product protection, and taking into account that inert materials are compatible with sustainable and organic food production, additional work is needed to quantify the effectiveness of these formulations when applied in empty storage facilities under a variety of application scenarios.

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References

1. Subramanyam, B.; Roesli, R. Inert dusts. In *Alternatives to Pesticides in Stored-Product IPM.*; Subramanyam, B., Hagstrum, D.W., Eds.; Kluwer Academic Publishers: Dordrecht, The Netherlands, 2000; pp. 321–380.
2. Athanassiou, C.G.; Vayias, B.J.; Dimizas, C.B.; Kavallieratos, N.G.; Papagregoriou, A.S.; Buchelos, C.T. Insecticidal efficacy of diatomaceous earth against *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) and *Tribolium confusum* du Val (Coleoptera: Tenebrionidae) on stored wheat: Influence of dose rate, temperature and exposure interval. *J. Stored Prod. Res.* **2005**, *41*, 47–55. [CrossRef]
3. Baldassari, N.; Prioli, C.; Martini, A.; Trotta, V.; Barionio, P. Insecticidal efficacy of a diatomaceous earth formulation against a mixed age population of adults of *Rhyzopertha dominica* and *Tribolium castaneum* as function of different temperature and exposure time. *Bul. Insectology* **2008**, *61*, 355–360.
4. Arthur, F.H. Immediate and delayed mortality of *Oryzaephilus surinamensis* (L.) exposed on wheat treated with diatomaceous earth: Effects of temperature, relative humidity, and exposure interval. *J. Stor. Prod. Res.* **2001**, *37*, 13–21. [CrossRef] [PubMed]
5. Mewis, I.; Ulrichs, C. Action of amorphous diatomaceous earth against different stages of the stored product pests *Tribolium confusum* (Coleoptera: Tenebrionidae), *Tenebrio molitor* (Coleoptera: Tenebrionidae), *Sitophilus granarius* (Coleoptera: Curculionidae) and *Plodia interpunctella* (Lepidoptera: Pyralidae). *J. Stored Prod. Res.* **2001**, *37*, 153–164.
6. Rumbos, C.I.; Sakka, M.; Berillis, P.; Athanassiou, C.G. Insecticidal potential of zeolite formulations against three major stored-grain insects: Particle size effect, adherence ratio and effect of test weight on grains. *J. Stored Prod. Res.* **2016**, *68*, 93–101. [CrossRef]
7. Campolo, O.; Romeo, F.V.; Malacrinò, A.; Laudani, F.; Carpinteri, G.; Fabroni, S.; Rapisarda, P.; Palmeri, V. Effects of inert dusts applied alone and in combination with sweet orange essential oil against *Rhyzopertha dominica* (Coleoptera: Bostrichidae) and wheat microbial population. *Industr. Crops Prod.* **2014**, *61*, 361–369. [CrossRef]
8. Zeni, V.; Baliota, G.V.; Benelli, G.; Canale, A.; Athanassiou, C.G. Diatomaceous Earth for Arthropod Pest Control: Back to the Future. *Molecules* **2021**, *26*, 7487. [CrossRef]
9. Eroglu, N.; Emekci, M.; Athanassiou, C.G. Applications of natural zeolites on agriculture and food production. *J. Sci. Food Agric.* **2017**, *97*, 3487–3499. [CrossRef]
10. Korunić, Z. Diatomaceous earths, a group of natural insecticides. *J. Stored Prod. Res.* **1998**, *34*, 87–97. [CrossRef]
11. Floros, G.D.; Kokkari, A.I.; Kouloussis, N.A.; Kantiranis, N.A.; Damos, P.; Filippidis, A.A.; Koveos, D.S. Evaluation of the natural zeolite lethal effects on adults of the bean weevil under different temperatures and relative humidity regimes. *J. Econ. Entomol.* **2018**, *111*, 482–490. [CrossRef]
12. Athanassiou, C.G.; Kavallieratos, N.G.; Vayias, B.J.; Tomanović, Ž.; Petrović, A.; Rozman, V.; Cornel, A.; Korunić, Z.; Milovanović, D. Laboratory evaluation of diatomaceous earth deposits mined from several locations in central and southeastern Europe as potential protectants against coleopteran grain pests. *Crop. Prot.* **2011**, *30*, 329–339. [CrossRef]
13. Liška, A.; Rozman, V.; Korunić, Z.; Halamić, J.; Galović, I.; Lucić, P.; Baličević, R. The potential of Croatian diatomaceous earths as grain protectant against three stored-product insects. *Integr. Prot. Stored Prod. IOBC-WPRS Bull.* **2015**, *111*, 107–113.
14. Ziaee, M.; Moharramipour, S.; Dadkhahipour, K. Effect of particle size of two Iranian diatomaceous earth deposits and a commercial product on *Sitophilus granarius* (Col. *: Dryophthoridae). J. Entom. Soc.* **2013**, *33*, 9–17.
