

Article

Timely Application of Four Insecticides to Control Corn Earworm and Fall Armyworm Larvae in Sweet Corn

Diego M. Viteri ^{1,*}  and Angela M. Linares-Ramírez ² 

¹ Department of Agro-Environmental Sciences, University of Puerto Rico, Isabela Research Substation, 2090 Ave. Militar, Isabela, PR 00662, USA

² Department of Agro-Environmental Sciences, University of Puerto Rico, Lajas Research Substation, Carr. 101 km 8.5 Barrio Palmarejo, Lajas, PR 00667, USA; angela.linares@upr.edu

* Correspondence: diego.viteri@upr.edu

Simple Summary: Corn earworm and fall armyworm are the most important Lepidopteran pests in sweet corn worldwide. Timely application of insecticides during pollination is crucial to control both species and produce kernels with higher quality. Our results showed that applications of insecticides should be conducted at 48 h after pollination. Emamectin benzoate and chlorantraniliprole provided an adequate control of fall armyworm and corn earworm larvae. Furthermore, emamectin benzoate should be applied to larvae in later instars (fourth–sixth) while chlorantraniliprole and spinetoram should be sprayed when infestations of larvae in earlier instars (first–third) are observed.

Abstract: Insecticide sprays are a common practice to control corn earworm, *Helicoverpa zea* (Boddie), and fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae), in corn (*Zea mays* L.) at reproductive stages. Our objectives were to determine (1) the most appropriate time for insecticide applications and (2) the effect of four insecticides on the survival of larvae as well as their weight. β -cyfluthrin (0.4 mL/L), chlorantraniliprole (0.6 mL/L), emamectin benzoate (0.2 g/L), and spinetoram (1.5 mL/L) were sprayed on silks of sweet corn planted in Isabela and Lajas, Puerto Rico 3 h before and 24 and 48 h after pollination. The number of kernels produced and the damage of larvae on kernels were quantified at harvest. In addition, percentages of mortality and changes on larval weight were noted at 96 h after insecticide applications. Insecticide sprays at 3 h before pollination reduced the number of kernels or were similar to the control in all treatments. However, emamectin benzoate sprayed in Lajas and chlorantraniliprole applied in Isabela at 48 h after pollination increased the number of kernels (281–294) and reduced the damage of larvae on kernels (<0.5%) compared to the control (201–229; >7%). Furthermore, applications of emamectin benzoate caused higher percentages of fall armyworm larval mortality (>70%). Conversely, β -cyfluthrin and chlorantraniliprole caused lower percentages of mortality (<30%) and only chlorantraniliprole and spinetoram reduced the weight of corn earworm and fall armyworm larvae collected in both locations. This information may help pest management programs and corn breeders to schedule insecticide sprays and pollination in the field.

Keywords: damage on kernels; insecticides; larvae; percentage of mortality; weight



Citation: Viteri, D.M.; Linares-Ramírez, A.M. Timely Application of Four Insecticides to Control Corn Earworm and Fall Armyworm Larvae in Sweet Corn. *Insects* **2022**, *13*, 278. <https://doi.org/10.3390/insects13030278>

Academic Editors: Andrew G. S. Cuthbertson and Lukasz L. Stelinski

Received: 3 February 2022

Accepted: 8 March 2022

Published: 11 March 2022

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

Corn earworm, *Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae), and fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae), are the most important pests affecting field/sweet corn (*Zea mays* L.) in the tropics, the United States, and worldwide [1–6]. Larvae of fall armyworm feed on leaves, tassels, and kernels [7] while corn earworm larvae are mostly observed feeding on kernels or cob tissue at reproductive stages [4]. Later larval instars (fourth to sixth) cause higher damage in the plant tissue, and yield losses over 18% have been reported for both species [7–9].

Puerto Rico is an important winter nursery location mostly for field corn with more than 1000 ha planted in each season. In fact, private companies and universities from North America use Puerto Rico to advance generations, and for genetics and breeding studies [10]. The tropical conditions of Puerto Rico, multiple plantings, and the presence of other hosts (e.g., soybean, rice, and vegetables) allow the development of multiple generations (>10) of both pest species during the year [11]. Furthermore, resistance of corn earworm and fall armyworm larvae to different insecticides including *Bacillus thuringiensis*, carbamates, and pyrethroids [2,10,12–14] were reported. All of these factors have caused biological, chemical, cultural, and mechanical controls plus fallow periods to be necessary to combat and decrease the populations of both species in each season.

Chemical control in combination with biological insecticides is the most common strategy to control Lepidopteran species in Puerto Rico [10,11]. In fact, some insecticides (e.g., β -cyfluthrin and spinetoram) have been used widely in direct applications to cobs after pollination to avoid the damage of corn earworm and fall armyworm larvae and other pests (e.g., corn sap beetle, *Carpophilus* spp. (Coleoptera: Nitidulidae); and silk fly *Euxesta* spp. (Diptera: Ulidiidae)) in the kernels (Viteri D, personal communication). Likewise, the efficacy of chlorantraniliprole, methoxyfenozide, and spinosyn to control corn earworm and fall armyworm larvae at reproductive stages by the reduction of the damage in the ears/kernels of sweet corn were reported in experimental trials in Florida and New York [15–17]. However, there are no previous studies, to the best of our knowledge, related to the appropriate time when the insecticides need to be sprayed (before or after pollination) to avoid any damage in the fertilization and kernel yields.

