COMPARATIVE EFFICACY OF SELECTED BOLL WEEVIL INSECTICIDES USING LABORATORY BIOASSAYS Boris A. Castro Texas AgriLife Extension Service Weslaco, TX Scott Armstrong USDA, ARS Weslaco, TX

Abstract

The efficacy of selected insecticides was evaluated for adult boll weevil mortality using treated leaf-disk bioassays in 2007. The experiments were conducted using native adult boll weevils collected either with pheromone traps or reared from infested squares obtained from commercial cotton fields in the Lower Rio Grande Valley of Texas. Boll weevil collections included those from the pre-squaring season (overwintered adult boll weevils), in-season boll weevil populations, and adult boll weevils collected after cotton harvest. Insecticides included malathion ULV, endosulfan, bifenthrin, encapsulated methyl parathion, oxamyl, carbaryl and cyfluthrin. Potted cotton plants were sprayed with a hand-boom sprayer except the malathion ULV formulation which was applied using a hand-held Micron ULVA+ controlled droplet applicator. Treated leaves were cut into disks and placed inside tissue culture plates. Individual adult boll weevils were placed inside culture plate cells in a no-choice situation and mortality assessed at 24, 48 and 72 hours. Boll weevil mortality in the malathion ULV, endosulfan, encapsulated methyl parathion and bifenthrin treatments was consistently at or near 100% at the end of the bioassays. Boll weevil mortality observed in the cyfluthrin and different rates of carbaryl was low and inconsistent throughout the bioassays. Boll weevil mortality observed in the oxamyl treatment was intermediate between the two groups. Highest boll weevil mortality after 24 hours of exposure was observed in the malathion ULV, endosulfan and bifenthrin treatments (97.9%, 86.6% and 80.2%, respectively). After 48 hours of exposure, boll weevil mortality reached 100% in the malathion ULV treatment but was not significantly different from those of encapsulated methyl parathion (96.1%), bifenthrin (95%) and endosulfan (94.9%). Results from this study indicate that, among the materials tested, malathion ULV is a highly effective material for boll weevil control and that encapsulated methyl parathion, bifenthrin and endosulfan also are effective in causing significantly high mortality to adult boll weevils.

Introduction

Malathion ULV is currently the only insecticide being used to control the boll weevil, *Anthonomus grandis grandis* (Boheman), in active Boll Weevil Eradication Programs. While substantial areas of the cotton belt have been declared either boll weevil-free or functionally-eradicated, most production regions of Texas and surrounding states remain in active eradication programs. The Lower Rio Grande Valley of Texas (LRGV) completed its second full season under the Texas Boll Weevil Eradication Program, and is not projected to attain functionally-eradicated status for several years. Availability of malathion ULV is critical for boll weevil eradication programs, both for achieving eradication and for subsequently maintaining that status. Should this effective and economical insecticide become unavailable, efforts toward eradication status could be jeopardized. There has been limited progress in identifying replacements for malathion ULV that meet the criteria of current registration, low environmental impact, low cost, and high level of efficacy against the boll weevil. Because all cotton areas in the United States are now engaged in eradication or maintenance efforts, opportunities to conduct efficacy studies are becoming limited.

Previous studies evaluated the efficacy of selected insecticide alternatives in an indirect manner by recording the number or percentages of punctured squares and yields (Sparks et al. 1997, Parker and Huffman 1997, Raulston et al. 1998) and not by a direct examination of the tested materials on boll weevil mortality. Field experiments indicate that azinphosmethyl (Guthion[®]), fipronil (Regent[®]), oxamyl (Vydate[®]), endosulfan (Phaser[®] and Thiodan[®]), lambda-cyhalothrin (Karate[®]), cyfluthrin (Baythroid[®]), and micro encapsulated methyl parathion (Penncap-M[®]) reduced boll weevil damage and increased cotton lint yields (Parker and Huffman 1997). The research provided valuable data for yield potential preservation of tested materials but limited information is revealed on the efficacy of the materials to eliminate the boll weevil. Additional research on field efficacy of selected materials to manage boll weevil damage often was confounded by multiple field and other environmental variables (England et al. 1997, Raulston et al. 1998). Field evaluations by Raulston et al. (1998) revealed that weekly applications of fipronil

