Research Article

Insecticidal Potential of Native Diatomaceous Earth Against *Sitophilus granarius* (Coleoptera: Curculionidae)

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Abstract | The granary weevil, *Sitophilus granarius* is one of the serious pests of cereal crops worldwide. To control this pest, diatomaceous earth (DE) formulation consisted of diatoms, obtained from Niğde Province, Turkey, was tested against adults of the granary weevil, *Sitophilus granarius* (L.) under laboratory conditions (25±1°C, 60±5% RH). Diatomaceous earth was applied to wheat grain at the rates of 100, 250, 300, 500, 750, 1000, 1500 and 2000 mg/Kg. Adult mortalities were assessed after 7, 14 and 21 days. Mortality rates rose with increases in dosage and length of exposure. Significantly higher mortality (89.75±3.66%) after one week occurred in grains treated with the highest concentration (2000 mg/Kg). At the highest concentration (2000 mg/Kg), the mortality reached as high as 100% after 2 weeks, while the lowest significant percent mortality (1.00±1.00%) was recorded in the control one week after exposure and only increased to 3.53±0.00% after 3 weeks. Percent weight loss in grain was also determined after 21 days. A significantly higher percent weight loss (7.29±0.11%) was found in the control, while no weight loss was observed in grains treated with the highest concentration (2000 mg/Kg). The grains were also observed for adult emergence for a period of 45 days. More than 90% reduction in adult emergence was recorded at concentrations of 1500 and 2000 mg/Kg. Conversely, the highest number of adults emerged at a dosage of 100 mg/Kg and in the control, i.e. 114.67±7.42 and 143.33±11.66 adults, respectively. The present study proposed that DE might potentially be effective in controlling *S. granarius* based on its ability to cause desiccation to the insects' protective cuticular wax layer. In order to develop environmentally sound control programs for *in sito* management of granary weevils, it will be necessary to expand the present study based on life table to get a comprehensive understanding and to determine the effect DE has on granary weevils and other grain pests when it is combined with plant extracts.

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Introduction

The granary weevil, *Sitophilus granarius* L. (Coleoptera: Curculionidae) is a major pest of stored rice in many developing countries. They attack wheat, corn, oats, rye, barley, sorghum, dried beans and other cereals. It causes economic losses and seed viability (Mebarkia et al., 2010).

To control this pest, different preventive measures have been applied to minimize the loss. These methods include cultural, physical and chemical methods. But farmers mostly rely on heavy insecticides uses (White and Leesch, 1996; Tomlin, 2000). The use of insecticides has many adverts effect like health problems, pollution, environment contamination, secondary pest outbreak, effect on non-target organism and residual effect. Moreover, the residual activity of insecticides applied to stored food has increased public concern worldwide (White and Leesch, 1995).

These limitations have stressed the researcher for the development of alternative pest control strategies such as use of botanicals, microbial control, biological control, insect growth regulators and inert dusts (Athanassiou et al., 2005)

Diatomaceous earth (DE) is one of the most promising alternatives to contact insecticides. Diatomaceous earth is a natural product. Because it can cause damage to insect cuticle, resulting in water loss and desiccation, therefore it has been used as insecticide (Subramanyam and Roesli, 2000). Uses of diatomaceous earth in field and in stored products have increased over the past few years (Mewis and Ulrichs, 2001). The nanoparticles of DE can be applied directly to the grain. Advantages of these nanoparticles include: 1) DE formulation offer physical control method without residual concern and are very effective against a wide range of pest species but are non-toxic to mammals, 2) They can be effortlessly removed from the stored product during processing and 3) No insecticide resistance recorded (Korunic,1998; Subramanyam and Roesli, 2000; Stathers et al., 2004).

Keeping in view the importance of DE against stored grain pests, the current study was carried out to determine the effect of nanoparticles diatomaceous earth against *S. granaries*. The objectives of the study were (a) to evaluate the insecticidal potential of local diatomaceous earth against *S. granaries* (b) to determine the influence of time of exposure and dose rate and (c) to monitor the ability of *S. granaries* for offspring production in the treated grain for a given period of time.

Materials and Methods

The study was conducted in the Entomology lab of the Department of Plant Production and Technologies, Faculty of Agricultural Sciences and Technologies, Nigde Ömer Halisdemir University, Niğde, Turkey, during 2015-16.

Insect culture

Adults of the *S. granarius* were mass-produced on wheat grains in plastic jars (1000 ml) covered with a piece of fine cloth. The cultures were maintained at 25±1°C and 60±5% R.H in a controlled environment chamber for three generations following Kljajić and Perić (2006).

DE formulation

The DE nanoparticles formulation was obtained from Department of Geological Engineering, Faculty of Engineering, Nigde Ömer Halisdemir University, Turkey. The DE formulation was stored in the laboratory at ambient temperature (25±2°C), until the beginning of experiments.

