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Source: Florida Entomologist, 94(2) : 272-278

Published By: Florida Entomological Society

URL: <https://doi.org/10.1653/024.094.0221>

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## LABORATORY TOXICITY AND FIELD EFFICACY OF SELECTED INSECTICIDES AGAINST FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE)<sup>1</sup>

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<sup>1</sup>Presented verbally at The Armyworm Symposium held in conjunction with the Entomological Society of America Southeastern Branch Meeting, March 6-10, 2010 in Atlanta.

### ABSTRACT

Fall armyworm, *Spodoptera frugiperda* (J. E. Smith), is an occasional but often serious pest of several row crops in the southern U.S., including cotton, field corn, and grain sorghum. The objective of these studies was to generate baseline dose-mortality responses for fall armyworm larvae in laboratory bioassays, to confirm field efficacy against natural infestations, and to determine residual efficacy of selected insecticides. These studies evaluated 4 recently developed insecticides (chlorantraniliprole, cyantraniliprole, flubendiamide, and spinetoram) and 5 commercial standards (indoxacarb, lambda-cyhalothrin, methoxyfenozide, novaluron, and spinosad). In diet-incorporated assays, the  $LC_{50}$  values of chlorantraniliprole and spinetoram were significantly lower than the  $LC_{50}$ 's of all other insecticides. The results of a field trial against a native fall armyworm infestation in grain sorghum indicated that chlorantraniliprole reduced the number of infested whorls below that in the non-treated control and the lambda-cyhalothrin- and methoxyfenozide-treated plots at 3 d after treatment (DAT). At 7 DAT, no insecticides significantly reduced the number of infested whorls below that in the non-treated plots. In residual efficacy studies, exposure of fall armyworm larvae to chlorantraniliprole- and cyantraniliprole-treated tissue resulted in significantly greater mortality compared to those exposed to non-treated tissue and lambda-cyhalothrin-, flubendiamide-, novaluron-, and methoxyfenozide-treated tissues at 7 DAT. In addition, chlorantraniliprole and cyantraniliprole were the only compounds that resulted in >40% mortality at 28 DAT. These results indicate that newer insecticides are equal to or more efficacious against fall armyworm than traditional insecticides.

Key Words: *Spodoptera frugiperda*, dose-mortality responses, chemical control, IPM

### RESUMEN

El gusano cogollero, *Spodoptera frugiperda* (J. E. Smith), es una plaga ocasional pero a menudo sería en varios cultivos de surcos en el sur de los Estados Unidos, incluyendo algodón, maíz de campo y sorgo de grano. El objetivo de estos estudios fue para generar una línea basal de respuestas a las dosis mortales para el gusano cogollero en bioensayos del laboratorio, para confirmar la eficacia en el campo contra infestaciones naturales, y para determinar la eficacia de residuos de insecticidas seleccionados. Estos estudios evaluaron 4 de los insecticidas recién desarrollados (chlorantraniliprole, cyantraniliprole, flubendiamide y spinetoram) y 5 productos comerciales estándares (indoxacarb, lambda-cyhalothrin, methoxyfenozide, novaluron, y spinosad). En ensayos de dietas incorporadas, los valores de  $CL_{50}$  de chlorantraniliprole y spinetoram fueron significativamente más bajos que los  $CL_{50}$  de los otros insecticidas. Los resultados de las pruebas de campo contra una infestación nativa del gusano cogollero en sorgo de grano indicaron que chlorantraniliprole redujo el número de los cogollos infestados y fue mas bajo que en las parcelas de control no-tratadas y tratadas con lambda-cyhalothrin- y methoxyfenozide a los 3 días después del tratamiento (con sus siglas en inglés - DAT). A los 7 DAT, ninguno de los insecticidas redujeron significativamente el número de cogollos infestados más bajo que en las parcelas no-tratadas. En estudios de la eficacia de residuo, larvas de gusano cogollero expuestas al tejido tratado con chlorantraniliprole y cyantraniliprole resultaron en una mortalidad significativamente mas alta comparada con tejidos no tratados y tejidos tratados con lambda-cyhalothrin, flubendiamide, novaluron, y methoxyfenozide a los 7 DAT. Además, el chlorantraniliprole y cyantraniliprole fueron los únicos compuestos que resultaron en >40% mortalidad a los 28 DAT. Estos resultados indican que los insecticidas más nuevos son iguales o más eficaces contra el gusano cogollero que los insecticidas tradicionales.