The intensive insecticide sprays (i.e., over 20 applications per season [2]) and the higher larvae pressure of both species in field/sweet corn in Puerto Rico have been important issues for pest management programs who want to avoid or delay the resistance to insecticides, and corn breeders who need to obtain higher numbers of viable seeds in their crosses or seed increases. In fact, an adequate synchronization at the time of pollination and insecticide sprays are necessary to meet the desired production of kernels. For this, insecticide rotations with different modes of action and short reentry periods (≤ 12 h) (e.g., chlorantraniliprole, emamectin benzoate, and spinetoram) should be used at the peak of pollinations. The objectives of this study were to determine (1) the most appropriate time for insecticide applications and (2) the effect of four insecticides on corn earworm and fall armyworm larval survival, as well as their weight.

2. Materials and Methods

2.1. Field Experiments

Eight experimental trials with sweet corn ‘Suresweet 2011’ were planted in Isabela and Lajas Research Substations at the University of Puerto Rico in September 2020 for emamectin benzoate treatment, and February, March, and April 2021 for spinetoram, chlorantraniliprole, and β -cyfluthrin treatments, respectively. The Isabela substation is located at latitude 18°30′00″ N, longitude 67°00′00″ W, and at 126 m above sea level. The temperature varies from 20 to 31 °C with a mean annual precipitation of 1592 mm, and 70% relative humidity. The soil at the Isabela Substation belongs to the Oxisol type (Coto series) [18]. The Lajas substation is located at 18°03′ 00″ N, 67°03′34″ W at 9 m above the sea level. Lajas has soils belonging to the Mollisol and Vertisol orders, and has temperatures between 19 to 33 °C, an annual precipitation of 1143 mm, and an average relative humidity of 80% [18].

Experimental plots were 25 × 15 m in size. One sweet corn seed was planted every 0.30 m in each row (25 m length) and the separation between rows was 0.76 m. Each treatment was placed in every 4 rows. Plants with cobs at R1 stage, without visible silk hairs, were covered with shoot bags to avoid their exposure to the pollen. Then, 5 mL of each insecticide solution was sprayed at higher dosages (Table 1) to four-day-old silk hairs, 3 h before pollination and 24 and 48 h after pollinations by the syringe Ideal® Prima® backpack (Neogen, Lansing, MI, USA). A total volume of 150 mL of insecticide solution

was used per treatment at each time of pollination. The control was not sprayed. Sixty milligrams of fresh pollen (50–55% of moisture content) per silk was used for pollinations and tassel bags replaced the shoot bags when plants were pollinated. A randomized complete block design with three replications was used in each location, and ten cobs were used per each insecticide treatment and the control per replication ($n = 120$ per location). Cobs were harvested 21 days after the pollination. The number of kernels produced and the percentage of corn earworm and fall armyworm damage on the kernels under natural infestation were recorded.

Table 1. Active ingredients and their percentages, insecticide group, commercial names, manufacturers and dosages of four insecticides used in field applications and bioassays to control corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) larvae in 2019 through 2021.

Insecticides	Major Group or Subgroup	Commercial Name	Manufacturers	Dosage (g or mL/L)
Active ingredient and percentage				
β -cyfluthrin 12.70%	3 A (pyrethroid)	Baythroid [®] XL	Bayer	0.40 mL
Chlorantraniliprole 18.40%	28 (diamide)	Coragen [®]	Dupont	0.60 mL
Emamectin benzoate 2.12%	6 (avermectin)	Proclaim [®] 05SG	Syngenta	0.20 g
Spinetoram 11.70%	5 (spinosyn)	Radiant [®] SC	Dow AgroSciences	1.50 mL

2.2. Bioassays

Larvae were collected from leaves and ears of sweet corn ‘Suresweet 2011’ planted in Isabela and Lajas Research Substations at the University of Puerto Rico in June 2019 for corn earworm and June 2020 for fall armyworm. The weight of each larva was recorded, and then one fourth or fifth instar larva of corn earworm or fall armyworm was placed separately in a 20 mL plastic cup (Lion, Santo Domingo, Dominican Republic) containing soybean–wheat germ-based artificial diet (Frontier Scientific Services Inc., Newark, DE, USA). Fifteen larvae of each species per replication were treated topically with 3 mL (200 μ L per larva) insecticide solution of β -cyfluthrin, chlorantraniliprole, emamectin benzoate, and spinetoram per replication at the dosages described in Table 1. A micropipette with a 200 μ L Eppendorf tip was used to deliver the insecticide solution over the head, thorax, and abdomen of each larva. The control was sprayed with 200 μ L of distilled water per larva. Treated cups and the control were held in a randomized complete block design with 4 replications (total $n = 60$ per location and species) in the laboratory at 18 to 20 °C, and a photoperiod of 12:12 h (L:D). The percentage of mortality and the weight of each larva were evaluated at 96 h after the insecticide application.

