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Combination of diatomaceous earth and temperature to control *Sitophilus zeamais* (Coleoptera: Curculionidae) in pearl millet seeds

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Abstract

Diatomaceous earth (DE) is an efficient grain protectant in IPM programs for controlling insects in stored seeds and grains. Pearl millet, *Pennisetum glaucum* has being produced in savanah areas of Brazil, known as Cerrado, as a soil cover crop and for direct cattle feeding during the dry season, whereas the grain is used for feed. The objective of this study was to verify the efficacy of DE under different temperatures for control ling of *Sitophilus zeamais* in stored millet. The tests were conducted with 20 unsexed adults in 100 g of pearl millet of the varieties ADR300 and ADR500 maintained in 500 ml glass flasks. The grain was treated with DE (KEEPDRY®) in the dosages of 500, 750, 1,000 and 1,500 g/t, under temperatures of 15, 25 and 30 °C. The insect mortality was evaluated on the 3rd, 7th and 14th days after treatment; after the adults were removed and the number of offspring were counted. The three highest doses of DE at 25 and 30 °C were significantly better at controlling *S. zeamais* compared to lower doses of DE. There was no significant difference in offspring production in DE treated samples, but it was lower than that in the control. Therefore, DE in dosages above 750 g/t is an efficient grain protectant against *S. zeamais* in stored pearl millet in tropical and subtropical climate.

Key words: Inert dust; insect biology; maize weevil; pearl millet seeds; stored product protection.

Introduction

Pearl millet, *Pennisetum glaucum* (Poaceae), has being cultivated as a soil cover crop, for direct cattle feeding during the dry season, and for feed (as grain). In Brazil, more than 2 million hectares are planted with pearl millet (Bonamigo, 2004), especially in Cerrado. Under optimal humidity and temperature conditions, millet plants can produce up to 70 ton/ha of forage and the grain production can reach 1,500 kg/ha.

The maize weevil, *Sitophilus zeamais* Motschulsky, 1855 (Coleoptera: Curculionidae), is the major primary pest of pearl millet in tropical areas. It also attacks corn, wheat, rice and sorghum grain and processed cereals, such as pasta and dehydrated cassava (Dobie et al., 1984). This species is very active and can infest the grain in the field and in storage (Pacheco and Paula, 1995) and always need to be controlled because of their high reproductive potential.

Diatomaceous earth (DE) has been used for insect control around the world and, according to Mital and Wrightman (1989) and Arthur (1996), interest in this technique has increased because the active ingredients for insect control in storage is restricted to four or five products, mainly due to insect resistance problems.

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According to Subramanyam and Roesli (2000), the insect death is attributed to desiccation caused by adsorption and abrasive properties of DE that absorbs the wax layer of the epicuticle, causing body water loss and death within hours or days. Aldryhim (1990, 1993) observed that the granary weevil, *Sitophilus granarius* (L.) (Coleoptera: Curculionidae) and the lesser grain borer, *Rhyzopertha dominica* (Fabr.) (Coleoptera: Bostrichidae), are more susceptible to DE at 30 ºC than at 20 ºC. However, *Tribolium confusum* (Duv.) (Coleoptera: Tenebrionidae) was more susceptible to DE at low temperatures.

When choosing between traditional chemical insecticides and DE for insect control in stored grains, the advantages and disadvantages of both should be evaluated. Chemical insecticides can eliminate insects very rapidly, reducing the chances of reproduction. However, the selective pressure of insecticides results in resistance development and contamination with residues that may cause human intoxication and environmental problems. Diatomaceous earth is not toxic to humans, domestic animals, and the environment, and does not leave toxic residues in the grain and by-products. However, its direct action on insects is slower than that of insecticides, permitting oviposition and offspring production, albeit in low number (Fields, 2000).

The objective of this research was to verify the efficacy of DE at different dosages and under different temperatures for controlling *S. zeamais* in stored millet grains.

Materials and methods

The specimens of *S. zeamais* were reared on corn at 13 % moisture content (wet basis) in an environmental chamber at 25 ± 2 ºC and 65 ± 5 % r.h., and 12 h of photophase.

The DE used for the experiments was the commercial product Keepdry®, which is composed of: 88 to 90 % of SiO_2, particles of 10-15 mm; apparent density of 200-230 g/L; color beige, aspect of a dry loose light powder, insoluble in water and free from foreign material. Four doses of diatomaceous earth were used: 500 g/ton (T1), 750 g/ton (T2), 1,000 g/ton (T3) and 1,500 g/ton (T4). The treatments were replicated three times, each one consisting of 100 g of pearl millet of the varieties ADR300 and ADR500 previously desinfested at -25 ºC for three days. The seeds were treated with DE in bags and homogenized by vigorous hand agitation for 2 minutes, and then placed in plastic flasks with a 500 mL volume. Twenty, unsexed, 7 to 14 days old adult *S. zeamais* were placed in each flask, which was covered with a screen lid. The flasks were kept in chambers maintained at 15, 25 and 30 °C, with a relative humidity of 65 ± 5 % and 12 h of photophase.