Translation provided by the authors.

The fall armyworm, *Spodoptera frugiperda* (J. E. Smith), is an occasional, but serious pest of cotton, *Gossypium hirsutum* (L.), field corn, *Zea mays* (L.), and grain sorghum, *Sorghum bicolor* (L.) Moench, across much of the mid-south and southeastern United States (Luginbill 1928; Buntin 1986; Meagher et al. 2004). Fall armyworm larvae feed on vegetative as well as reproductive structures in these crops (Buntin 1986; Adamczyk et al. 1997). The significance of this pest in crops has been related to the inconsistent performance of many insecticide strategies across a range of plant growth stages.

Ovipositional preference and larval behavior for this species within host plants greatly reduces susceptibility to many insecticides. Adults may deposit clusters of 10-500 eggs throughout the plant canopy, but often prefer to oviposit in the lower two-thirds of cotton plants or in the whorls of corn or sorghum. First instars can be observed in an aggregate near the site of the egg mass, however late instars aggressively disperse within and across adjacent plants (Ali et al. 1989, 1990). Control with insecticides in broad-leaved crops such as cotton can often be difficult due to a lack of sufficient deposition in the lower region of the cotton canopy. As larvae age, they feed inside fruiting structures, or deeper in the whorls of grass crops further reducing their exposure to insecticide applications (Morrill & Greene 1973; Young 1979; Martin et al. 1980; Pitre 1986). In addition, larvae become more tolerant to insecticides as larval age/size increases (Yu 1983; Mink & Luttrell 1989). This tolerance further compounds problems in effectively controlling fall armyworm, as infestations of this pest are typically not discovered until large larvae are common across crop fields.

The development of dose-mortality responses to insecticides is necessary to provide baseline data for future resistance monitoring efforts for pests (Cook et al. 2004). Insecticide resistance surveys exist for bollworm, *Helicoverpa zea* (Boddie), and tobacco budworm, *Heliothis virescens* (F.), but no such coordinated program currently exists for fall armyworm. In addition, several new insecticides have been developed in recent years which exhibit activity against Lepidopteran pests. In most instances, the most appropriate time in the life of an insecticide to establish baseline responses is prior to the widespread use of these products in crops.

Many of these compounds exhibit novel modes of action to which the insect has not yet been exposed. One such group of insecticides is the diamides and includes chlorantraniliprole, cyantraniliprole, and flubendiamide. These molecules are described as ryanodine receptor modulators and affect nerve and muscle action (IRAC Mode of Action Working Group 2009). Spinetoram is another new compound in the chemical class known as

spinosyns. Spinosyns are nicotinic acetylcholine receptor allosteric activators that affect nerve action. The modes of action for diamides and spinosyns differ greatly from that of products currently recommended for control of fall armyworm in crops. Examples of registered products used against this pest are novaluron, a benzoylurea which inhibits chitin biosynthesis and acts as an insect growth regulator (IGR); methoxyfenozide, a diacylhydrazine that is an ecdysone receptor agonist also acting as an IGR; lambda-cyhalothrin, a pyrethroid which acts as a sodium channel modulator affecting nerve action; indoxacarb, a blocker of voltage dependent sodium channels in the nervous system; and spinosad, an older spinosyn with a similar mode of action to that of spinetoram. Newer compounds with novel modes of action have the potential to improve integrated pest management (IPM) and delay insect resistance in row crops in southern states by providing growers with additional tools to control fall armyworm.

The objective of these studies was to generate insecticide dose-mortality responses for fall armyworm larvae in diet-incorporation bioassays, confirm field efficacy against natural infestations, and determine residual properties in the field environment. These results will provide reference data for future insecticide susceptibility surveys and give support to IPM recommendations for the use of insecticides against field infestations of fall armyworm.

## MATERIALS AND METHODS

### Laboratory Bioassays

Fall armyworms were obtained from a laboratory colony (LSU-FAW) maintained at the Louisiana State University Department of Entomology, Baton Rouge, LA. This colony was established in 2005 from multiple collections in cotton, and supplemented with additional samples from field corn during 2006 and 2008. Based on mitochondrial markers, the colony was validated as the corn strain of fall armyworm (Unpublished communication, R. Nagoshi, USDA-ARS, Gainesville, FL).