resulted in lower percentages of punctured squares than did weekly applications of azinphosmethyl. However, fipronil plots had higher numbers of both punctured and non-punctured squares than did the azinphosmethyl plots. The authors concluded that the higher numbers of total squares in the fipronil plots probably resulted from product efficacy. However, the higher numbers of punctured squares likely resulted from a combination of a higher number of squares (offering increased opportunity for population recruitment and increased attractiveness relative to adjacent plots) and inter-plot movement of boll weevils. Similar results were obtained by Sparks et al. (1997) evaluating also fipronil and azinphosmethyl. Fipronil provided the highest residual activity in leaf-disk exposure bioassays. However, oviposition-punctured squares were highest in fipronil-treated plants in small replicated plots. The objective of eradication programs (elimination of the pest) is different from those of management programs (preservation of yield potential), and because neither fipronil nor azinphosmethyl are available as alternatives to malathion ULV, there is a critical need for direct assessment of efficacy corresponding to materials currently labeled for cotton. The objective of this project was to directly evaluate efficacies of selected insecticides as alternatives to malathion ULV on native, field collected adult boll weevils using laboratory bioassays.

Materials and Methods

Insecticides. Selected insecticides included oxamyl (Vydate[®] C-LV), carbaryl (Sevin[®] XLR Plus), encapsulated methyl parathion (Penncap[®] M), malathion ULV (Fyfanon[®] ULV), bifenthrin (Capture[®] 2EC), endosulfan (Thionex[®] 3EC) and cyfluthrin (Baythroid[®] 1.0L). Insecticide rates are included in tables.

Boll weevil source. The first and sixth bioassays (on April 6th and Sept 20th, 2007, respectively) were conducted using adult boll weevils from pheromone traps because cotton fields either had no infested squares to rear the adults (April 6^{th}) or were already harvested (Sept 20^{th}). Boll weevil trapping for the first bioassay was conducted for overwintered boll weevils. Trapping location, setup and maintenance followed recommendations outlined in previous studies in the LRGV (Armstrong et al. 2006). Traps were baited with grandlure pheromone and adult boll weevils collected one day later. Boll weevils were transported to the laboratory and were provided with ten percent sucrose water in moistened cotton wicks. Bioassays were conducted one day later after selecting only apparently healthy boll weevils. All other assays were conducted on adult boll weevils reared out from infested squares collected in the LRGV. Squares with single boll weevil oviposition punctures were collected from commercial cotton fields. The squares were collected from the plant as opposed to infested squares on the ground to prevent pathogen contamination. Infested squares collected from plants also increased uniformity of adult boll weevil emergence. Squares were taken to the laboratory and placed in a growth chamber at 85 °F, 50% RH and 14:10 (L:D) photoperiod regime. After 5 to 6 days, squares were dissected and boll weevils that had pupated were placed in 15cm-diameter Petri dishes lined with a thin layer of vermiculite moistened with RO water. Newly emerged adult boll weevils were placed in plexiglass cages and provided with pesticide-free cotton squares at a rate of one square per boll weevil per day. A ten percent sucrose solution was supplied using moistened cotton wicks. Two-day old adult boll weevils were used in bioassays.

Cotton leaf source. Leaves were obtained from greenhouse potted plants from conventional cotton variety Stoneville 474. Cotton was planted in the greenhouse at different intervals during the study to be used both as a leaf source for bioassays and square source to feed emerging adult boll weevils. Insect pests such as aphids, whiteflies and spider mites on greenhouse plants were kept in check by washing the plants with water almost on a daily basis.

Bioassay procedure. Procedures for leaf-disk bioassays are similar to those used by Spurgeon et al. (1997). Expanded leaves from the upper half of the plant were chosen for bioassays. In the first bioassay (April 6th), leaves were excised, sealed in plastic bags and carried to the laboratory. Leaves were placed horizontally on a spray table in an automated spraying system equipped with a 'Microfit' controlled-droplet applicator calibrated to deliver 15 gpa and the specified insecticide rate. A malfunction of the 'Microfit' controlled-droplet applicator prevented the inclusion of the malathion ULV treatment in the first bioassay. In all other bioassays, potted plants were sprayed directly using a hand-held boom equipped with a CO₂-charged system calibrated to deliver 15 gpa at 48 psi through four Tee Jet 8002VS nozzles. Malathion ULV was applied at 12 oz/acre of the ULV formulation using a hand-held Micron ULVA+ controlled droplet applicator (Bromyard Industrial Estate, Bromyard, Herefordshire, HR7 4HS, UK). An untreated check was included by spraying water only. Leaves in all insecticide treatments and the untreated (water) check were allowed to air dry before cutting leaf disks. Leaf disks were cut to fit inside 6-well tissue culture plates (35 mm diameter per well, six wells per culture plate). The bottom of each culture well was lined with a moistened paper filter. Sprayed cotton leaf disks were placed on culture wells and one adult boll weevil was exposed

