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JOURNAL OF THE KANSAS ENTOMOLOGICAL SOCIETY
59(2), 1986, pp. 350-355***Bacillus thuringiensis* Applied to Stored Corn
Using Grain Drying Fans¹**WM. H. MCGAUGHEY
U.S. Grain Marketing Research Laboratory,
Agric. Res. Serv., USDA, 1515 College Ave.,
Manhattan, Kansas 66502

ABSTRACT: *Bacillus thuringiensis* was applied to the surface layer of stored corn as an airborne dust carried into the grain by the downward air flow of drying fans. At air flow rates of typical farm-bin grain drying systems approximately 25% of the dust penetrated 2.5 to 12.5 cm into the corn and prevented infestation by Indianmeal moths, *Plodia interpunctella* (Hübner). Data are presented on the distribution of spores and toxic activity among particle size fractions of the dust formulation, the effects of air flow rate on depth of dust penetration, and the depth of dust penetration in farm grain drying bins.

Dust and WP (wetttable powder) formulations of *Bacillus thuringiensis* (BT) have been tested in farm and commercial grain storage facilities for the prevention of Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), infestations (McGaughey, 1985). However, the auger and rake-in methods of application to the top 10 cm of grain are so labor intensive, they possibly discourage use of the insecticide for this purpose. A more convenient method of application is needed.

Quinlan (1972, 1979, 1980) achieved limited success applying malathion to shelled corn as an aerosol propelled through the grain mass by the drying and aeration systems. Particles with a mass median diameter of ca. 0.5 μm penetrated as far as 3.8 m into the grain. This was advantageous because there was no need to turn or mix the grain. This paper discusses the application of a dust formulation of BT to the surface layer of corn by the downward air flow of drying fans.

Methods and Materials

Top-Side® Dipel dust, a formulation containing 4000 International Units of BT subsp. *kurstaki* per mg in a wheat flour carrier is labeled for application to grain at a rate of 39 g/m² of surface area. The range of particle sizes in the formulation was determined by passing the dust through a series of U.S. standard sieves (125, 88, 63, and 44 μm) and weighing each fraction. The <44 μm fraction was separated using a sonic sifter with 20 and 10 μm sieves.

The fractions were analyzed to determine the proportions of spores, crystal toxin, and insecticidal activity in each one. Viable spores were counted by suspending 10 mg of each fraction in 10 ml of sterile water, preparing serial 1:10 dilutions of the suspensions, and spreading 0.1 ml of each dilution on half-strength nutrient agar plates. Colonies were counted after incubation at 27°C for 24 hr. The crystal toxin content was determined using the modification of the rocket immunoelectrophoresis technique described by Andrews et al. (1980) for mea-

¹ This paper presents the results of research only. Mention of a proprietary product or pesticide does not constitute an endorsement or a recommendation for its use by USDA.

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suring the toxin in mixtures of crystals, spores, cell debris, and carrier materials. Insecticidal activity was determined by bioassay using neonate larvae of the tobacco hornworm, *Manduca sexta* (L.), as described by Schesser et al. (1977).

Tests were conducted in the laboratory to determine the depth of dust penetration into shelled corn using six different air flow rates. A 30.5 cm diameter × 38 cm deep metal cylinder was constructed with a perforated floor 7.5 cm above the solid bottom to simulate a grain bin with a perforated floor. A vacuum hose was attached to the side of the cylinder below the perforated floor and routed through a flow meter to an adjustable vacuum pump to produce a downward air flow. Air flow rates of 0, 457, 914, 1524, 2286, and 3048 liters/m²/min were tested. These rates are equivalent to 0, 1.5, 3.0, 5.0, 7.5, and 10 CFM/ft².

The cylinder was filled level with the top with shelled corn that was dockage free. Dust deposition in the top five 2.54 cm layers of corn was measured. In order to withdraw samples from each depth without physically disturbing the grain and causing the dust to sift farther into the bin, five 2.54 cm deep × 16 cm square wire mesh baskets were constructed and embedded below one another in the corn. The wire mesh had 6.3 mm openings to provide unrestricted air flow yet not allow corn kernels to fall through. After treatment the five baskets were removed and the corn from each was placed in a separate container for later analysis. The bin was cleaned with compressed air and refilled with clean corn for each treatment. Three replications were done for each air flow rate.

Equipment was not available for dispersing the small amount of dust appropriate for this column in a confined space above the grain. Therefore, the column was placed on the floor in a larger 1.435 m diameter bin and an Atomite® model 1907 electrically operated duster was used to disperse dust inside the larger bin. Approximately 65 g of dust was applied intermittently over a 9-min period for each treatment. Vacuum was applied to the container of corn for 20 min during and after dispersion of the dust.