Larvae were fed a meridic semi-solid diet (Ward's Natural Science, Rochester, NY) prepared according to manufacturer's recommendations. Rearing conditions consisted of a 14:10 light-dark photoperiod, 23.9 to 29.4°C, and 80% relative humidity (Cook et al. 2004).

Insecticides used in the bioassay included chlorantraniliprole (Coragen 200 g/L Soluble Concentrate [SC], DuPont Crop Protection, Wilmington, DE), cyantraniliprole (HGW-86, 200 g/liter SC, DuPont Crop Protection, Wilmington, DE), flubendiamide (Belt 480 g/L SC, Bayer Crop Science, Research Triangle Park, NC), indoxacarb (Steward 150 g/L Emulsifiable Concentrate [EC],

DuPont Crop Protection, Wilmington, DE), lambda-cyhalothrin (Karate-Z 250 g/liter EC, Syngenta Crop Protection, Greensboro, NC), methoxyfenozide (Intrepid 240 g/L Flowable [F], Dow AgroSciences, Indianapolis, IN), novaluron (Diamond 100 g/liter EC, Makhteshim Agan of North America, Inc., Raleigh, NC), spinetoram (Radiant 120 g/L SC, Dow AgroSciences, Indianapolis, IN), and spinosad (Tracer 480 g/L SC, Dow AgroSciences, Indianapolis, IN). Formulated products were used to create all initial concentrations.

Procedures similar to Temple et al. (2009) were used for preparing diet-incorporated insecticide bioassays. Insecticides were dissolved in distilled water to create a stock solution of 100 µg/mL. Serial dilutions of desired concentrations were standardized to 30 mL for each insecticide: water mixture. The insecticide solution was mixed with meridic diet to yield 200 mL of a diet/insecticide mixture. This mixture of diet and insecticide solution was agitated for 30-45 s in a 2.0-L bowl with a hand mixer. Insecticide-treated diet was then placed in 30-mL plastic cups with approximately 7 mL of diet per cup. Insecticide concentrations in the diet ranged from 0.25 µg/mL to 30.0 µg/mL diet. The insecticide-treated diet was stored in a refrigerator and used within 7 d of preparation. Four to 7 replicates (30-105 larvae per dose) were used for each insecticide. Fall armyworms (L3 stage; 30-45 mg) were placed on insecticide-treated and non-treated (control) diet. Insect mortality was evaluated at 96 h after exposure (HAE). A larva was considered dead if it could not right itself after being placed on its dorsal surface. Data were corrected for control mortality (0-5%) (Abbott 1925) and analyzed by probit analysis with Polo-Plus (LeOra Software 2006) to obtain  $LC_{50}$  values. Non-overlapping confidence limits (95%) were used to indicate significant differences among insecticides. Values are reported as concentration of insecticide (µg/mL diet).

#### Field Experiments

Insecticide screening studies on grain sorghum were conducted during 2009 at the LSU AgCenter Macon Ridge Research Station (Franklin Parish, LA). Plots were planted to sorghum var. Terral TV 1050 (Terral Seed, Inc., Lake Providence, LA) on 8 Jun 2009. Plots consisted of 8 rows on 1-m centers and 15.24 m long. Treatments were arranged in a randomized complete block design with 4 replications. Cultural practices recommended by the LSU AgCenter were used to maintain plots in a consistent manner within the trial.

The treatments included chlorantraniliprole, cyantraniliprole, flubendiamide, lambda-cyhalothrin, methoxyfenozide, novaluron, and a non-treated control. Insecticides were applied on 24

Jul 2009, with a high-clearance sprayer and a CO<sub>2</sub>-charged spray system calibrated to deliver 89.78 L per ha through TX-8 hollow cone nozzles (Spraying Systems Company, Wheaton, IL). Pre-treatment samples across the test area indicated that >50% of plant whorls were infested with 1 or more fall armyworm in several stages of larval development.

Treatment efficacy was determined 3 and 7 d after treatment (DAT). Within each plot, a single plant was randomly selected on 1 of the center rows (Rows 4 or 5). That plant and the next 9 consecutive plants were destructively sampled and examined for fall armyworm infested whorls. Number of infested whorls was calculated as percent infestation of plants within each plot. Data were analyzed by PROC GLM and means separated according to Tukey's Studentized Range Test (SAS Institute 2004).