15. Korunić, Z. Overview of undesirable effects of using diatomaceous earths for direct mixing with grains. *Pestic. Phytomed.* **2016**, *31*, 9–18. [CrossRef]
16. Korunić, Z. Rapid assessment of the insecticidal value of diatomaceous earths without conducting bioassays. *J. Stored Prod. Res.* **1997**, *33*, 219–229. [CrossRef]
17. Marantos, I.; Christidis, G.E.; Ulmanu, M. Zeolite formation and deposits. In *Handbook of Natural Zeolites*; Bentham Science Publishers: Hilversum, The Netherlands, 2012; pp. 28–51. [CrossRef]
18. Reháková, M.; Čuvanová, S.; Dzivák, M.; Rimár, J.; Gavalová, Z. Agricultural and agrochemical uses of natural zeolite of the clinoptilolite type. *Curr. Opin. Solid State Mater. Sci.* **2004**, *8*, 397–404. [CrossRef]
19. Vayias, B.J.; Athanassiou, C.G.; Korunić, Z.; Rozman, V. Evaluation of natural diatomaceous earth deposits from south-eastern Europe for stored-grain protection: The effect of particle size. *Pest Manag. Sci.* **2009**, *65*, 1118–1123. [CrossRef]
20. Athanassiou, C.G.; Kavallieratos, N.G.; Andris, N.S. Insecticidal effect of three diatomaceous earth formulations against adults of *Sitophilus oryzae* (Coleoptera: Curculionidae) and *Tribolium confusum* (Coleoptera: Tenebrionidae) on oat, rye, and triticale. *J. Econ. Entomol.* **2004**, *97*, 2160–2167. [CrossRef]
21. Eroglu, N.; Sakka, M.K.; Emekci, M.; Athanassiou, C.G. Effects of zeolite formulations on the mortality and progeny production of *Sitophilus oryzae* and *Oryzaephilus surinamensis* at different temperature and relative humidity levels. *J. Stored Prod. Res.* **2019**, *81*, 40–45. [CrossRef]

22. Dowdy, A.K.; Fields, P.G. Heat combined with diatomaceous earth to control the confused flour beetle (Coleoptera: Tenebrionidae) in a flour mill. *J. Stored Prod. Res.* **2002**, *38*, 11–22. [CrossRef]
23. Dowdy, A.K. Mortality of red four beetle, *Tribolium castaneum* (Coleoptera: Tenebrionidae) exposed to high temperature and diatomaceous earth combinations. *J. Stored Prod. Res.* **1999**, *35*, 175–182. [CrossRef]
24. Bohinc, T.; Horvat, A.; Andrić, G.; Pražić-Golić, M.; Kljajić, P.; Trdan, S. Natural versus synthetic zeolites for controlling the maize weevil (*Sitophilus zeamais*)–like Messi versus Ronaldo? *J. Stored Prod. Res.* **2020**, *88*, 101639. [CrossRef]
25. Athanassiou, C.G.; Kavallieratos, N.G.; Arthur, F.H.; Nakas, C.T. Rating knockdown of flour beetles after exposure to two insecticides as an indicator of mortality. *Sci. Rep.* **2021**, *11*, 1145. [CrossRef] [PubMed]
26. Nayak, M.K.; Daglish, G.J.; Phillips, T.W.; Ebert, P.R. Resistance to the fumigant phosphine and its management in insect pests of stored products: A global perspective. *Annu. Rev. Entomol.* **2020**, *65*, 333–350. [CrossRef]
27. Sakka, M.K.; Athanassiou, C.G. Insecticidal effect of diatomaceous earth and pirimiphos-methyl against phosphine-susceptible and phosphine-resistant populations of two stored product beetle species. *Environm. Sci. Poll. Res.* **2021**, *28*, 33181–33191. [CrossRef]
28. Baliota, G.V.; Athanassiou, C.G. Evaluation of a Greek diatomaceous earth for stored product insect control and techniques that maximize its insecticidal efficacy. *Appl. Sci.* **2020**, *10*, 6441. [CrossRef]
29. Rojht, H.; Horvat, A.; Athanassiou, C.G.; Vayias, B.J.; Tomanović, Ž.; Trdan, S. Impact of geochemical composition of diatomaceous earth on its insecticidal activity against adults of *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae). *J. Pest. Sci.* **2010**, *83*, 429–436. [CrossRef]
30. *JMP®Software*, version 17.0.0; JMP Statistical Discovery LLC: Car, NC, USA, 2022.
31. Arthur, F.H. Structural pest management for stored product insects. In *Recent Advances in Stored Product Protection*; Athanassiou, C., Arthur, F., Eds.; Springer GmbH: Berlin/Heidelberg, Germany, 2018; pp. 65–81.