2.3. Data Analysis

A combined data analysis [19] was performed to determine the effect of the location and the interactions location \times time for pollination and location \times larval weight per each insecticide. A fixed model was used for data analysis where the two locations, time of pollination, and larval weight were fixed effects and the replication was a random effect [19]. In addition, simple analysis of variance for the number of kernels produced, the percentage of corn earworm and fall armyworm damage on kernels, and the larval weight were conducted in each location. Similar criterion was used for the percentage of larval mortality in the bioassays. However, the four insecticide treatments (also a fixed effect) were analyzed together in the combined and simple analysis. Data were analyzed using SAS 9.4 PROC GLIMMIX [20] and a TUKEY HRS test was conducted to discriminate differences among treatments versus the control for each variable. In addition, Abbott’s formula [21] was used to correct larval mortality attributed to the insecticide effect in the bioassays.

3. Results

Significant differences ($p \leq 0.05$) between the time of application were observed for all insecticides in the number of kernels produced (Table 2). The interaction location \times time of application showed significant differences for β -cyfluthrin, chlorantraniliprole, and spinetoram for the same variable. On the contrary, significant differences for the time of application and its interaction with the location were only observed for chlorantraniliprole and emamectin benzoate for the variable damage on kernels (Table 2). The simple analysis of variance also showed differences between time of application for all insecticide treatments in the number of kernels produced in both locations (Table 3). Emamectin benzoate showed differences for the time of application in Isabela and Lajas for the damage on kernels (Table 3).

Table 2. Combined analysis of variance for the number of kernels produced and percentages of damage of corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) per each insecticide in sweet corn (*Zea mays* L.) ‘Suresweet 2011’ planted at Isabela and Lajas, Puerto Rico in 2020 and 2021.

Effect	df	Number of Kernels Produced				Damage on Kernels			
		β -cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram	β -cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram
		<i>F</i> values							
Location (L)	1, 2	43.97 *	5.18	12.71	4.58	3.38	3.49	2.37	6.24
Time of application (T)	3, 228	39.92 ***	27.09 ***	7.18 ***	11.64 ***	0.89	10.15 ***	3.46 *	3.67 *
L \times T	3, 228	30.04 ***	9.56 ***	1.08	18.88 ***	2.13	10.15 ***	4.00 *	0.92

* Significant at $p \leq 0.05$, and *** $p \leq 0.001$.

Table 3. Simple analysis of variance for the number of kernels produced and percentages of damage of corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) per each insecticide in sweet corn (*Zea mays* L.) ‘Suresweet 2011’ planted at Isabela and Lajas, Puerto Rico in 2020 and 2021.

Variable	Effect	df	Isabela				Lajas			
			β -cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram	β -cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram
			<i>F</i> values							
Number of kernels produced	Time of application	3, 114	100.67 ***	22.04 ***	4.11 *	11.17 ***	10.44 ***	6.62 ***	4.10 *	20.68 ***
	Time of application	3, 114	3.30 *	10.15 ***	2.83 *	2.12	1.38	_ ^a	3.81 *	4.23 *

* Significant at $p \leq 0.05$, and *** $p \leq 0.001$. ^a There was no infestation of corn earworm and fall armyworm larvae. Thus, statistical analyses were not conducted.

The combined analysis for the percentage of mortality had significant differences between the location and location \times treatment interaction for corn earworm and fall armyworm (Table 4). Similar results were observed for the simple analysis of variance for corn earworm ($F = 6.68$; $df = 4, 12$; $p \leq 0.005$ and $F = 79.47$; $df = 4, 12$; $p \leq 0.0001$) and fall armyworm ($F = 45.64$; $df = 4, 12$; $p \leq 0.0001$ and $F = 154.49$; $df = 4, 12$; $p \leq 0.0001$) in Isabela and Lajas, respectively. In addition, significant differences were observed between larval weight and location \times larval weight interaction for β -cyfluthrin for corn earworm (Table 5). Conversely, larval weight and location \times larval weight showed significant differences for the control, chlorantraniliprole, and spinetoram treatments for fall armyworm (Table 5). Furthermore, significant differences were noted for chlorantraniliprole and spinetoram in the simple analysis of variance carried out for both species in Isabela and Lajas (Table 6).

Table 4. Combined analysis of variance for the percentages of mortality of corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) larvae evaluated at 96 h after insecticide application in 2019 and 2020.

Effect	df	Corn Earworm	Fall Armyworm
F values			
Location (L)	1, 3	20.11 *	9.29
Treatment (T)	4, 24	47.07 ***	157.58 ***
L × T	4, 24	12.69 ***	12.71 ***

* Significant at $p \leq 0.05$, and *** $p \leq 0.001$.

Table 5. Combined analysis of variance for the location and weight of corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) larvae evaluated at 96 h after insecticide application in 2019 and 2020.