In all treatments, mortality was recorded on the 3rd, 7th and 14th days , after which the adults were removed from the grain. Insects that didn't move after being touched with a brush, after two minutes, were considered dead. On the 56th day, the flasks were opened and the progeny were counted.

During the experiment the temperature and relative humidity inside the chambers were recorded. The moisture content of the grain was determined by the oven method (Lazzari, 1997).

The mean mortality and the standard error (SE) were calculated for each sampling date for each treatment. The data were analyzed using analysis of variance and means were compared by Tukey´s multiple range test at 5 % probability.

Results

The mortality was significantly affected by the doses of DE interacting with temperature and exposure time ($p < 0.05$). The mean mortality was higher at 25 and 30 ºC for all three doses of DE in the varieties ADR300 and ADR 500 (Table 1 and Table 2). Figure 1 shows DE particles adsorbed on the maize weevil, causing the wax layer removal and the insect death.

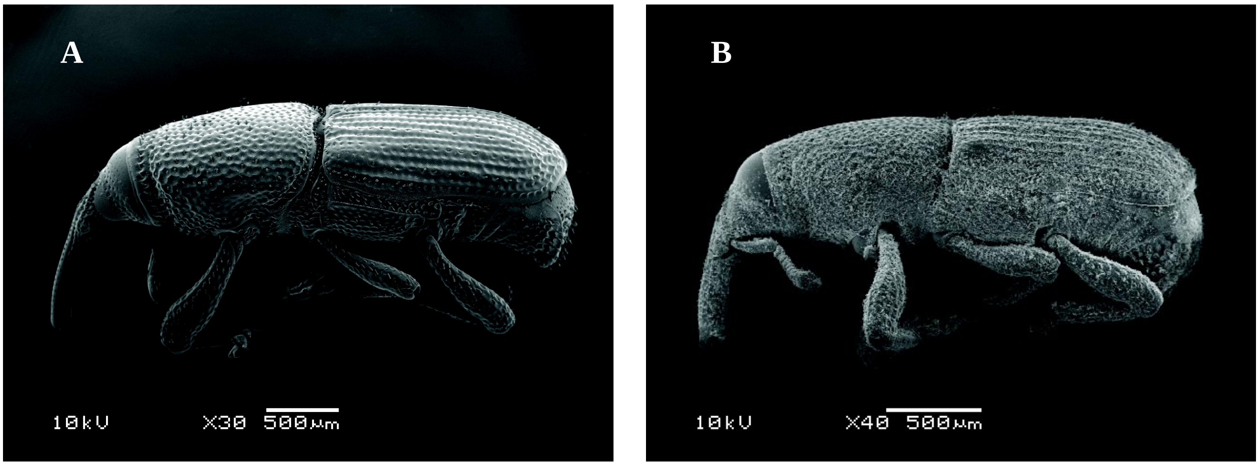

Figure 1. Electronmicrographs of *S. zeamais*. A. Without diatomaceous earth treatment; B. Treated with diatomaceous earth at 750 g/ton.

Table 1. Mean mortality (%) of *Sitophilus zeamais* in pearl millet seeds of variety ADR 300 treated with different dosages of diatomaceous earth and temperature; 65 ± 5 % r.h. and 12 h of photophase.

	Mean mortality of insects (%)								
ADR 300	15 ºC			25 ºC			30 ºC		
Days after treatment	3	7	14	3	7	14	3	7	14
Control	0 d	0 c	11 c	0 c	2 c	14 c	0 c	3 c	14 b
500 g/ton DE (T1)	2 c	75 b	94 ab	8 b	80 b	96 ab	11 b	89 ab	100 a
750 g/ton DE (T2)	5 b	86 ab	98 a	10 ab	88 ab	99 a	13 ab	96 a	100 a
1,000 g/ton DE (T3)	8 a	87 a	99 a	13 a	91 a	100 a	17 a	99 a	100 a
1,500 g/ton DE (T4)	9 a	90 a	100 a	15 a	93 a	100 a	21 a	100 a	100 a

Table 2. Mean mortality (%) of *Sitophilus zeamais* in pearl millet seeds of variety ADR 500 treated with different dosages of diatomaceous earth and temperature; 65 ± 5 % r.h. and 12 h of photophase.

