APPLICATION OF BACILLUS THURINGIENSIS THROUGH CENTER-PIVOT IRRIGATION SYSTEMS FOR CONTROL OF SOUTHWESTERN AND EUROPEAN CORN BORER

by

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INTRODUCTION

Current control methods for second generation southwestern corn borer (SWCB), Diatraea grandiosella Dyar, and European corn borer (ECB), Ostrinia nubilalis Hubner, in southcentral Kansas depend on applications of an effective chemical timed to cover the duration of oviposition periods. The oviposition periods may last 3-6 weeks and these periods tend to coincide within a given field. The height of the corn at the time of infestation and the number of chemical treatments necessary to cover the oviposition period create problems currently dealt with only by aerial application. Likewise, proper timing of chemical applications is critical to achieve SWCB and ECB control. Proper timing of aerial applications is difficult because of its dependence on appropriate weather conditions (i.e., temperature, humidity, and wind speed). On those occasions when conditions are suitable, peak demands on the applicator's services often result in delays in application and subsequent loss of optimum chemical efficacy. Center-pivot irrigation systems where in use, may provide a possible alternative to aerial application (Raun 1979). In addition to the advantage of proper application timing, the grower reduces his costs. Diesel engine operated center-pivot application costs are determined by the amount of water applied, depth from which water is pumped, application pressure and cost of diesel fuel. For southcentral Kansas, the combination of these factors require the use of approximately 567 l of diesel fuel to apply 7.6 mm water per unit area to 54.6 ha in a 24 h period. Application costs for a center-pivot, assuming the price of diesel fuel to be 0.26 dollars per l, would be 2.74 dollars per ha compared to 7.41 dollars per ha for aerial application.

The selection of an insecticide for application through center-pivot irrigation systems must be based on several criteria in addition to efficacy for SWCB and ECB. The insecticide must have low mammalian toxicity to allow field entry for irrigation system maintenance. The insecticide should be selective and represent minimum hazard to beneficial arthropods or wildlife. Likewise, it should not cause phytotoxic effects on other plant species because of the high potential for particle drift. The pathogen, *Bacillus thuringiensis* Berlinger, meets these criteria. *B. thuringiensis* is efficacious for both SWCB (Sikorowski and Davis 1970) and ECB (Raun 1963). The pathogen is virtually nontoxic to man and wildlife, exhibits no phytotoxic effects on any plant species, and is selective for arthropod pests (Bond and Boyce 1971, Cooksey 1971, Norris 1971).

Another potential problem with center-pivot application of insecticides involves the washing effect that accompanies applications with large amounts of water. Irrigation systems are designed to put water on the soil surface rather than the plant foliage.

During 1978 and 1979 we conducted studies to: 1) determine the efficacy of *B. thuringiensis* applied through center-pivot sprinkler irrigation systems for SWCB and ECB larvae, 2) determine the effect of carrier rates (liquid output per area) on the efficacy of *B. thuringiensis* for SWCB and ECB control, and 3) delineate potentially detrimental sublethal effects of *B. thuringiensis* on SWCB and ECB populations.

METHODS AND MATERIALS

Center-Pivot Application Study

A commercial preparation of *B. thuringiensis* (Dipel®) was applied to corn (ca. 54.6 ha) using center-pivot sprinkler irrigation systems in southcentral Kansas during 1978 and 1979 for second generation SWCB and ECB larval control. The 1978 study was arranged in a randomized complete block design, 2 blocks each with 2 treatments. Applications, delivering 7.17 billion International Units of potency/ha (IU/ha) of a wettable powder (wp) formulation (16,000 IU/mg), were initiated on July 25, 1978, 3 days before peak SWCB oviposition as predicted by a phenology model (Poston et al. 1978). Three subsequent applications were made at 5 day intervals. Treatment rates were set such that 4 applications of Dipel were equivalent in material costs to 2 applications of carbofuran at 1.12 kg active ingredients/ha. Although applications were not timed directly for ECB oviposition, ECB and SWCB oviposition periods almost coincide in southcentral Kansas (Poston unpublished data). Dipel was introduced into the center-pivot via a single piston injection pump fed by a continuously agitated mix tank. Irrigation was applied as needed. One normal irrigation cycle (one revolution of the center pivot) required 48 h and delivered 16 mm of water per unit area.

During the Dipel application, revolution time was reduced to 24 h delivering 7.6 mm of water per unit area.