#### Residual Efficacy in a Field Environment

Larvae were removed from the same colony (LSU-FAW) previously described for the laboratory experiments. At 0 (4 HAT), 7, 14, 21, and 28 DAT, sorghum leaf tissue (non-treated and insecticide-treated) was removed from plants in the previously described field trial. Plants were mapped for leaf collars at the time of treatment application to ensure that the leaves selected during all time periods of the study were present at the time insecticides were applied. Leaf tissue was harvested on each date from the uppermost fully-expanded leaf that was present at the time of treatment application. Leaves were immediately transported to the laboratory and dissected into tissue sections averaging 2.5 cm<sup>2</sup>. Two second instars (3-4-d-old) were placed into each cell of a plastic bioassay tray (CD International, Pitman, NJ), each containing 3 pieces of leaf tissue. Thirty-two larvae were infested on each treatment at each infestation timing (8 larvae per replication). Larvae were evaluated for mortality 72 HAE on leaf tissue. Larval mortality was determined by methods previously described in the laboratory bioassays. Percent mortality data was analyzed and compared among treatments at each DAT interval according to methods described for the field trials.

## RESULTS AND DISCUSSION

#### Laboratory Bioassays

The  $LC_{50}$  values among insecticides ranged from 0.066 µg/mL for spinetoram to 5.27 µg/mL for lambda-cyhalothrin (Table 1). The newer insecticides, chlorantraniliprole, cyantraniliprole, flubendiamide, and spinetoram had  $LC_{50}$ 's ranging from 0.066 µg/mL to 0.93 µg/mL and were generally lower than those observed for the older traditional

TABLE 1. DOSE-MORTALITY RESPONSES OF FALL ARMYWORM (LSU-FAW) LARVAE IN DIET-INCORPORATED ASSAYS 96 H AFTER EXPOSURE.

Insecticide <sup>1</sup>	<i>n</i> <sup>2</sup>	LC <sub>50</sub> <sup>3</sup>	95% C.L. <sup>3,4</sup>	Slope ± SE	χ <sup>5</sup>	<i>df</i> <sup>6</sup>
Diamide (28)						
Chlorantraniliprole	685	0.068	0.060-0.077	2.55 ± 0.23	2.92	6
Cyantraniliprole	310	0.118	0.097-0.141	2.88 ± 0.34	1.95	4
Flubendiamide	420	0.930	0.775-1.126	1.99 ± 0.18	2.87	4
Indoxacarb (22A)						
Indoxacarb	300	0.392	0.317-0.481	2.35 ± 0.25	4.09	5
Pyrethroid (3A)						
Lambda-cyhalothrin	210	5.270	4.028-6.797	2.02 ± 0.25	3.29	4
Diacylhydrazine (18)						
Methoxyfenozide	225	0.875	0.658-1.037	3.13 ± 0.62	2.33	3
Benzoylurea (15)						
Novaluron	270	0.166	0.112-0.220	1.74 ± 0.25	1.75	3
Spinosyn (5)						
Spinetoram	210	0.066	0.053-0.081	2.54 ± 0.36	2.43	4
Spinosad	210	0.557	0.382-0.879	2.21 ± 0.30	4.21	4

<sup>1</sup>IRAC Mode of Action classification\_v6.3.3\_28July09.pdf Working Group 2009, <http://www.irac-online.org/wp-content/uploads/2009/09/MoA->

<sup>2</sup>Number of insects tested.

<sup>3</sup>µg/mL.

<sup>4</sup>Confidence Limits.

<sup>5</sup>Chi square values (no significant values).

<sup>6</sup>Degrees of freedom.

insecticides (indoxacarb, lambda-cyhalothrin, methoxyfenozide, novaluron, and spinosad) with LC<sub>50</sub>'s ranging from 0.166 µg/mL to 5.27 µg/mL. Fall armyworm larvae were significantly less susceptible to lambda-cyhalothrin than all other insecticides. Spinetoram (0.066 µg/mL) and chlorantraniliprole (0.068 µg/mL) were significantly more toxic to fall armyworm than all other insecticides.

These results represent initial efforts to develop baseline data for new insecticides with reference data for several commercial products that are currently used against fall armyworm. Although the use of diet-incorporated bioassays may not provide the optimum measure of the toxicity for all compounds, the procedure appeared to perform well for those products that require ingestion. Evaluations for mortality at 96 HAE may not allow sufficient time to accurately gauge the maximum effectiveness of the IGR's, novaluron and methoxyfenozide, but did allow for comparisons among several chemistries. This standard methodology and baseline data should assist in monitoring for changes in susceptibility to these new insecticides as their use becomes widespread across multiple crops in the southern United States.