to the treated surface in a no-choice situation. Experimental units consisted of five or six boll weevils per treatment. Culture plates were arranged in a complete randomized block design with four to six replicates per trial. Mortality was assessed at 24, 48 and 72 hours of continued exposure to the treated leaf-disk surface. The mortality criterion was the absence of response after gently stroking the sides and antennae of the boll weevil for 30 seconds with a camel's hair brush.

Data analysis. Data was corrected for the mortality observed in the untreated checks using Abbott's (1925) formula. Because mortality data were measured in percentages, they were transformed [arcsine $\sqrt{(x/100)}$] for analysis of variance and means separation to normalize the variance distribution. Mortality data were subjected to ANOVA and analyzed using the REPEATED statement within SAS MIXED procedure (SAS Institute Inc. 2001). Differences among treatment means were determined by using the Tukey-Kramer's procedure for a confidence level of $\alpha = 0.05$.

Results

Six sprayed leaf-disk bioassays were conducted during the 2007 cotton season. These assays were conducted on overwintered adult boll weevils collected from pheromone traps along cotton fields (April 6th bioassay, Table 1), from adult boll weevils reared from infested squares (June 20th, 25th, 30th and July 17th bioassays, Tables 2-5) and adult boll weevils from pheromone traps during the post-harvest season (Sept. 20th bioassay, Table 6). Based on the results from the six bioassays, boll weevil mortality in the malathion ULV, endosulfan, encapsulated methyl parathion and bifenthrin treatments was consistently at or near 100% at the end of the bioassays. Efficacy of cyfluthrin and different rates of carbaryl was low and inconsistent throughout the bioassays. Boll weevil mortality observed in the oxamyl treatment was intermediate between the two groups (Fig. 1). Results were consistent in most bioassays except on June 30th (Table 4) when intermittent drizzling was experienced during insecticide spraying of potted plants except during the application of malathion ULV. This could have caused the reduced mortality observed on some insecticidal treatments compared to that of the malathion ULV. Malathion ULV was the only treatment that accomplished 100% boll weevil mortality in all bioassays after 48 hours of treated leaf-disk exposure (Fig. 1). Significantly higher boll weevil mortality after 24 hours was observed in the malathion ULV, endosulfan and bifenthrin treatments (97.9%, 86.6% and 80.2%, respectively, see Fig. 1). At 48 hours of exposure, boll weevil mortality reached 100% in the malathion ULV treatment but was not significantly different from those of encapsulated methyl parathion (96.1%), bifenthrin (95%) and endosulfan (94.9%). A similar trend of significance was observed after 72 hours of exposure (Fig. 1). Data analysis from these experiments did not unveil efficacy problems in the malathion ULV treatment. However, given possible repellency effects of malathion ULV and other insecticides, additional bioassays using cages will be helpful to further evaluate efficacy of insecticides for boll weevil control. Results from this study indicate that, among the materials tested, malathion ULV is highly effective for boll weevil control and encapsulated methyl parathion, bifenthrin and endosulfan also are effective in causing high mortality to adult boll weevils.

Treatment				
	Rate	24 h	48 h	72 h
Encap. methyl parathion (Penncap $M^{\mathbb{R}}$)	0.5 lb a.i./acre	100.0 a	100.0 a	100.0 a
Endosulfan (Thionex 3EC [®])	0.5 lb a.i./acre	100.0 a	100.0 a	100.0 a
Bifenthrin (Capture 2EC®)	0.10 lb a.i./acre	97.2 a	97.2 a	97.2 a
Oxamyl (Vydate C-LV [®])	0.25 lb a.i./acre	36.1 bc	36.1 c	50.0 b
Cyfluthrin (Baythroid XL®)	0.0205 lb a.i./acre	0.0 c	8.3 c	11.1 bc
Carbaryl (Sevin XLR Plus®)	0.25 lb a.i./acre	5.6 c	5.6 c	8.3 c

Table 1. Efficacy of selected insecticides on adult boll weevils using a sprayed leaf-disk bioassay, Lower Rio Grande Valley of Texas, April 6th, 2007 (bioassay 1)¹.

