

ARTHROPOD MANAGEMENT

Comparative Efficacy of Selected Insecticide Alternatives for Boll Weevil (Coleoptera: Curculionidae) Control Using Laboratory Bioassays

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ABSTRACT

The boll weevil, *Anthonomus grandis grandis* (Boheman), is a major pest of cotton (*Gossypium hirsutum* L.), and responsible for an estimated \$300 million in annual losses (National Cotton Council of America 2009, Texas Boll Weevil Eradication Foundation, Inc. 2009). Current boll weevil eradication programs depend on malathion ULV to achieve and maintain eradication status. Should this effective and economical insecticide become unavailable, eradication efforts could be jeopardized. The objective of this project was to evaluate the efficacy of selected insecticides as alternatives to malathion ULV on field collected boll weevils. The study was conducted in the Lower Rio Grande Valley of Texas in 2007. Insecticides included malathion ULV, endosulfan, bifenthrin, encapsulated methyl parathion, oxamyl, carbaryl and cyfluthrin. Malathion ULV was applied using an ULV, controlled-droplet applicator. Other insecticides were applied with a hand-held, CO₂-charged sprayer. Leaf disks were removed from treated cotton, placed in culture plates and infested with individual adult boll weevils. Boll weevil mortality in the malathion ULV, endosulfan, encapsulated methyl parathion and bifenthrin treatments was at or near 100%. Mortality with cyfluthrin and carbaryl was low and inconsistent. Mortality in the oxamyl treatment was intermediate between the two above groups. Highest mortality after 24 h was observed with malathion ULV (97.9%), endosulfan (86.6%) and bifenthrin (80.2%). After 48 h, mortality reached 100% with malathion ULV but was not significantly different from those of encapsulated methyl parathion (96.1%), bifenthrin (95%) and endosulfan (94.9%). Results indicate that malathion ULV is a highly effective mate-

rial for boll weevil control and that encapsulated methyl parathion, bifenthrin and endosulfan also cause high mortality.

The boll weevil, *Anthonomus grandis grandis* (Boheman), is a major cotton pest, responsible for an estimated \$300 million in cotton crop losses every year including \$70 million spent annually on control efforts (National Cotton Council of America 2009, Texas Boll Weevil Eradication Foundation, Inc. 2009). Malathion ULV is currently the only insecticide being used to control the boll weevil 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 Mexico remain in active eradication programs. The Lower Rio Grande Valley of Texas (LRGV) completed its third full season in 2008 under the Texas Boll Weevil Eradication Program, and is not projected to attain functionally-eradicated status for a few years. The continued availability of malathion ULV is critical to boll weevil eradication programs to both achieve and maintain eradication. 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 on field collected boll weevils are limited.

Previous studies provided important information on the efficacy of selected insecticide alternatives on boll weevil infestations (Sparks et al. 1997, Parker and Huffman 1997, Raulston et al. 1998). Although they do not report direct boll weevil mortality over time caused by insecticides, they increased knowledge on boll weevil management by recording the percentage of oviposition-punctured squares and by measuring impact on yields (Sparks et al. 1997, Parker and Huffman 1997, Raulston et al. 1998). Field experiments

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indicated 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). Some research on field efficacy of selected insecticides 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 higher number of punctured squares than did weekly applications of azinphosmethyl. However, fipronil-treated plots also had higher number of total squares. This caused the actual percentage of infested squares to be lower in fipronil-treated plots than in azinphosmethyl-treated plots. The authors concluded that the higher numbers of total squares in fipronil-treated plots probably resulted from product efficacy. They added that the higher numbers of punctured squares likely resulted from a combination of inter-plot movement of boll weevils towards fipronil-treated plots which contained higher number of squares. It was concluded that fipronil-treated plots offered increased opportunities for boll weevil population recruitment and increased attractiveness relative to adjacent plots. Similar results were observed by Sparks et al. (1997) while evaluating fipronil and azinphosmethyl. Fipronil provided the highest residual activity in leaf-disk exposure bioassays. More oviposition-punctured squares were observed in fipronil-treated plants in small replicated plots. Despite this valuable information, fipronil and azinphosmethyl are no longer available as alternatives

to malathion ULV. Previous research increased our understanding of insecticide options for improved boll weevil management and yield protection. They not only illustrate difficulties in controlling field and other environmental variables but also the continued trend for reduction in insecticidal options on which previous efficacy work was conducted. Moreover, information continues to be limited on the ability of currently available insecticides to cause direct and swift mortality, which is the ultimate objective of the boll weevil eradication program. Therefore, there is still a critical need for direct assessment of efficacy corresponding to materials currently labeled for cotton. The objective of these experiments was to directly evaluate efficacies of selected insecticides as alternatives to malathion ULV on native, field collected adult boll weevils using a simple and repeatable laboratory bioassay.