Treatment efficacy in 1978 was assessed through infestation counts, tunneling data, girdling counts and yield. Five consecutive plants were sampled from 6 randomly selected locations within each plot on September 9 and 13. Plants were dissected, and live SWCB and ECB larvae and the number of tunnels per plant were recorded. Tunnel lengths were measured to the nearest cm. SWCB girdling was sampled on September 9, 13, 22 and 28 from subplots (15 rows x 30.4 m) located in each plot. Ten consecutive plants in each of the 15 rows were examined for signs of SWCB girdling. The corn was hand harvested on September 13, after physiological maturity.

Applications in 1979 began July 25, 6 days before peak oviposition. Two circles were treated in 1979; field 1 with the same wettable powder formulation used in 1978 and field 2 with a flowable (f) formulation (8.4 million IU/ml). Application procedures used in 1978 were also used in 1979. Infestation counts and tunneling measurements (field 1 only, field 2 harvested) were collected from 8 five-plant samples on September 17-18 and September 21, respectively. SWCB girdling was sampled on September 12, 19, 25 and October 2 in field 1 using the sampling method from 1978. Field 2 was sampled on the same dates except October 2, because the crop was harvested. The corn plots were harvested on September 19 and 21 for fields 1 and 2, respectively, using a mechanical harvester operating over areas averaging 0.18 ha for field 1 and 0.05 ha for field 2.

Carrier Rate Study

A Hagie® high clearance tractor modified to deliver high gallonages was used to simulate center-pivot applications of Dipel in 1979. The study was arranged in a randomized complete block, 4 blocks each with 4 treatments. Plots 4 rows by 15.2 m with 2 border rows between them were located under a center-pivot irrigation system on Sandyland Experimental Field, St. John, Kansas. Dipel (7.17 billion IU/ha) was applied with 7.6, 12.7, or 25.4 mm of water per unit area. Check plots received no treatments. The gradient of carrier rates was chosen such that the lower water application rates of center pivots were represented. Applications (4) were made at 5-day intervals beginning on July 28. The first treatment was applied 4 days before peak SWCB oviposition, predicted by the SWCB phenology model. Infestation counts (SWCB and ECB larvae) taken on September 13 and 14 were sampled from 20 consecutive plants per plot.

Sublethal Effects Study

A range of dosages for this study was obtained from preliminary bioassays conducted on SWCB and ECB neonate larvae. A 10 to 1 serial dilution beginning with 587.3 mg of Dipel wp (17.025 IU/mg), brought to 100 ml, was used to prepare 5 treatments of *B. thuringiensis* (0, 1, 10, 100, 1000 IU/ml). Sections (ca. 0.5 cm diam. x 0.5 cm) of meristematic tissue from whorl stage corn were submerged for 5 minutes, with periodic agitation, in one of the 5 treatments. The sections were then removed and the excess liquid allowed to dry. Individual tissue sections were placed with a single neonate SWCB or ECB larva in a 30-ml plastic cup. The cup was sealed with cellophane and a paper lid to prevent larval desiccation. The larvae were maintained at 30% Rh and 26°C with a 16L:8D photoperiod throughout the study. Larvae were removed from the treated corn after 24 h and placed on untreated corn sections (ca. 2 cm x 0.75 cm diam.) for 4 days, to flush the larva's intestine of any treated materials. The larvae were then placed on an artificial diet (Davis 1976) until pupation. Pupal weights and larval developmental times were recorded for SWCB and ECB males and females. A subsequent study was conducted with some modifications of dosage levels and environmental conditions. Treatment levels were revised to 0, 10, 100, 500 and 1000 IU/ml, and climatic conditions to 60% Rh and 29°C. Larvae were exposed in groups (10) to the treated and untreated corn sections which numbered 3 per cup. Larvae fed on the untreated corn for 3 days before being transferred to an artificial diet. Pupal weights and developmental times were recorded as before. Head capsule widths of the final larval instar were measured for SWCB and ECB larvae. Feces deposited while on the artificial diet were collected, dried, and weighed as an index of SWCB and ECB feeding.

RESULTS

Center-Pivot Application Study

SWCB and ECB infestation levels for the 3 fields treated in 1978 and 1979 are presented in Table 1. The percentage plants infested by SWCB in 1978 and SWCB/ECB in 1978 and 1979 (field 1) were significantly reduced in the Dipel treated plots. Trends toward fewer infested plants were observed for the ECB in 1978, the SWCB and ECB in fields 1 and 2 in 1979, and SWCB/ECB in field 2 in 1979, but no significant control was achieved. Reductions in the number of SWCB per plant seen in 1978 were not observed in 1979. ECB and SWCB/ECB numbers per plant in treated areas were not significantly reduced from those of nontreated areas in the 3 fields. No significant differences between treatments were observed for the numbers of SWCB or ECB larvae per infested plant. Results of the tunneling measurements taken in 1978 and 1979 (Table 2) indicated a significant reduction in mean number of tunnels per plant in 1979 and a similar trend in 1978, although it was not significant. Also of significance was the reduction in mean tunnel length per plant found in 1978 and 1979. The decrease was attributed to the reduction in percentage plants infested by SWCB/ECB (Table 1). No significant differences between treatments were observed in the mean tunnel length per tunnel. No significant reductions in girdling were observed in