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \le 0.05$; Tukey-Kramer's test). Values are non-transformed data. Significance is based on arcsin [$\sqrt{(x/100)}$] transformed data. Actual mortality percentages are shown after Abbott's (1925) adjustment.

¹Adult boll weevils collected from pheromone traps in the LRGV prior to cotton squaring.

Table 2. Efficacy of selected insecticides on adult boll weevils usi	sing a sprayed leaf-disk bioassay, Lower Rio
Grande Valley of Texas, June 20^{th} , 2007 (bioassay 2) ¹ .	

Treatment	Rate	24 h	48 h	72 h
Endosulfan (Thionex 3EC [®])	0.5 lb a.i./acre	100.0 a	100.0 a	100.0 a
Malathion ULV (Fyfanon [®] ULV)	12 fl oz/acre	92.0 ab	100.0 a	100.0 a
Bifenthrin (Capture 2EC [®])	0.10 lb a.i./acre	88.0 abc	100.0 a	100.0 a
Encap. methyl parathion (Penncap M [®])	0.5 lb a.i./acre	76.0 bc	96.0 a	100.0 a
Oxamyl (Vydate C-LV®)	0.25 lb a.i./acre	60.0 c	64.0 bc	66.0 bc
Carbaryl (Sevin XLR Plus®)	0.5 lb a.i./acre	8.0 d	16.0 d	16.0 d

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \le 0.05$; Tukey-Kramer's test). Values are non-transformed data. Significance is based on arcsin [$\sqrt{(x/100)}$] transformed data. Actual mortality percentages are shown after Abbott's (1925) adjustment.

¹Adult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

Table 3. Efficacy of selected insecticides on adult bol	weevils using a sprayed leaf-disk bioassay, Lower Rio
Grande Valley of Texas, June 25 th , 2007 (bioassay 3) ¹ .	

		Mortality (%)		
Treatment	Rate	24 h	48 h	72 h
Endosulfan (Thionex 3EC [®])	0.5 lb a.i./acre	100.0 a	100.0 a	100.0 a
Malathion ULV (Fyfanon [®] ULV)	12 fl oz/acre	100.0 a	100.0 a	100.0 a
Bifenthrin (Capture 2EC®)	0.10 lb a.i./acre	64.2 bc	90.0 ab	100.0 a
Encap. methyl parathion (Penncap $M^{\mathbb{R}}$)	0.5 lb a.i./acre	61.7 bc	96.7 a	96.7 a
Oxamyl (Vydate C-LV®)	0.25 lb a.i./acre	16.7 d-g	31.7 c-f	50.0 cd
Carbaryl (Sevin XLR Plus®)	1.0 lb a.i./acre	6.7 efg	16.7 d-g	36.7 cde
Cyfluthrin (Baythroid 1.0L®)	0.0205 lb a.i./acre	3.3 fg	10.0 efg	24.1 d-g
Carbaryl (Sevin XLR Plus®)	0.5 lb a.i./acre	0.0 g	3.3 fg	10.8 efg

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \le 0.05$; Tukey-Kramer's test). Values are non-transformed data. Significance is based on arcsin [$\sqrt{(x/100)}$] transformed data. Actual mortality percentages are shown after Abbott's (1925) adjustment.