Insect contact bioassay

For this purpose, the wheat grains were autoclaved at 121°C for 30 minutes. After sterilization, the grains were air dried and then DE was applied to sterilized wheat grain at the rates of 100, 250, 300, 500, 750, 1000, 1500 and 2000 mg/Kg in 50 ml glass vials separately (Athanassiou et al., 2005). Seven hundred milligrams of wheat were used in each replication and each treatment was replicated thrice. DE was mixed thoroughly with the grains by continuous shaking. When the DE was fully admixed with the grains, twenty *S. granarius* adults per 50ml glass vials were released in each treatment. There were nine treatments including control followed Randomized Complete Block design. Adult mortalities were assessed after 7, 14 and 21 days. After 21 days, all the insects (dead and alive insects) were removed from each treatment in order to trace the fresh adult emergence. These glass vials were then kept under the same experimental condition for checking of F1 adult emergence. These vials were checked daily for 45 days until no adults emerged.

When the adults were removed after 21 days, these grains were also observed for the percent weight loss. After 21 days, the treated grains were again weighed and the percent weight loss were recorded by formula as suggested by Khattak et al. (1987).

$$\%\text{Weight loss} = \frac{(\text{weight of the control sample} - \text{weight of the sound} + \text{damaged grains in the test sample})}{\text{weight of the control sample}} \times 100$$

Statistical analyses

The data recorded for mean percent mortality, weight loss and adult's emergence in different treatments were subjected to statistical analysis by ANOVA. LSD test was used for means comparison in different treatments (Steels and Torrie, 1960). The mortality data were processed and corrected by probit analysis as proposed methodology by Finney (1978) using computer software developed by Chi (1997).

Results and Discussion

Adult mortality

Higher mortality was observed with increasing concentration and length of exposure. Significantly higher mortality (*DF* 3,8; *F* 9.77; *P* 0.00) (89.75±3.70%) after one week occurred in grains treated with the highest concentration (2000 mg/Kg), followed by 72.75±13.75% mortality in grains treated with 1500 mg/Kg concentration (Table 1). The lowest mortality of *S. granarius* adults was recorded in the control (1.00±1.00%). Similar trends in % mortality was observed after 2nd and 3rd week of treatments (*DF* 3,8; *F* 33.46; *P* 0.00). At the higher concentration (2000 mg/Kg and 1.50 g/Kg of DE) % mortality reached 98.75±1.25% after 2 weeks, while the mean percent mortality in control after 3 weeks reached only 3.55±0.00%).

Table1: *Mean percent mortality of S. granarius in wheat grains treated with different concentrations of Diatomaceous earth (DE).*

Concentrations (mg/Kg)	Percent mortality		
	7 days	14 days	21 days
100	13.17±1.66de	20.21±4.41e	20.21±4.41e
250	16.50± 0.00de	61.88±2.89d	61.88±2.89d
300	19.83±1.66de	66.88±2.89cd	66.88±2.89cd
500	33.00±13.47cd	77.75±12.11bcd	77.75±12.11bcd
750	52.75± 8.32bc	91.25±5.15ab	91.25±5.95ab
1000	54.25±15.85bc	81.25±2.39bc	81.25±2.76bc
1500	72.75±13.75ab	98.75±1.25a	98.75±1.44a
2000	89.75±3.66a	98.75±1.25a	98.75±1.44a
Control	1.00± 1.00e	3.55±1.67e	3.55±0.00e
LSD value	21	20	20

Mean values followed by different letter in each column are significantly different at α=0.05

Determination of LC_{50}

LC_{50} value of 780 mg/Kg was calculated for *S. granarius* adults after 1st week exposure to DE formulation (Figure 1). LC_{50} value of 220 mg/Kg and 150 mg/Kg were calculated for *S. granarius* adults after 2nd and 3rd weeks exposure to DE formulation. LC_{50} value of DE decreased with increases in time of exposure.

Percentage weight loss and adult emergence of sitophyllus granarius

Increasing DE concentration has a significant effect on percent wheat grain loss (*DF* 2,8; *F* 83.11; *P* 0.00). High concentration of DE formulation significantly reduces the weight loss caused by *S. granarius* infestation and no weight loss was observed in grains treated with the highest dosage (2000 mg/Kg) (Figure 2). Significantly higher percent weight loss (*DF* 2,8; *F* 60.71; *P* 0.00). (7.29±0.11%) was found in the control. Exposure of *S. granarius* adults to different concentrations of DE nanoparticles significantly reduced F1 adult emergence. At high concentration of 2000 mg/Kg, more than 90% reduction (only 2.70%) in F1 adult emergence was recorded. At a concentration of 1500 mg/Kg DE, only 21.00±4.16% adult emerged after 45 days. Conversely, the highest number of adults emerged at a concentration of 100 mg/Kg and in the control (114.67±7.42 and 143.33±11.66 adults, respectively) after 45 days.