Several insecticides representing various classes of chemistry have been evaluated against fall armyworm in recent years with bioassays of meridic diet surface-treated with insecticides. Adamczyk et al. (1999) exposed third instar fall armyworms to insecticide-treated diet and devel-

oped LC<sub>50</sub> values for methoxyfenozide (197.9 ppm, ppm = parts per million) and spinosad (4.4 ppm), both of which represent toxicity values significantly higher than those found in the current study. Cook et al. (2001) conducted a study similar to Adamczyk et al. (1999) using first instars on indoxacarb-treated diet (LC<sub>50</sub> = 0.59 ppm). This value is similar to data presented herein, although for smaller larvae. Argentine et al. (2002) also exposed first instars to diet-overlay assays using chlorfenapyr (1.2 ppm), emamectin benzoate (0.0029 ppm), fipronil (2.4 ppm), and tebufenozide (0.95 ppm). Results from these studies suggest significant effects on insecticide toxicity are present between the surface-treated (diet-overlay) assays and diet incorporated assays.

Field Trials

Fall armyworm infested whorls in the insecticide-treated plots ranged from 10 to 45% at 3 DAT and from 2.5 to 40% at 7 DAT (Table 2). Chlorantraniliprole (10.0%), cyantraniliprole (12.5%), and novaluron (15.0%) significantly reduced fall armyworm infested whorls compared to that in the non-treated control (50.0%) and lambda-cyhalothrin-treated (45.0%) plots at 3 DAT. Chlorantraniliprole also significantly reduced infestations below that in the methoxyfenozide-treated (40.0%) plots. At 7 DAT, no significant treatment effect was detected compared to the non-treated control. However, the

TABLE 2. EFFICACY OF SELECTED INSECTICIDES AGAINST FALL ARMYWORM IN A GRAIN SORGHUM FIELD TRIAL.

Treatment	Rate per ha (kg AI)	Percent ( $\pm$ SE) fall armyworm-infested whorls	
		3 DAT <sup>1</sup>	7 DAT <sup>1</sup>
Chlorantraniliprole	0.101	10.0 c $\pm$ 5.8	2.5 b $\pm$ 2.5
Cyantraniliprole	0.098	12.5 bc $\pm$ 6.3	5.0 b $\pm$ 5.0
Flubendiamide	0.106	32.5 abc $\pm$ 7.5	10.0 b $\pm$ 7.1
Lambda-cyhalothrin	0.034	45.0 a $\pm$ 6.5	40.0 a $\pm$ 7.1
Methoxyfenozide	0.101	40.0 ab $\pm$ 9.1	22.5 ab $\pm$ 6.3
Novaluron	0.088	15.0 bc $\pm$ 6.5	2.5 b $\pm$ 2.5
Non-treated control	—	50.0 a $\pm$ 4.1	35.0 ab $\pm$ 2.9
<i>df</i>		6, 18	6, 18
<i>F</i> value		6.78	12.45
( <i>P</i> > <i>F</i> ) ANOVA		0.0007	<0.0001

Means within columns followed by a common letter are not significantly different ( $P \leq 0.05$  Tukey's Studentized Range Test).  
<sup>1</sup>Days after treatment.

newer compounds (chlorantraniliprole, cyantraniliprole, and flubendiamide) reduced fall armyworm infestations by >2.5-fold below that in the non-treated control. The newer insecticides displayed efficacy equal to or greater than standard insecticides (indoxacarb, lambda-cyhalothrin, methoxyfenozide, novaluron, and spinosad) currently recommended for control of fall armyworm (Baldwin et al. 2010; Catchot 2010; Studebaker 2010). Although additional research with these insecticides is needed, the results presented in this study should aid producers in making fall armyworm management decisions. The poor control with lambda-cyhalothrin in this study was not surprising given that previous by Guillebeau & All (1990) evaluating a range of insecticides for control of fall armyworm in whorl-stage corn and sorghum showed considerable variability in the effectiveness of several pyrethroids.

Mink & Luttrell (1989) exposed fall armyworm larvae to insecticide-treated cotton tissue in the laboratory. Their findings indicated significant levels of mortality when larvae were directly exposed to organophosphate, carbamate, and pyrethroid-treated tissue. However, against natural infestations in a field environment, the performance of these insecticides on cotton may not be as consistent; especially if larvae are located low in the plant canopy and insecticide deposition is an issue.