Dust deposits were measured by making viable spore counts on grain from each sample depth. Five grams of corn were washed in 50 ml sterile water, the wash water was serially diluted (1:10), and 0.1 ml of each dilution was spread on half-strength nutrient agar plates. Colonies were counted after incubation at 27°C for 24 hr.

Corn was treated in 4 circular metal corn drying bins on farms in Merrick County, NE, to evaluate dust penetration using typical rates of fixed air flow. The normally upward air flow of the drying systems was reversed to a downward flow by reversing the fans. The bin sizes, air flow rates, and dockage content of the corn are summarized in Table 3. Air flow rates were determined by taking static pressure measurements with a magnehelic differential pressure gage attached to a probe at 0.61 m intervals at 9 locations within each bin and converting the measurements to air flow rates using charts published by Shedd (1953). The dust was dispersed into the headspace of each bin through the roof opening using the Atomite duster. The fans were operated for 40–60 min during and after treatment until all the dust had settled from the air.

The top five 2.54 cm deep layers of grain were sampled at 3 locations in each bin for determining the proportion of spores deposited in each layer. The wire mesh sample baskets and spore counting technique described for the previous test were used. Composite 10 cm deep samples were taken at 5 locations in each

Table 1. Percentages of the total dust, crystal toxin (CT), spores, and toxicity associated with each particle size range of Top-Side Dipel dust.^a

Size (μm)	Dust ^b	CT	Spores	Toxicity
>125	2.45	1.24	2.65	1.78
89-125	19.66	6.18	9.57	10.96
64-88	11.42	3.64	9.15	6.17
45-63	12.09	7.91	15.53	13.99
21-44	43.76	77.71	60.91	63.29
10-20	6.27	3.14	2.05	3.73
<10	0.64	0.19	0.14	0.08

^a Means of 3 samples.

^b 96.29% of sample accounted for; 3.71% lost in separation process.

bin for bioassay of the toxicity of the deposits. One hundred eggs from a laboratory colony of Indianmeal moths were placed on 250 g of each sample in a mason jar. The samples were held at 25°C and 60–70% RH until adults emerged. Posttreatment Indianmeal moth infestation levels in the bins were monitored by counting larvae, pupae, and empty cocoons in 2 × 50 cm strips of corrugated paper that were placed on the grain surface in each bin at a rate of 1 strip/2.32 m² of surface. The strips were replaced monthly. Counts were expressed as insects per strip per 30 days. Infestation levels were compared with those in 10 untreated bins in the same area.

Results and Discussion

The particle size, crystal toxin, spore, and toxicity distributions for the dust formulation are summarized in Table 1. The mass median particle size was 44 μm and 44% of the formulation was in the 21–44 μm size range. The amounts of viable spores, crystal toxin, and toxic activity were even more concentrated in the predominant size range. Sixty-one percent of the spores, 63% of the toxic activity, and 78% of the crystal toxin were in the 44% of the formulation with particle sizes of 21–44 μm . The obvious relationship between spore count, toxic activity, and crystal toxin content makes it possible to use viable spore deposits, which are easily measured, as an indicator of the amounts of dust deposited on grain.

The size range of particles in this formulation is far above the 0.5 μm aerosol particles that Quinlan (1972, 1979, 1980) applied to corn, but because BT is intended for application to only the top 10 cm of the grain mass it seemed appropriate to determine whether the dust would penetrate a few centimeters into the grain. The concentration of the toxic components in the smaller size fractions of the formulation increases the likelihood of sufficient penetration into a grain mass to control insects. Earlier tests with auger and rake in application have shown that uniform incorporation of BT with the surface layer of grain may not be necessary (McGaughey, 1985).

BT deposits in the surface layers of corn treated in the 30.5 cm diameter column using different air flow rates are summarized in Table 2. The mean spore deposits in the 12.5 cm surface layer compared favorably with the mean spore deposits of 52.85×10^4 spores/g in bins of corn treated in an earlier study using auger and

Table 2. Spore deposits ($\times 10^4/g$) in the top five 2.54-cm layers of grain when BT was applied as an airborne dust in a 30.5-cm-diameter column of grain using different air flow rates.^a

Air flow (liters/m ² /min)		Depth ^b					\bar{x}
		1	2	3	4	5	
0	\bar{x}	217.88	8.50	2.05	1.07	0.70	46.04
	SD	104.40	6.44	2.88	1.00	0.54	
	%	94.6	3.7	0.9	0.5	0.3	
457	\bar{x}	190.90	8.34	1.79	2.80	1.31	41.03
	SD	92.50	1.83	0.99	2.24	0.62	
	%	93.1	4.1	0.9	1.4	0.6	
914	\bar{x}	150.87	26.54	17.13	3.07	1.25	39.77
	SD	76.00	13.40	21.62	2.45	0.45	
	%	75.9	13.3	8.6	1.5	0.6	
1524	\bar{x}	197.50	45.43	17.95	4.34	3.58	53.76
	SD	80.55	33.16	22.58	2.14	1.68	
	%	73.5	16.9	6.7	1.6	1.3	
2286	\bar{x}	185.67	35.70	6.31	4.71	3.25	47.13
	SD	54.37	11.92	2.15	0.20	0.88	
	%	78.8	15.2	2.7	2.0	1.4	
3048	\bar{x}	191.77	35.83	5.72	3.82	3.91	48.21
	SD	49.64	21.33	1.07	0.78	1.34	
	%	79.6	14.9	2.4	1.6	1.6	

^a Means of 6 replications without air flow; 3 replications with each air flow rate.