Table 1. Percentage plants infested, number per plant and number per infested plant for Southwestern corn borer (SWCB) and European corn borer (ECB) larvae, 1978 and 1979 Center-Pivot Application Study.[a]

		Plants infested (%)			No./plant			No./infested plant		
Year	Treatment	SWCB[b]	ECB	SWCB/ECB	SWCB	ECB	SWCB/ECB	SWCB	ECB	SWCB/ECB
1978	Dipel(wp)	36.6 a	54.9 a	71.6 a	0.41 a	0.85 a	1.26 a	1.13 a	1.54 a	1.76 a
	Check	70.0 b	86.6 a	93.3 b	0.98 b	2.56 a	3.53 a	1.40 a	2.96 a	3.78 a
1979	Dipel(wp) field 1	38.7 a	33.7 a	60.0 a	0.41 a	0.37 a	0.78 a	1.05 a	1.10 a	1.31 a
	Check	55.0 a	51.2 a	82.5 b	0.65 a	1.02 a	1.67 a	1.17 a	1.99 a	2.02 a
1979	Dipel(f) field 2	20.0 a	40.0 a	52.5 a	0.25 a	0.56 a	0.81 a	1.24 a	1.40 a	1.54 a
	Check	33.7 a	60.0 a	67.5 a	0.36 a	0.86 a	1.22 a	1.06 a	1.43 a	1.80 a

[a]Numbers in a column for each year or formulation within a year followed by the same letter are not significantly different ($P > 0.05$).

[b]Significant at ($P < 0.06$).

Table 2. Number of tunnels per plant, tunnel length per plant, tunnel length per tunnel, and yield, 1978 and 1979 Center-Pivot Application Study.[a]

Year	Treatment	No. Tunnels/ Plant	Tunnel length/ Plant (cm)	Tunnel length/ Tunnel (cm)	Yield kg/ha
1978	Dipel	1.2 a	9.7 a	5.25 a	8340 a
	Check	2.4 a	21.0 b	8.46 a	8325 a
1979	Dipel field 1	1.2 a	13.5 a	8.95 a	8562 a
	Check	2.7 b	23.2 b	7.88 a	8519 a

[a]Numbers in a column within the same year followed by the same letter are not significantly different ($P > 0.05$).

the fields in 1978 or 1979 except on the last sample date (September 25, 1979) in field 2 (7.6% treated versus 18.3% nontreated).

Carrier Rate Study

Results of the carrier rate study are presented in Table 3. The application of Dipel with 7.6, 12.7 or 25.4 mm of water per unit area significantly reduced the percentage of infested plants and number of larvae per plant of ECB and SWCB/ECB compared to the check. The percentage of plants infested with SWCB and the number of SWCB per plant was significantly reduced only at the 12.7 mm rate. No reductions were found in the number of SWCB per infested plant. Significantly fewer ECB per infested plant were observed in the treatments receiving 12.7 and 25.4 mm water per unit area. Numbers of SWCB/ECB per infested plant were significantly reduced over all carrier rate treatments compared to the check.

Sublethal Effects Study

SWCB larvae exposed to 100 IU/ml of *B. thuringiensis* in study 1 had significantly slower developmental rates than larvae treated with 0 or 1 IU/ml (Table 4). In study 2, SWCB larvae treated with 100 or 500 IU/ml had significantly ($P < 0.06$) longer developmental times than those receiving no treatment. No differences in time spent in the larval stadia could be found for the ECB. Larval mortality for SWCB and ECB in study 1 were not found to be different for dosage levels 0, 1, and 10 IU/ml, but these 3

Table 3. Percentage plants infested, number per plant and number per infested plant for Southwestern corn borer (SWCB) and European corn borer (ECB) larvae, 1979 Carrier Rate Study.[a]

	Plants infested (%)			No./plant			No./infested plant		
Treatment (mm)	SWCB	ECB	SWCB/ECB	SWCB	ECB	SWCB/ECB	SWCB	ECB	SWCB/ECB
7.6	16.2 ab	31.2 a	42.5 a	0.16 ab	0.43 a	0.58 a	1.0 a	1.35 ab	1.37 a
12.7	6.2 a	40.0 a	42.5 a	0.06 a	0.49 a	0.55 a	1.0 a	1.21 a	1.29 a
25.4	13.7 ab	32.5 a	43.7 a	0.14 ab	0.39 a	0.52 a	1.0 a	1.18 a	1.19 a
No Treatment	25.0 b	76.2 b	82.5 b	0.24 b	1.25 b	1.48 b	1.0 a	1.63 b	1.79 b

[a]Numbers in a column followed by the same letter are not significantly different ($P > 0.05$).