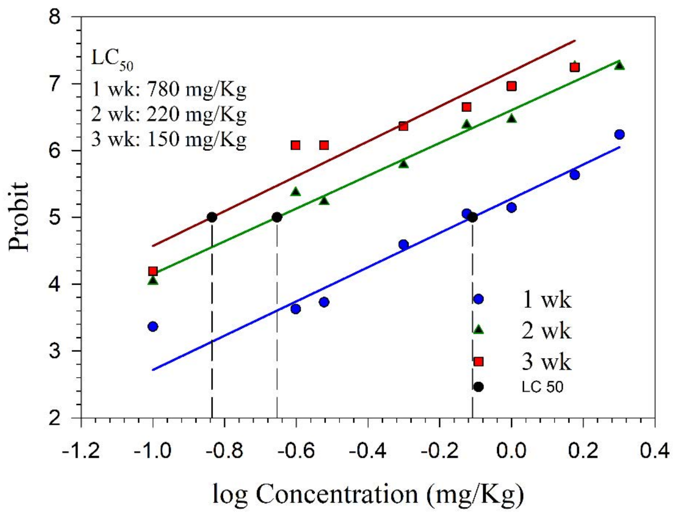

Figure 1: *Determination of the LC50 values for S. granarius adults after different weeks exposure to DE formulation.*

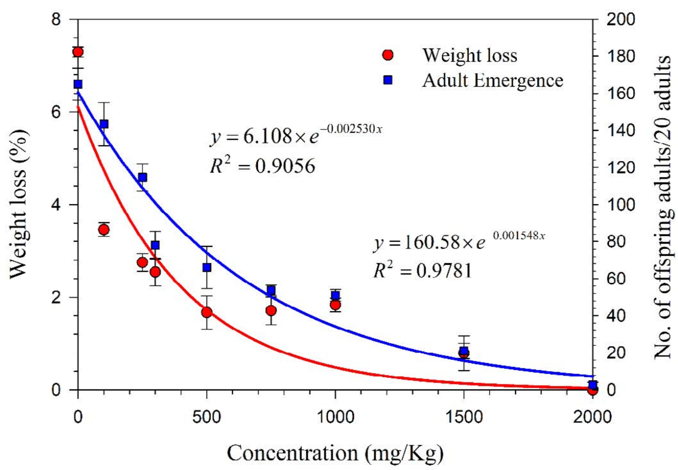

Figure 2: *Determination of weight loss in wheat grains and percent adult emergence of S. granarius exposed to different concentration of DE formulation.*

In this study different dosages of diatomaceous earth nanoparticles were tested against *S. granarius*. The data showed that higher mortality was observed with increasing concentration and length of exposure. High concentration of 2000 mg/Kg gave 100% mortality after 2 weeks while the lowest mortality of 3.5 % was recorded in control after 3 weeks. The data further showed that increase in time of exposure, the LC_{50} value of diatomaceous earth decreased from 780 mg/Kg to 150 mg/Kg.

The effectiveness of DE increases with the increase in time exposure because movement of adult insects increases the chances of contact with the DE particles. The contact of DE particles with the insect cuticles causes desiccation which lead to the death of the insect (Fields and Korunic, 2000; Arthur, 2000). Athanassiou et al. (2006) reported that mortality of the stored grain pests increases with the increase in dose concentration. They reported 100 % mortality of *C. ferrugineus* at 150 ppm DE formulation.

Adults of *S. granarius* caused significant weight loss in the control whereas no weight loss was observed in grains treated with high concentration of 2000 mg/Kg of DE formulation. More than 80-100% reduction in *S. granarius* adult emergence was recorded at concentrations of 1500 and 2000 mg/Kg, while the lowest concentration and control had significantly high adults emergence. No progeny of the *S. oryzae* were produced at the highest concentration of 250 mg/Kg of DE because all of the exposed adults died after 14 days of exposure (Athanassiou et al., 2005). They further reported that wheat grains treated with DE formulations (100 and 1500 mg/Kg of wheat) offered a significance control against *S. oryzae* and *T. confusum* infestations.

The present study suggests that DE nanoparticles could effectively control *S. granarius* infestation in stored grain. Similar studies with DE nanoparticles can be carried out for other stored-product pests. For a comprehensive understanding of the long term effect, it is suggested to expand the scope of present work to include effects of DE on life table of *S. granarius*. These studies will help us to predict potential damage and ways to extend the grains shelf life during storage.

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Novelty Statement

This study highlights the importance of diatomaceous earth formulation against *Sitophyllus granarius*. DE consisted of diatoms inert dust particles which are non-poisonous to human being. They kill insects by desiccation. The present study suggests that DE nanoparticles could effectively control stored grain pests infestation in stored grain.

Author's Contribution

MS and MNN conducted the research trial, compiled the data and wrote the article, AG supervised the whole research and OE provide the nano particle formulation of DE for experiments.

Conflict of interest

The authors have declared no conflict of interest.

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