Effect	df	Corn Earworm					Fall Armyworm				
		No insecticide	β-cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram	No insecticide	β-cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram
F values											
Location (L)	1, 3	6.57	0.74	28.05 *	17.27 *	10.09	169.52 **	27.82 *	190.54 ***	20.08 *	511.86 ***
Larval weight (W)	1, 230	300.47 ***	18.36 ***	15.11 ***	0.01	16.37 ***	7.75 **	2.01	74.40 ***	1.63	81.94 ***
L × W	1, 230	0.08	8.10 **	0.86	0.46	0.10	9.58 **	9.05 **	28.09 ***	0.60	43.81 ***

* Significant at $p \leq 0.05$, ** $p \leq 0.01$, and *** $p \leq 0.001$.

Table 6. Simple analysis of variance for the weight of corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) larvae evaluated at 96 h after insecticide application in 2019 and 2020.

Effect	df	Location	Corn Earworm					Fall Armyworm				
			No insecticide	β-cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram	No insecticide	β-cyfluthrin	Chlorantraniliprole	Emamectin benzoate	Spinetoram
F values												
Larval weight	1, 115	Isabela	100.44 ***	0.90	3.84 *	0.15	5.13 *	0.04	1.05	63.09 ***	1.65	80.56 ***
	1, 115	Lajas	71.80 ***	30.03 ***	13.40 ***	0.17	14.83 ***	23.45 ***	12.40 ***	11.94 ***	0.17	6.22 **

* Significant at $p \leq 0.05$, ** $p \leq 0.01$, and *** $p \leq 0.001$.

In general, insecticide applications at 3 h before pollination or 24 h after pollination reduced the number of kernels produced or were similar to the control, especially in lower corn earworm and fall armyworm infestations (Table 7). In contrast, β-cyfluthrin and emamectin benzoate sprayed at 48 h after pollination increased the number of kernels (means of 268 and 281, respectively). However, only the applications of emamectin benzoate reduced the damage of corn earworm and fall armyworm larvae in the kernels (<0.5%) compared to the control (229; 7.3%) in Lajas. Likewise, applications of chlorantraniliprole at the same time increased the yield (294) and produced kernels without damage compared to the control (201; 9%) in Isabela.

The applications of emamectin benzoate caused 80 and 94% of larval mortality from fall armyworm populations collected in Isabela and Lajas, respectively. However, corn earworm larvae collected in Isabela had lower percentages of mortality (36%) to this insecticide. Spinetoram caused between 53 to 70% of larval mortality in Lajas and 30 to 36% in Isabela for both species. Conversely, applications of β-cyfluthrin and chlorantraniliprole

caused percentages of mortality below 27% in both populations (Tables 8 and 9). Corn earworm and fall armyworm larvae treated with chlorantraniliprole and spinetoram had a significant weight reduction in both locations (Tables 8 and 9). In contrast, there were not significant differences in the weight in larvae of both populations applied with β -cyfluthrin and emamectin benzoate, while corn earworm and fall armyworm larvae treated with water had similar or higher weight at 96 h after insecticide application in both locations as expected (Tables 8 and 9).

Table 7. Mean (\pm SE) of the number of kernels produced at different time of insecticide applications and percentages of damage of corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) and fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) larvae in kernels of sweet corn (*Zea mays* L.) ‘Suresweet 2011’ planted at Isabela and Lajas, Puerto Rico in 2020 and 2021.

Treatment ⁺	Time of Application	Isabela		Lajas	
		Number of Kernels	Damage on Kernels %	Number of Kernels	Damage on Kernels %
β -cyfluthrin	Control (no insecticide)	264 \pm 7.06 a	0.80 \pm 0.34 a	190 \pm 12.09 a	3.60 \pm 1.62 a
	3 h before pollination	86 \pm 8.68 b	2.10 \pm 0.73 b	202 \pm 9.54 a	2.20 \pm 1.13 a
	24 h after pollination	140 \pm 9.91 b	0.50 \pm 0.13 a	257 \pm 13.09 b	3.20 \pm 1.21 a
	48 h after pollination	234 \pm 6.84 b	0.70 \pm 0.21 a	268 \pm 13.55 b	6.30 \pm 2.05 a
Chlorantraniliprole	Control (no insecticide)	201 \pm 22.90 a	8.60 \pm 2.72 a	151 \pm 13.83 a	0.00 \pm 0.00 a
	3 h before pollination	50 \pm 7.85 b	0.10 \pm 0.06 b	94 \pm 8.37 b	0.00 \pm 0.00 a
	24 h after pollination	159 \pm 25.57 b	0.00 \pm 0.00 b	157 \pm 12.28 a	0.00 \pm 0.00 a
	48 h after pollination	294 \pm 25.27 b	0.00 \pm 0.00 b	159 \pm 13.02 a	0.00 \pm 0.00 a
Emamectin benzoate	Control (no insecticide)	219 \pm 12.65 a	0.40 \pm 0.14 a	229 \pm 17.59 a	7.30 \pm 3.21 a
	3 h before pollination	169 \pm 14.44 b	1.70 \pm 0.95 a	214 \pm 12.98 a	1.00 \pm 0.54 b
	24 h after pollination	202 \pm 12.79 a	0.00 \pm 0.00 a	228 \pm 13.38 a	1.80 \pm 0.61 b
	48 h after pollination	226 \pm 9.25 a	0.00 \pm 0.00 a	281 \pm 13.78 b	0.40 \pm 0.19 b
Spinetoram	Control (no insecticide)	197 \pm 23.12 a	8.78 \pm 2.47 a	232 \pm 10.98 a	0.91 \pm 0.65 a
	3 h before pollination	75 \pm 22.96 b	2.82 \pm 2.18 a	93 \pm 10.62 b	0.30 \pm 0.27 a
	24 h after pollination	199 \pm 12.12 a	4.50 \pm 5.65 a	211 \pm 14.41 a	0.00 \pm 0.00 b
	48 h after pollination	217 \pm 17.68 b	2.52 \pm 1.32 a	224 \pm 15.34 a	0.00 \pm 0.00 b