MATERIALS AND METHODS

Insecticides. Selected insecticides evaluated in this study included oxamyl (Vydate[®] C-LV, DuPont Crop Protection, Wilmington, DE), carbaryl (Sevin[®] XLR Plus, Bayer CropScience, Research Triangle Park, NC), encapsulated methyl parathion (PennCap[®] M, United Phosphorus, Inc., King of Prussia, PA), malathion ULV (Fyfanon[®] ULV AG, Cheminova, Wayne, NJ), bifenthrin (Capture[®] 2EC, FMC Corporation, Philadelphia, PA), endosulfan (Thionex[®] 3EC, Makhteshim Agan of North America, Inc., New York, NY) and cyfluthrin (Baythroid[®] 1.0L, Bayer CropScience, Research Triangle Park, NC). The insecticide rates were applied in accordance with the label and are included in Tables 1 through 6.

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

Treatment	Rate	Mortality (%)		
		24 h	48 h	72 h
Encap. methyl parathion	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	100.0 a	100.0 a	100.0 a
Endosulfan	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	100.0 a	100.0 a	100.0 a
Bifenthrin	112 g ai ha ⁻¹ (0.10 lb a.i./acre)	97.2 a	97.2 a	97.2 a
Oxamyl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	36.1 bc	36.1 c	50.0 b
Cyfluthrin	23 g ai ha ⁻¹ (0.0205 lb a.i./acre)	0.0 c	8.3 c	11.1 bc
Carbaryl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	5.6 c	5.6 c	8.3 c

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \leq 0.05$; 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.

^zAdult boll weevils collected from pheromone traps in the LRGV prior to cotton squaring.

Table 2. Efficacy of selected insecticides on adult boll weevils using a sprayed leaf-disk bioassay, Lower Rio Grande Valley of Texas, 20 June 2007 (bioassay 2)^z.

Treatment	Rate	Mortality (%)		
		24 h	48 h	72 h
Endosulfan	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	100.0 a	100.0 a	100.0 a
Malathion ULV	1035 g ai ha ⁻¹ (12 fl oz/acre)	92.0 ab	100.0 a	100.0 a
Bifenthrin	112 g ai ha ⁻¹ (0.10 lb a.i./acre)	88.0 abc	100.0 a	100.0 a
Encap. methyl parathion	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	76.0 bc	96.0 a	100.0 a
Oxamyl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	60.0 c	64.0 bc	66.0 bc
Carbaryl	560 g ai ha ⁻¹ (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 ($P \leq 0.05$; 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.

^zAdult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

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

Treatment	Rate	Mortality (%)		
		24 h	48 h	72 h
Endosulfan	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	100.0 a	100.0 a	100.0 a
Malathion ULV	1035 g ai ha ⁻¹ (12 fl oz/acre)	100.0 a	100.0 a	100.0 a
Bifenthrin	112 g ai ha ⁻¹ (0.10 lb a.i./acre)	64.2 bc	90.0 ab	100.0 a
Encap. methyl parathion	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	61.7 bc	96.7 a	96.7 a
Oxamyl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	16.7 d-g	31.7 c-f	50.0 cd
Carbaryl	1120 g ai ha ⁻¹ (1.0 lb a.i./acre)	6.7 efg	16.7 d-g	36.7 cde
Cyfluthrin	23 g ai ha ⁻¹ (0.0205 lb a.i./acre)	3.3 fg	10.0 efg	24.1 d-g
Carbaryl	560 g ai ha ⁻¹ (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 ($P \leq 0.05$; 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.

^zAdult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

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

Treatment	Rate	Mortality (%)		
		24 h	48 h	72 h
Malathion ULV	1035 g ai ha ⁻¹ (12 fl oz/acre)	100.0 a	100.0 a	100.0 a
Encap. methyl parathion	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	50.0 b-e	75.0 abc	100.0 a
Bifenthrin	112 g ai ha ⁻¹ (0.10 lb a.i./acre)	40.0 c-f	70.0 a-d	85.0 ab
Cyfluthrin	23 g ai ha ⁻¹ (0.0205 lb a.i./acre)	45.0 b-e	65.0 a-d	75.0 a-d
Endosulfan	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	50.0 b-e	60.0 a-d	70.0 a-d
Oxamyl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	20.0 def	25.0 def	30.0 c-f
Carbaryl	1120 g ai ha ⁻¹ (1.0 lb a.i./acre)	0.0 f	5.0 ef	5.0 ef

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \leq 0.05$; 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.

^zAdult 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, 17 July 2007 (bioassay 5)^z.

Treatment	Rate	Mortality (%)		
		24 h	48 h	72 h
Malathion ULV	1035 g ai ha ⁻¹ (12 fl oz/acre)	100.0 a	100.0 a	100.0 a
Bifenthrin	112 g ai ha ⁻¹ (0.10 lb a.i./acre)	86.1 abc	100.0 a	100.0 a
Encap. methyl parathion	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	60.0 bcd	100.0 a	100.0 a
Endosulfan	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	57.2 cd	93.9 ab	93.9 ab
Cyfluthrin	23 g ai ha ⁻¹ (0.0205 lb a.i./acre)	33.3 def	50.0 cde	64.4 a-d
Oxamyl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	34.5 def	46.1 cde	62.8 bcd
Carbaryl	1120 g ai ha ⁻¹ (1.0 lb a.i./acre)	11.1 ef	53.9 cd	65.6 bcd
Carbaryl	560 g ai ha ⁻¹ (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 ($P \leq 0.05$; 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.