Table 4. Days in larval stadia and percentage mortality for Southwestern corn borer (SWCB) and European corn borer (ECB), Sublethal Effects Studies 1 and 2.

	Treatment (IU/ml)	Days in larval stadia SWCB	ECB	Mortality (%) SWCB	ECB
Study 1	0	21.4 b	19.1 a	15.0 a	17.5 a
	1	21.6 b	18.9 a	5.0 a	7.5 a
	10	22.1 ab	19.4 a	7.5 a	9.0 a
	100	23.1 a	21.0 a	35.0 b	46.5 b
	1000	22.1 ab	23.4 a	65.0 c	94.0 c
Study 2[b]	0	18.1 b	16.9 a	21.7 ab	28.0 a
	10	18.5 ab	17.9 a	8.4 a	36.0 a
	100	19.0 a	17.7 a	33.4 b	74.0 b
	500	19.0 a	20.0 a	68.4 c	98.0 c
	1000	18.7 ab	--	80.0 c	100.0 c

[a]Numbers in a column within a study followed by the same letter are not significantly different ($P > 0.05$).

[b]Environmental conditions adjusted to 29°C and 60% Rh for study 2; study 1, 26°C and 30% Rh.

levels had significantly less mortality than larvae exposed to the 100 or 1000 IU/ml. SWCB or ECB larvae treated with 1000 IU/ml had significantly greater mortality than those exposed to 100 IU/ml. In study 2 mortality for SWCB larvae exposed to 10 IU/ml and ECB larvae exposed to 0 and 10 IU/ml was significantly less than the mortality of larvae exposed to 100, 500 or 1000 IU/ml. No significant differences in SWCB or ECB mortality were observed between the 500 and 1000 IU/ml dosage levels. However, the mortality observed in the 2 levels for both SWCB and ECB was significantly greater than that found in the 0, 10, or 100 IU/ml levels. No significant differences between dosages were observed for larval head capsule widths, fecal weight, or pupal weight.

DISCUSSION

The use of Dipel through center-pivot irrigation systems for SWCB and ECB control is impractical. Increasing efficacy via higher rates or more frequent applications was not economically feasible. Increasing the concentration by decreasing the amount of water applied was not possible either. The carrier rate study demonstrated that Dipel's efficacy did not increase as the carrier rate decreased within a range of practical rates for center-pivot systems.

Sublethal effects on feeding and girdling behavior were not observed in the laboratory study or field studies except for field 2 in 1979. Logically the girdling reduction observed in field 2 could be accounted for in one of two ways. A decrease in the percentage of SWCB infested plants because of increased larval mortality from the Dipel treatment would cause a reduction in girdling. However, no significant reduction in SWCB infestation was detected. The second possibility would be a sublethal response such that treated larvae girdled less. In this case one should observe a reduction in the percentage girdled plants per percentage SWCB infested plants in treated versus nontreated areas. Analysis of the data indicated no significant ($P > 0.05$) differences between treatments for this variable. Thus we attribute the girdling reduction observed in field 2 in 1979 to fortuitous error. Any benefits derived from sublethal concentrations of *B. thuringiensis* encountered by larvae in the field is minimal. The slower developmental times for the SWCB are not of great enough magnitude to be detected or beneficial in a field situation.

The use of a center-pivot irrigation system for the application of *B. thuringiensis* has many desirable attributes. However, considering the cost of the pathogen, the low efficacy attained and the problem with excessive water applications, this method does not appear to be feasible.

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Center-pivot sprinkler irrigation systems were used during 1978 and 1979 to apply Bacillus thuringiensis Berlinger for second generation control of Diatraea grandiosella Dyar and Ostrinia nubilalis Hubner. Although a reduction in larval density and subsequent damage for both species was attained, the level of suppression was insufficient to warrant commercial use. Reduction of irrigation rates to the minimum attainable with commercial sprinkler systems provided no increase in borer control. Larval development time for D. grandiosella was increased from exposure to selected concentrations of B. thuringiensis in laboratory studies. This change, however, was of no measurable benefit to corn production in field situations. Although B. thuringiensis has many desirable attributes (low mammalian toxicity, etc.) for suppression of D. grandiosella and O. nubilalis populations when applied through center-pivot irrigation systems, the cost of the pathogen and its low relative efficacy in large quantities of water make it impractical.