32. Babamir-Satehi, A.; Ziaee, M.; Ashrafi, A. Toxicity of chlorpyrifos against *Rhyzopertha dominica* and *Tribolium confusum* adults on different surfaces. *Toxin Reviews* **2017**, *36*, 57–62. [CrossRef]
33. Arthur, F.H.; Ghimire, M.N.; Myers, S.W.; Phillips, T.W. Evaluation of pyrethroid insecticides and insect growth regulators applied to different surfaces for control of *Trogoderma granarium* (Coleoptera: Dermestidae) the khapra beetle. *J. Econ. Entomol.* **2018**, *111*, 612–619. [CrossRef]
34. Arthur, F.H. Aerosols and contact insecticides as alternatives to methyl bromide in flour mills, food production facilities, and food warehouses. *J. Pest Sci.* **2012**, *85*, 323–329. [CrossRef]
35. Collins, P.J.; Nayak, M.K.; Kopittke, R. Residual efficacy of four organophosphate insecticides on concrete and galvanized steel against three liposcelid psocid species (Psocoptera: Liposcelidae) infesting stored products. *J. Econ. Entomol.* **2000**, *93*, 1357–1363. [CrossRef] [PubMed]
36. Vassilakos, T.N.; Athanassiou, G.G. Long-term residual efficacy of spinetoram on concrete and steel surfaces for the management of three stored product beetle species. *J. Econ. Entomol.* **2015**, *108*, 2090–2097. [CrossRef] [PubMed]
37. Rumbos, C.I.; Dutton, A.C.; Athanassiou, C.G. Efficacy of two formulations of pirimiphos-methyl as surface treatment against *Sitophilus granarius*, *Rhyzopertha dominica*, and *Tribolium confusum*. *J. Pest Sci.* **2014**, *87*, 507–519. [CrossRef]
38. Vassilakos, T.N.; Athanassiou, C.G.; Chloridis, A.S.; Dripps, J.E. Efficacy of spinetoram as a contact insecticide on different surfaces against stored-product beetle species. *J. Pest Sci.* **2014**, *87*, 485–494. [CrossRef]
39. Toews, M.D.; Subramanyam, B.; Rowan, J.M. Knockdown and mortality of adults of eight species of stored-product beetles exposed to four surfaces treated with spinosad. *J. Econ. Entomol.* **2003**, *96*, 1967–1973. [CrossRef] [PubMed]
40. Carlson, S.D.; Ball, H.J. Mode of action and insecticidal value of a diatomaceous earth as a grain protectant. *J. Econ. Entomol.* **1962**, *55*, 964–970. [CrossRef]
41. Arthur, F.H.; Athanassiou, C.G.; Morrison, W.R., III. Mobility of stored product beetles, after exposure to a combination insecticide containing deltamethrin, methoprene, and a piperonyl butoxide synergist depends on species, concentration, and exposure time. *Insects* **2020**, *11*, 151. [CrossRef]
42. Agrafioti, P.; Brabec, D.; Morrison, W.R., III.; Campbell, J.F.; Athanassiou, C.G. Scaling recovery of susceptible and resistant stored product insects after exposure to phosphine by using automated video tracking software. *Pest Manag. Sci.* **2020**, *77*, 1245–1255. [CrossRef]
43. Athanassiou, C.G.; Kavallieratos, N.G.; Rumbos, C.I.; Stavropoulos, D.J.; Boukouvala, M.C.; Nika, E.P. Laboratory studies of the behavioral responses of *Tribolium confusum* and *Ephestia kuehniella* to surfaces treated with diatomaceous earth and spinosad formulations. *J. Pest Sci.* **2017**, *99*, 299–311. [CrossRef]
44. Cook, D.A.; Collins, D.A.; Collins, L.E. Efficacy of diatomaceous earths, applied as structural treatments, against stored product insects and mites. *HGCA Project Rep.* **2004**, *344*, 50.
45. Collins, D.A.; Cook, D.A. Laboratory evaluation of diatomaceous earths, when applied as dry dust and slurries to wooden surfaces, against stored-product insect and mite pests. *J. Stored Prod. Res.* **2006**, *42*, 197–206. [CrossRef]
46. Collins, D.A.; Cook, D.A. Laboratory studies evaluating the efficacy of diatomaceous earths, on treated surfaces, against stored-product insect and mite pests. *J. Stored Prod. Res.* **2006**, *42*, 51–60. [CrossRef]
47. Schöller, M.; Reichmuth, C. Field trials with the diatomaceous earth SilicoSec®for treatment of empty rooms and bulk grain. *Julius-Kühn-Archiv.* **2010**, *425*, 899–905.

48. Ertürk, S.; Atay, T.; Toprak, U.; Alkan, M. The efficacy of different surface applications of wettable powder formulation of Detech®diatomaceous earth against the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae). *J. Stored Prod. Res.* **2020**, *89*, 101725. [CrossRef]
49. Arthur, F.H. Toxicity of diatomaceous earth red flour beetles and confused flour beetles (Coleoptera: Tenebrionidae): Effects of temperature and relative humidity. *J. Econ. Entomol.* **2000**, *93*, 526–532. [CrossRef]
50. Vayias, B.J.; Athanassiou, C.G. Factors affecting the insecticidal efficacy of the diatomaceous earth formulation SilicoSec against adults and larvae of the confused flour beetle, *Tribolium confusum* DuVal (Coleoptera: Tenebrionidae). *Crop. Prot.* **2004**, *23*, 565–573. [CrossRef]