	Mean mortality of insects (%)								
ADR 500	15 ºC			25 ºC			30 ºC		
Days after treatment	3	7	14	3	7	14	3	7	14
Control	0 c	0 b	12 b	0 d	0 c	8 c	0 e	0 c	18 b
500 g/ton DE (T1)	2 bc	76 ab	92 a	2 c	80 b	98 ab	8 d	91 b	100 a
750 g/ton DE (T2)	3 b	85 ab	99 a	8 b	86 ab	100 a	12 c	94 ab	100 a
1,000 g/ton DE (T3)	3 b	91 a	100 a	9 b	93 a	100 a	15 ab	96 a	100 a
1,500 g/ton DE (T4)	12 a	92 a	100 a	18 a	96 a	100 a	20 a	99 a	100 a

For ADR 300 (Table 1), the treatments with DE had significantly higher mortality than the controls after the 3rd day of exposure. At 15 ºC, the accumulated mortality was significantly higher than the controls by the 7th day of exposure for treatments with the two highest doses, T3 and T4, with mortality of 87 and 90 %, respectively. By the 14th day, the difference among the doses of DE were not significantly different, independent of temperature. In the control, the accumulated mortality was only 11 % by the 14th day, when the insects were removed to evaluate the number of 2nd generation progeny. At 25 ºC, the accumulated mortality by the 7th day was significantly different from the control for treatments T3 and T4, with mortality of 91 and 93 %, respectively. By the 14th day there were no significant difference among the higher doses of DE, with 99 % of mortality for T2, and 100 % for T3 and T4. The mortality in the control was 14 % after 14 days of monitoring. At 30 ºC, the mortality by 3rd day was 21% for the treatment with 1,500 g/ton. By the 7th day of exposure there were no significant differences in mortality among the highest doses of DE. By the 14th day, all treatments caused 100 % insect mortality.

For ADR 500 (Table 2), the mortality was already 91 % at 1,000 g/ton and 92 % to 1,500 g/ton by the 7th day at 15 ºC. By the 14th day, there were no significant differences in the mortality at all DE doses. In the control, the accumulated mortality was only 12 % by the 14th day of the experiment. At 25 ºC, by the 3rd day, the accumulated mortality was significant ly higher than the control among the three lowest doses of DE and the dose of 1,500 g/ton with 18 % mortality. The mortality in the control was 8 % after 14 days of monitoring. At 30 ºC, the accumulated mortality was significantly different among the lowest doses of DE. The accumulated mortality after the 7th day of exposure was 100 % for all DE treatments. In the control, the mortality of *S. zeamais* after the 14th day was only 14 %.

Discussion

The results showed that the mortality of maize weevils in seeds of pearl millet treated with DE was much higher than in corn grain with the same doses and temperatures (Ceruti et al., 2005). High mortality in the control suggests that the adaptation of the insect to the kernel seed coat may change with variety or plant species.

Previous studies showed that DE is less effective against insects at low temperatures (Aldryhim, 1990, 1993; Collins et al., 2001), whichwas probably due to reduced adsorption of DE particles when insect mobility decreases. Highest temperatures would increase insect movement, increase contact with DE, greater cuticular damage. Increased temperature would also lead to a higher metabolic rate, which would result in increased water needs. In this study, there was no significant difference for the effectiveness of DE combined with temperature.

The results of this research agree with those obtained by Pinto Jr. (1994), who observed a correlation between the doses of DE and the time of exposure of *Sitophilus* spp. in corn, reaching 100 % mortality after 19 days of exposure to 500 and 750 g/ton of DE. Lorini and Schneider (1994), testing DE in doses of 500 and 1,000 g/ton with *S. oryzae*, obtained, after seven days of treatment, a mortality of 87 and 100 %, respectively.

At 25 and 30 ºC, after 56 days the number of offspring in the control was significantly higher than at 15 ºC, in all treatments. For ADR 300, progeny in the control was smaller than that in the ADR 500 parcels at 25 ºC and 30 ºC and 37 insects at 15 ºC, but, at 15 ºC, it was greater. The cause of such differences might be explored to point out the mechanisms involved. At low temperature, as 15 ºC, insect reproduction is reduced, however an interaction with DE was not observed. These results showed that infestation may be severe in both varieties of pearl millet seeds under high temperatures, as those prevailing in the Brazilian savanah. DE has proven to be an efficient grain protectant under such conditions.

Paula (2001) observed that the number of progeny of *Sitophilus* spp. in paddy rice was inversely proportional to the dose of DE. Mewis and Reichmuth (1998) observed similar results in a laboratory experiment with *S. granarius* exposed to DE treated wheat grains at 25 ºC and 14.5 % r.h. They reported that the adults died within a few days, which was, however, time enough to produce progeny resulting in a considerable population increase after 42 days.

The moisture content of the seed was 11 % at the beginning of the experiment in all treatments. At the end, the moisture content decreased slightly to 10.8 % at 15 ºC; 10.2 % at 25 ºC and 9.5 % at 30 ºC with no significant difference among them, showing that DE keeps the moisture content down. In the control, the MC at the end of the experiment was 11.5 %, showing a slight increase without the protectant DE particles adhered to the kernel.

Conclusion

Temperatures of 25 ºC and 30 ºC, which are the prevailing conditions in silos and storage units year round in tropical areas, favors the action of DE against *S. zeamais* in stored pearl millet.

The treatments with DE at 750, 1,000 and 1,500 g/ton provide a satisfactory control of *S. zeamais* by the 7th day after treatment. Also, long-term control is achieved because the survival of insect progeny is reduced.

The grain moisture content is maintained in a safe range when grain is protected with DE, avoiding deterioration.

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