⁺ TUKEY HRS test was conducted separately for each insecticide per location. Different letters denote significant differences at $p \leq 0.05$ for the number of kernels produced and damage on kernels between the time of each insecticide application versus the control.

Table 8. Mean (\pm SE) of percentages of mortality for corn earworm (*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)) larvae and their initial and final weight evaluated at 96 h after insecticide application in 2019.

Treatment	% of Mortality ⁺	Isabela		Lajas		
		Initial Weight (g)	Final Weight (g)	% of Mortality	Initial Weight (g)	Final Weight (g)
Control	0.00 \pm 0.00 a	0.22 \pm 0.01 a	0.42 \pm 0.02 b	0.00 \pm 0.00 a	0.18 \pm 0.01 a	0.38 \pm 0.01 b
β -cyfluthrin	26.52 \pm 8.08 b	0.25 \pm 0.01 a	0.27 \pm 0.02 a	21.33 \pm 6.01 b	0.18 \pm 0.01 a	0.30 \pm 0.02 b
Chlorantraniliprole	10.24 \pm 6.43 a	0.22 \pm 0.01 a	0.18 \pm 0.01 b	16.67 \pm 5.89 b	0.17 \pm 0.01 a	0.12 \pm 0.01 b
Emamectin benzoate	32.74 \pm 7.68 b	0.28 \pm 0.02 a	0.27 \pm 0.02 a	91.90 \pm 3.00 b	0.20 \pm 0.01 a	0.21 \pm 0.01 a
Spinetoram	35.92 \pm 3.42 b	0.21 \pm 0.01 a	0.17 \pm 0.01 b	53.77 \pm 3.70 b	0.18 \pm 0.01 a	0.13 \pm 0.01 b

⁺ Different letters denote significant differences at $p \leq 0.05$ between the percentages of mortality of larvae treated with each insecticide versus the control according to the TUKEY HRS test. Different letters denote significant differences at $p \leq 0.05$ between the initial and the final weight according to the TUKEY HRS test.

Table 9. Mean (\pm SE) of percentages of mortality for fall armyworm (*Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae)) larvae and their initial and final weight evaluated at 96 h after insecticide application in 2020.

Treatment	Isabela			Lajas		
	% of Mortality ⁺	Initial Weight (g)	Final Weight (g)	% of Mortality	Initial Weight (g)	Final Weight (g)
Control	0.00 \pm 0.00 a	0.29 \pm 0.01 a	0.29 \pm 0.02 a	0.00 \pm 0.00 a	0.09 \pm 0.01 a	0.16 \pm 0.01 b
β -cyfluthrin	8.70 \pm 4.15 a	0.26 \pm 0.01 a	0.24 \pm 0.02 a	5.77 \pm 3.68 a	0.12 \pm 0.01 a	0.18 \pm 0.02 b
Chlorantraniliprole	22.14 \pm 7.51 b	0.30 \pm 0.01 a	0.20 \pm 0.06 b	9.62 \pm 5.77 a	0.11 \pm 0.01 a	0.08 \pm 0.005 b
Emamectin benzoate	79.80 \pm 4.31 b	0.22 \pm 0.01 a	0.20 \pm 0.01 a	94.24 \pm 3.68 b	0.10 \pm 0.01 a	0.09 \pm 0.01 a
Spinetoram	30.00 \pm 3.74 b	0.29 \pm 0.01 a	0.18 \pm 0.01 b	70.36 \pm 0.36 b	0.09 \pm 0.004 a	0.07 \pm 0.01 b

⁺ Different letters denote significant differences at $p \leq 0.05$ between the percentages of mortality of larvae treated with each insecticide versus the control according to the TUKEY HRS test. Different letters denote significant differences at $p \leq 0.05$ between the initial and the final weight according to the TUKEY HRS test.