Residual Efficacy Experiments

Fall armyworm mortality on all insecticide-treated tissue at 0 DAT (4 HAT) (90.6 to 100%) and at 7 DAT (28.1 to 96.9%) was significantly higher than that on non-treated control tissue (Table 3). Mortality on chlorantraniliprole

TABLE 3. RESIDUAL EFFICACY OF SELECTED INSECTICIDES AGAINST FALL ARMYWORM (LSU-FAW) LARVAE ON GRAIN SORGHUM TISSUE.

Insecticide	Rate per ha (kg AI)	% Mortality 72 HAE <sup>1</sup>				
		0 DAT <sup>2</sup>	7 DAT <sup>2</sup>	14 DAT <sup>2</sup>	21 DAT <sup>2</sup>	28 DAT <sup>2</sup>
Chlorantraniliprole	0.101	100.0 a	96.9 a	85.9 a	82.8 a	53.1 a
Cyantraniliprole	0.098	100.0 a	93.8 a	75.0 ab	75.0 a	43.8 ab
Flubendiamide	0.106	93.8 a	53.1 cd	26.6 c	9.4 bc	— <sup>3</sup>
Lambda-cyhalothrin	0.034	90.6 a	28.1 d	5.6 cd	6.3 bc	— <sup>3</sup>
Methoxyfenozide	0.101	92.2 a	89.1 ab	53.1 b	29.7 b	20.3 bc
Novaluron	0.088	92.2 a	65.6 bc	23.4 cd	14.1 bc	12.5 bc
Non-treated control	—	9.4 b	1.6 e	1.6 d	0.0 c	0.0 c
<i>df</i>		6, 38	6, 38	6, 39	6, 30	4, 20
<i>F</i> value		175.45	41.30	34.58	40.22	7.51
( <i>P</i> > <i>F</i> ) ANOVA		<0.0001	<0.0001	<0.0001	<0.0001	0.0007

Means within columns followed by a common letter are not significantly different ( $P \leq 0.05$  Tukey's Studentized Range Test).  
<sup>1</sup>h after exposure.  
<sup>2</sup>d after treatment.  
<sup>3</sup>Not included in the analysis due to low sample number of larvae.

(96.9%) and cyantraniliprole-treated tissue (93.8%) significantly differed from that for all other treatments, except methoxyfenozide (89.1%) at 7 DAT. At 14 DAT, mortality on chlorantraniliprole (85.9%), cyantraniliprole (75.0%), flubendiamide (26.6%), and methoxyfenozide-treated tissue (53.1%) was significantly different from that of larvae on the non-treated tissue. In addition, chlorantraniliprole caused significantly higher mortality than all insecticides except cyantraniliprole. At 21 DAT, chlorantraniliprole (82.8%) and cyantraniliprole (75.0%) caused significantly higher mortality than the non-treated control (0.0%) and all other insecticide treatments (6.3 to 14.1%) except methoxyfenozide (29.7%). Only chlorantraniliprole (53.1%) and cyantraniliprole (43.8%) caused mortality significantly higher than that on the non-treated control (0%) at 28 DAT. These results suggest that the newer insecticides generally exhibited longer residual efficacy compared to that for several of the standard insecticides (lambda-cyhalothrin, methoxyfenozide, and novaluron) currently recommended for fall armyworm management.

Long (>21 DAT) residual efficacy provided by compounds may help to reduce insecticide application frequency necessary to achieve satisfactory control of persistent fall armyworm infestations. Additional research is necessary to determine the ecological effects of the persistent nature of these products in a row-crop ecosystem. Further fieldwork is also needed to compliment these laboratory studies to determine the most effective rates of compounds given their respective residual properties. Finally, research is needed to understand the most appropriate timing for applications of these insecticides in order to maximize their effectiveness in various cropping systems.

#### ACKNOWLEDGMENTS

The authors thank Trey Price, Ralph Sheppard, Karla Emfinger, and the numerous student workers at the Macon Ridge Research Station for assistance with laboratory and field studies, and colony maintenance. The authors thank the LSU AgCenter, Cotton Incorporated, and the Louisiana Soybean and Grain Promotion Board for financial support. This article was approved for publication by the Director of the Louisiana Agricultural Experiment Station as manuscript No. 2010-258-9502.

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