¹Adult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

Treatment	Rate	24 h	48 h	72 h
Malathion ULV (Fyfanon [®] ULV)	12 fl oz/acre	100.0 a	100.0 a	100.0 a
Encap. methyl parathion (Penncap M®)	0.5 lb a.i./acre	50.0 b-e	75.0 abc	100.0 a
Bifenthrin (Capture 2EC [®])	0.10 lb a.i./acre	40.0 c-f	70.0 a-d	85.0 ab
Cyfluthrin (Baythroid 1.0L®)	0.0205 lb a.i./acre	45.0 b-e	65.0 a-d	75.0 a-d
Endosulfan (Thionex 3EC [®])	0.5 lb a.i./acre	50.0 b-e	60.0 a-d	70.0 a-d
Oxamyl (Vydate C-LV®)	0.25 lb a.i./acre	20.0 def	25.0 def	30.0 c-f
Carbaryl (Sevin XLR Plus [®])	1.0 lb a.i./acre	0.0 f	5.0 ef	5.0 ef

Table 4. Efficacy of selected insecticides on adult boll weevils using a sprayed leaf-disk bioassay, Lower Rio Grande Valley of Texas, June 30^{th} , 2007 (bioassay 4)¹.

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \le 0.05$; Tukey-Kramer's test). Values are non-transformed data. Significance is based on arcsin [$\sqrt{(x/100)}$] transformed data. Actual mortality percentages are shown after Abbott's (1925) adjustment.

¹Adult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

Table 5. Efficacy of selected insecticides on adult boll	weevils using a sprayed leaf-disk bioassay, Lower Rio
Grande Valley of Texas, July 17 th , 2007 (bioassay 5) ¹ .	

		Mortality (%)			
Treatment	Rate	24 h	48 h	72 h	
Malathion ULV (Fyfanon [®] ULV)	12 fl oz/acre	100.0 a	100.0 a	100.0 a	
Bifenthrin (Capture 2EC®)	0.10 lb a.i./acre	86.1 abc	100.0 a	100.0 a	
Encap. methyl parathion (Penncap M [®])	0.5 lb a.i./acre	60.0 bcd	100.0 a	100.0 a	
Endosulfan (Thionex 3EC [®])	0.5 lb a.i./acre	57.2 cd	93.9 ab	93.9 ab	
Cyfluthrin (Baythroid 1.0L®)	0.0205 lb a.i./acre	33.3 def	50.0 cde	64.4 a-d	
Oxamyl (Vydate C-LV®)	0.25 lb a.i./acre	34.5 def	46.1 cde	62.8 bcd	
Carbaryl (Sevin XLR Plus [®])	1.0 lb a.i./acre	11.1 ef	53.9 cd	65.6 bcd	
Carbaryl (Sevin XLR Plus®)	0.5 lb a.i./acre	2.8 f	50.0 cde	53.3 bcd	

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \le 0.05$; Tukey-Kramer's test). Values are non-transformed data. Significance is based on arcsin [$\sqrt{(x/100)}$] transformed data. Actual mortality percentages are shown after Abbott's (1925) adjustment.

¹Adult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

		Mortality (%)		
Treatment	Rate	24 h	48 h	72 h
Endosulfan (Thionex 3EC [®])	0.5 lb a.i./acre	100.0 ab	100.0 ab	100.0 ab
Malathion ULV (Fyfanon [®] ULV)	12 fl oz/acre	96.0 ab	100.0 ab	100.0 ab
Bifenthrin (Capture 2EC [®])	0.10 lb a.i./acre	92.0 ab	100.0 ab	100.0 ab
Encap. methyl parathion (Penncap M [®])	0.5 lb a.i./acre	72.0 bc	92.0 ab	100.0 a
Oxamyl (Vydate C-LV®)	0.25 lb a.i./acre	76.0 abc	80.0 abc	80.0 abc
Carbaryl (Sevin XLR Plus [®])	1.0 lb a.i./acre	12.0 d	21.0 d	37.0 cd
Cyfluthrin (Baythroid 1.0L®)	0.0205 lb a.i./acre	4.0 d	8.0 d	17.0 d

Table 6. Efficacy of selected insecticides on adult boll weevils using a sprayed leaf-disk bioassay, Lower Rio Grande Valley of Texas, Sept. 20^{th} , 2007 (bioassay 6)¹.

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \le 0.05$; Tukey-Kramer's test). Values are non-transformed data. Significance is based on arcsin [$\sqrt{(x/100)}$] transformed data. Actual mortality percentages are shown after Abbott's (1925) adjustment.

¹Adult boll weevils collected from pheromone traps in the LRGV during cotton post-harvest season.



Fig. 1. Adult boll weevil susceptibility to selected insecticides on treated-leaf bioassays in cotton, LRGV, 2007

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