4. Discussion

Our data suggest that safe insecticide sprays on silks should be conducted at 48 h or longer after pollination. Applications of commercial dosages of β -cyfluthrin and chlorantraniliprole at 3 h before pollination and 24 h after pollination reduced the number of kernels produced > 20% in Isabela. Likewise, applications of emamectin benzoate and spinetoram in Isabela and chlorantraniliprole and spinetoram in Lajas at 3 h before pollination reduced the seed yield from 23 to 60%. Apparently, the water or the insecticide solution applied on the silks may affect the pollen germination, female receptivity or the syngamy process which occurs in the next 28 h after pollination [22,23]. In fact, the extracellular surface of corn silks has lipids with non-polar molecules [24]. This hydrophobic property of the silks caused bubbles of water to form on the surface when insecticides were sprayed (especially at earlier insecticide applications). The residues of water on the silk hairs/trichomes may kill some pollen grains, avoiding proper fertilization. In addition, corn silks provide pollen hydration for its germination when pollen is in contact with the stigma [23], and the water or insecticide solutions may also affect these biological procedures. Further research is necessary to estimate the time required for the water evaporation on the silks or if the insecticide residues in the corn plant tissue cause a detrimental effect on the pollen–pistil interactions. Residues of acephate, acetamiprid, chromafenozide, and etofenprox were reported on silks and husks of sweet corn [25]. Furthermore, the residues of the four insecticides used in this study vary from 5 to 11 days in the plant tissue [26–29]. However, our data suggest that residues of insecticide solutions on silks up to 24 h may cause a negative effect in the pollen, fertilization, and/or kernel formation. In vitro studies should be carried out with pollen germination media [30] amended with β -cyfluthrin, chlorantraniliprole, emamectin benzoate, and spinetoram at different dosages to verify if these molecules cause any damage in the pollen. It has been reported that corn pollen germination and tube elongation were inhibited or reduced by the combination of trichlorfon with adjuvants [31].

Chlorantraniliprole was the most effective treatment in the field in Isabela at 48 after pollination. Likewise, spinetoram reduced the damage of Lepidopteran species on kernels at the same period although there were not statistical differences compared with the control. The direct application of both insecticides on the corn earworm and fall armyworm larvae caused lower percentages of mortality. However, there was a marked effect in the weight reduction in the larvae treated with these active ingredients. It has been reported that diamides cause an impaired regulation of muscle contraction [32] while spinosyns produce larvae paralysis attributed to fatigue in the nervous system [33]. Future studies are necessary to determine the rate of larvae feeding and their relationship with the damage in kernels from larvae treated with chlorantraniliprole and spinetoram. It is important to mention that larvae in earlier instars (first–third) of corn earworm (80%) and fall armyworm (20%) were observed on silks at the time of applications. Thus, these stages probably were

more susceptible to chlorantraniliprole and spinetoram and/or their indirect effect in the weight reduction compared to the larvae in fifth instar that were used in the bioassays.

Emamectin benzoate sprayed at 48 after pollination was effective to control Lepidopteran species in Lajas where the rate of corn earworm and fall armyworm populations in cobs reached up to 1:1, as reported previously [10]. In fact, our data showed that both larvae species treated with emamectin benzoate had higher percentages of mortality in Lajas. On the contrary, corn earworm populations from Isabela did have lower percentages of mortality, and this may be associated with the prolonged use of this pesticide to control corn earworm in other crops (e.g., common bean), compared to Lajas where most of the applications are conducted with biological insecticides. In addition, the percentages of mortality reached 55% when corn earworm larvae in the fourth instar were treated with emamectin benzoate in Isabela (Viteri D, unpublished). Sarmiento et al. (2022) [34] also reported percentages of corn earworm mortality between 50 to 75% of populations from Isabela, Lajas, and Juana Díaz in previous years.

In summary, insecticides should be applied at least 48 h after pollination to control severe infestations of fall armyworm and corn earworm larvae on cobs/kernels of sweet corn. Emamectin benzoate provided an adequate control for both Lepidopteran species. If a second application is required or under infestations of earlier larvae instars (first–third), spinetoram or chlorantraniliprole could be used in rotation. Furthermore, the evaluation of synthetic insecticides in combination with biological control agents [14,35] is necessary in targeted applications on silks and cobs.

Author Contributions: D.M.V.: Supervision, conceptualization, data collecting, methodology, investigation, project administration, review and editing, validation, writing—original draft; A.M.L.-R.: Data collecting, methodology, supervision, resources, review and editing. All authors have read and agreed to the published version of the manuscript.

Funding: This project was supported by the USDA-NIFA-HATCH (Project # 1012972) funds.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: The authors thank the personnel of Isabela and Lajas Research Substations for their support in the field and bioassays.