^b Each depth equals a 2.54-cm increment.

rake in application (McGaughey, 1985). With no air flow 95% of the spores were deposited in or on the top 2.54 cm layer of grain. Most of the remaining 5% were deposited in the 2nd layer; <2% reached the 3rd to 5th layers. With an air flow rate of 457 liters/m²/min (1.5 CFM/ft²) penetration was only slightly better, but at higher air flows penetration improved considerably. The best penetration was attained at air flows of 914 and 1524 liters/m²/min (3 and 5 CFM/ft²), with no further improvement at higher rates. At these air flows 25% of the dust penetrated below the surface layer. The proportion of the deposits in each of the subsurface layers increased 4 to 5 fold over the levels with no air flow. Deposits in the 4th and 5th layers were ca. 4×10^4 spores/g.

BT deposits in the surface layers of corn treated in farm bins are summarized in Table 3. Several differences are apparent between these results and those from the small-column tests. The average spore deposits in the layers sampled were only 30–35% of the levels observed in the earlier study (McGaughey, 1985) and in the small-column test (Table 2). Most of the deposit deficit was apparent in the 1st and 2nd layers of the grain. The reason for this was not apparent. Deposits in the 3rd to 5th layers were comparable to those in the small-column tests, ranging from 2 to 6×10^4 spores/g of grain. Undoubtedly some of the dosage was lost through the roof openings of the bins during application. Also, some of the dust may have penetrated deeper than the levels sampled.

The air flow rates in the bins differed widely, but all of them were at or above the optimum rates indicated in the column tests. The differences in dust pene-

Table 3. Spore deposits ($\times 10^4/g$) in the top five 2.54-cm layers of grain when BT was applied as an airborne dust in farm bins equipped with drying fans.^a

Bin no.	Bin diameter (m)	Fan diameter (cm)	Air flow (liters/m ² /min) ^b	Depth ^c					\bar{x}	
				1	2	3	4	5		
1	12.8	45.7 ^d	1402	\bar{x}	40.04	8.89	6.58	6.33	4.99	13.37
				SD	16.41	0.73	0.56	0.86	1.09	
				%	59.9	13.3	9.8	9.5	7.5	
2	11.0	45.7	853	\bar{x}	83.00	5.30	4.58	2.67	1.78	19.47
				SD	43.48	1.95	3.95	2.04	1.36	
				%	85.3	5.4	4.7	2.7	1.8	
3	8.5	30.5	640	\bar{x}	54.67	4.63	6.03	4.37	4.17	14.77
				SD	16.74	0.83	2.65	1.08	0.97	
				%	74.0	6.3	8.2	5.9	5.6	
4	9.1	71.1	3505	\bar{x}	59.30	6.73	5.07	6.50	5.07	16.53
				SD	17.10	1.98	1.79	2.55	2.06	
				%	71.7	8.1	6.1	7.9	6.1	

^a The corn in bins 1-3 contained 1.8% (wt/wt) dockage; corn in bin 4 contained 0.9% dockage.

^b 304.8 liters/m²/min = 1 CFM/ft².

^c Each depth equals a 2.54-cm increment.

^d Bin equipped with 2 fans, each 45.7 cm in diameter.

tration between the bins were attributed more to characteristics of the corn, such as the amount and distribution of fine material and the degree of settling or packing of the grain, than to differences in air flow rates.

Mortality of Indianmeal moths was 100% in the composite samples from the top 10 cm layer of grain in the bins. Infestations of Indianmeal moths during the summer and fall months following treatment were significantly lower than in untreated bins in the same area ($P = 0.05$). The average count of larvae and pupae in corrugated paper strips placed in the bins was 0.38 insects/strip/month, while in untreated bins the average was 5.37, indicating 93% control of infestations.

Based on the extent of dust penetration and the levels of insect control observed in these tests it appears that this method of applying BT to stored corn has sufficient potential to merit further study. Tests should be conducted over a wider range of conditions to more clearly define the results that may be expected in terms of insecticide deposition and insect control in different types of storage facilities with different kinds and sizes of aeration equipment. If proven effective the method should significantly reduce the time and labor required for treating corn that is already in storage.

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