Conflicts of Interest: The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

References

- Deshmukh, S.; Pavithral, H.B.; Kallelshwaraswamy, C.M.; Shivanna, B.K.; Maruthi, M.S.; Mota-Sanchez, D. Field efficacy of insecticides for management of invasive fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) on maize in India. *Fla. Entomol.* **2020**, *103*, 221–227. [[CrossRef](#)]
- Gutiérrez-Moreno, R.; Mota-Sanchez, D.; Blanco, C.A.; Whalon, M.E.; Terán-Santofimio, H.; Rodríguez-Maciél, J.C.; DiFonzo, C. Field-evolved resistance of the fall armyworm (Lepidoptera: Noctuidae) to synthetic insecticides in Puerto Rico and Mexico. *J. Econ. Entomol.* **2019**, *112*, 792–802. [[CrossRef](#)] [[PubMed](#)]
- Moore, V.; Tracy, W. Survey of organic sweet corn growers identifies corn earworm prevalence, management and opportunities for plant breeding. *Renew. Agric. Food Syst.* **2021**, *36*, 126–129. [[CrossRef](#)]
- Reay-Jones, F.P.F. Pest status and management of corn earworm (Lepidoptera: Noctuidae) in field corn in the United States. *J. Integr. Pest Manag.* **2019**, *10*, 1–9. [[CrossRef](#)]
- Reisig, D.D.; Akin, D.S.; All, J.N.; Bessin, R.T.; Brewer, M.J.; Buntin, D.G.; Catchot, A.L.; Cook, D.; Flanders, K.L.; Huang, F.N.; et al. Lepidoptera (Crambidae, Noctuidae, and Pyralidae) injury to corn containing single and pyramided Bt traits, and blended or block refuge, in the southern United States. *J. Econ. Entomol.* **2015**, *108*, 157–165. [[CrossRef](#)]
- Zhang, D.; Xiao, Y.; Xu, P.; Yang, X.; Wu, Q.; Wu, K. Insecticide resistance monitoring for the invasive populations of fall armyworm, *Spodoptera frugiperda* in China. *J. Integr. Agric.* **2021**, *20*, 783–791. [[CrossRef](#)]
- Sisay, B.; Tefera, T.; Wakgari, M.; Ayalew, G.; Mendesil, E. The efficacy of selected synthetic insecticides and botanicals against fall armyworm, *Spodoptera frugiperda*, in Maize. *Insects* **2019**, *10*, 45. [[CrossRef](#)]

8. Aguirre, L.; Hernández-Juárez, A.; Flores, M.; Cena, E.; Landeros, J.; Frías, G.; Harris, M. Evaluation of foliar damage by *Spodoptera frugiperda* (Lepidoptera: Noctuidae) to genetically modified corn (Poales: Poaceae) in Mexico. *Fla. Entomol.* **2016**, *99*, 276–280. [[CrossRef](#)]
9. Bohnenblust, E.; Breining, J.; Fleischer, S.; Roth, G.; Tooker, J. Corn earworm (Lepidoptera: Noctuidae) in northeastern field corn: Infestation levels and the value of transgenic hybrids. *J. Econ. Entomol.* **2013**, *106*, 1250–1259. [[CrossRef](#)]
10. Viteri, D.M.; Linares, A.M.; Cabrera, I.; Sarmiento, L. Presence of corn earworm and fall armyworm (Lepidoptera: Noctuidae) populations in sweet corn and their susceptibility to insecticides in Puerto Rico. *Fla. Entomol.* **2019**, *102*, 451–454. [[CrossRef](#)]
11. Blanco, C.A.; Chiaravalle, W.; Dalla-Rizza, M.; Farias, J.R.; García-Degano, M.F.; Gastaminza, G.; Mota-Sánchez, D.; Murúa, M.G.; Omoto, C.; Pieralisi, B.K.; et al. Current situation of pests targeted by Bt crops in Latin America. *Curr. Opin. Insect Sci.* **2016**, *15*, 131–138. [[CrossRef](#)] [[PubMed](#)]
12. da Silva, R.; Trujillo, D.; Bernardi, O.; Rodriguez, J.C.; Bailey, W.D.; Gilligan, T.M.; Carrillo, D. Comparative toxicity of *Helicoverpa armigera* and *Helicoverpa zea* (Lepidoptera: Noctuidae) to selected insecticides. *Insects* **2020**, *11*, 431. [[CrossRef](#)] [[PubMed](#)]
13. Schlum, K.; Lamour, K.; Tandy, P.; Emrich, S.J.; de Bortoli, C.P.; Rao, T.; Viteri-Dillon, D.M.; Linares-Ramirez, A.M.; Jurat-Fuentes, J.L. Genetic screening to identify candidate resistance alleles to Cry1F corn in fall armyworm using targeted sequencing. *Insects* **2021**, *12*, 618. [[CrossRef](#)] [[PubMed](#)]
14. Viteri, D.M.; Linares, A.M.; Flores, L. Use of the entomopathogenic nematode *Steinernema carpocapsae* in combination with low-toxicity insecticides to control fall armyworm (Lepidoptera: Noctuidae) larvae. *Fla. Entomol.* **2018**, *101*, 327–329. [[CrossRef](#)]
15. Harding, R.S.; Nault, B.A.; Seaman, A.J. Lepidopteran pest control in sweet corn with insecticides allowed for organic production, 2018. *Arthropod Manag. Tests* **2020**, *45*, 1–2. [[CrossRef](#)]
16. Olmstead, D.L.; Shelton, A.M. Effects of timing and insecticide on management of *Helicoverpa zea* (Lepidoptera: Noctuidae) in sweet corn (Poales: Poaceae). *Fla. Entomol.* **2016**, *102*, 451–454.
17. Qureshi, J.; Kostyk, B.C. Control of fall armyworm with labeled and experimental biopesticides and insecticides in sweet corn, fall 2020. *Arthropod Manag. Tests* **2021**, *46*, 1–2. [[CrossRef](#)]
18. Muñoz, M.A.; Lugo, W.I.; Santiago, C.; Matos, M.; Rios, S.; Lugo, J. Taxonomic classification of the soils of Puerto Rico, 2017. In *Bulletin 313*; Agricultural Experimental Station: San Juan, Puerto Rico, 2018; pp. 14–26.
19. McIntosh, M.S. Analysis of combined experiments. *Agron. J.* **1983**, *75*, 153–155. [[CrossRef](#)]
20. SAS Institute. *SAS/STAT User's Manual, version 9.4*; SAS Institute: Cary, NC, USA, 2012.
21. Fleming, R.; Retnakaran, A. Evaluating single treatment data using Abbott's formula with reference to insecticides. *J. Econ. Entomol.* **1985**, *78*, 1179–1181. [[CrossRef](#)]
22. Møl, R.; Matthys-Rocho, E.; Dumas, C. The kinetics of cytological during double fertilization in *Zea mays* L. *Plant J.* **1994**, *5*, 197–206. [[CrossRef](#)]
23. Tao, T.Y.; Ouellet, T.; Dadej, K.; Miller, S.S.; Johnson, D.A.; Singh, J. Characterization of a novel glycine-rich protein from the cell wall of maize silk tissues. *Plant Cell Rep.* **2006**, *25*, 848–858. [[CrossRef](#)] [[PubMed](#)]
24. Loneman, D.M.; Peddicord, L.; Al-Rashid, A.; Nikolau, B.J.; Yandeu-Nelson, M.D. A robust and efficient method for the extraction of plant extracellular surface lipids as applied to the analysis of silks and seedling leaves of maize. *PLoS ONE* **2017**, *12*, e0180850. [[CrossRef](#)] [[PubMed](#)]
25. Yajima, T.; Fujita, M.; Lijima, K.; Sato, K. Influence of various parts of sweet corn ears on pesticide residue levels. *J. Pestic. Sci.* **2017**, *42*, 52–57. [[CrossRef](#)] [[PubMed](#)]
26. European Food Safety Authority (EFSA); Anastassiadou, M.; Bernasconi, G.; Brancato, A.; Carrasco-Cabrera, L.; Greco, L.; Jarrah, S.; Kazocina, A.; Leuschner, R.; Magrans, J.O.; et al. Review of the existing maximum residue levels for spinetoram according to Article 12 of Regulation (EC) No 396/2005. *ESEA J.* **2020**, *18*, e05997.
27. He, M.; Song, D.; Jia, H.C.; Zheng, Y. Concentration and dissipation of chlorantraniliprole and thiamethoxam residues in maize straw, maize, and soil. *J. Environ. Sci. Health* **2016**, *51*, 594–601. [[CrossRef](#)]
28. Nuessly, G.S.; Hentz, M.G. Contact and leaf residue activity of insecticides against the sweet corn *Euxesta stigmatias* (Diptera: Otitidae). *J. Econ. Entomol.* **2004**, *97*, 496–502. [[CrossRef](#)]
29. Sial, A.A.; Brunner, J.F. Toxicity and residual efficacy of chlorantraniliprole, spinetoram, and emamectin benzoate to obliquebanded leafroller (Lepidoptera: Tortricidae). *J. Econ. Entomol.* **2010**, *103*, 1277–1285. [[CrossRef](#)]
30. Tushabe, D.; Rosbakh, S. A compendium of in vitro germination media for pollen research. *Front. Plant Sci.* **2021**, *12*, 709945. [[CrossRef](#)]
31. Gentile, A.G.; Vaughan, A.W.; Richman, S.M.; Eaton, A.T. Corn pollen germination and tube elongation inhibited or reduced by commercial and experimental formulations of pesticides and adjuvants. *Environ. Entomol.* **1973**, *2*, 473–476. [[CrossRef](#)]
32. Córdova, D.; Benner, E.A.; Sacher, M.D.; Rauh, J.J.; Sopa, J.S.; Lahm, G.P.; Selby, T.P.; Stevenson, T.M.; Flexner, L.; Gutteridge, S.; et al. Anthranilic diamides: A new class of insecticides with a novel mode of action, ryanodine receptor activation. *Pestic. Biochem. Phys.* **2006**, *84*, 196–214. [[CrossRef](#)]
33. Salgado, V. Studies on the mode of action of spinosad: Insect symptoms and physiological correlates. *Pestic. Biochem. Physiol.* **1998**, *60*, 91–102. [[CrossRef](#)]

34. Sarmiento, L.; Viteri, D.M.; Linares, A.M.; Cabrera, I. Bio-ensayos de insecticidas biológicos y orgánicos sintéticos en larvas del gusano de la mazorca del maíz [*Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae)]. *J. Agric. Univ. P. R.* **2022**, *105*. in press.
35. Negrisoli, A.S.; Garcia, M.S.; Barbosa, C.R.C.; Bernardi, D.; da Silva, A. Efficacy of entomopathogenic nematodes (Nematoda: Rhabditida) and insecticide mixtures to control *Spodoptera frugiperda* (Smith, 1797) (Lepidoptera: Noctuidae) in corn crops. *Crop Protect.* **2010**, *29*, 677–683. [[CrossRef](#)]