^zAdult boll weevils reared out from infested squares collected from commercial cotton fields in the LRGV.

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

Treatment	Rate	Mortality (%)		
		24 h	48 h	72 h
Endosulfan	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	100.0 ab	100.0 ab	100.0 ab
Malathion ULV	1035 g ai ha ⁻¹ (12 fl oz/acre)	96.0 ab	100.0 ab	100.0 ab
Bifenthrin	112 g ai ha ⁻¹ (0.10 lb a.i./acre)	92.0 ab	100.0 ab	100.0 ab
Encap. methyl parathion	560 g ai ha ⁻¹ (0.5 lb a.i./acre)	72.0 bc	92.0 ab	100.0 ab
Oxamyl	280 g ai ha ⁻¹ (0.25 lb a.i./acre)	76.0 abc	80.0 abc	80.0 abc
Carbaryl	1120 g ai ha ⁻¹ (1.0 lb a.i./acre)	12.0 d	21.0 d	37.0 cd
Cyfluthrin	23 g ai ha ⁻¹ (0.0205 lb a.i./acre)	4.0 d	8.0 d	17.0 d

Means within columns and across rows are significantly different when followed by a different small-case letter ($P \leq 0.05$; 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.

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

Boll weevil source. The first and sixth bioassays (on 6 April 2007 and 20 September 2007, respectively) were conducted using adult boll weevils from pheromone traps because commercial cotton fields in the LRGV either had no infested squares to rear the adults (6 April) or were already harvested (20 September). Overwintered boll weevils were used in the first bioassay (6 April). Bioassays conducted on 20 September were performed on adult boll weevils from the post-harvest season of the LRGV. Trapping location, setup and maintenance for bioassays on 6 April and 20 September 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 10% sucrose water in moistened cotton wicks. Bioassays were conducted one day later after selecting only healthy boll weevil adults. Bioassays conducted on 20 June, 25 June, 30 June, and 17 July were conducted on adult boll weevils reared from infested squares collected in the LRGV during the cotton squaring season. 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 15-cm-diameter Petri dishes lined with a thin layer of vermiculite moistened with reverse osmosis 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 10% 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 cultivar "Stoneville 474". Cotton was planted in the greenhouse at different intervals during the study to be used as leaf source for bioassays. Separate sets of plants were used as 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 (6 April), 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 140 L ha⁻¹ (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 140 L ha⁻¹ (15 gpa) at 331 kPa (48 psi) through four Tee Jet 8002VS nozzles. Malathion ULV was applied at 1035 g ai ha⁻¹ (12 fl 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 "Falcon 3046" (BD Falcon, Billerica, MA) tissue culture plates equipped with 6, 35 mm diameter wells. The bottom of each culture well was lined with a moistened paper filter. Sprayed cotton leaf disks were placed in 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 adult 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 h 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 s 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 [$\arcsin \sqrt{(x/100)}$] 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 for each bioassay are presented in Tables 1 through 6 to differentiate adult boll weevil mortality as the cotton season progressed.

RESULTS AND DISCUSSION

Six sprayed leaf-disk bioassays were conducted during the 2007 cotton season at the LRGV. These assays were conducted on overwintered adult boll weevils collected from pheromone traps along cotton fields (6 April bioassay, Table 1), from adult boll weevils reared from infested squares (20 June, 25 June, 30 June, and 17 July bioassays, Tables 2-5) and adult boll weevils from pheromone traps during the post-harvest season (20 September 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 each bioassay. Efficacies of cyfluthrin and different rates of carbaryl were low and inconsistent throughout the bioassays. A season average of boll weevil mortality revealed that susceptibility to oxamyl was intermediate between the two above groups (Fig. 1). An intermittent drizzling event was experienced during the outdoor plant spraying for the fourth bioassay on 30 June. Despite this event, high mortality (at or near 100%) was consistently observed in the Malathion ULV and the encapsulated methyl parathion treatments (Fig. 2). Performance of cyfluthrin did not seem to be affected by the drizzling event. However, overall cyfluthrin performance was poor. Conversely, bifenthrin, endosulfan, oxamyl

and carbaryl did not appear to be as rainfast as other materials, as indicated by a decline in boll weevil mortality in the 30 June bioassay (Fig. 2). After removing the 30 June bioassay data, boll weevil susceptibility to each evaluated insecticide appears to be constant across the season (Fig. 2). Malathion ULV was the only treatment to accomplish 100% boll weevil mortality in all bioassays after 48 h of treated leaf-disk exposure (Fig. 1). Significantly higher boll weevil mortality after 24 h was observed in the malathion ULV, endosulfan and bifenthrin treatments (97.9%, 86.6% and 80.2%, respectively, see Fig. 1). At 48 h 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 h of exposure (Fig. 1).

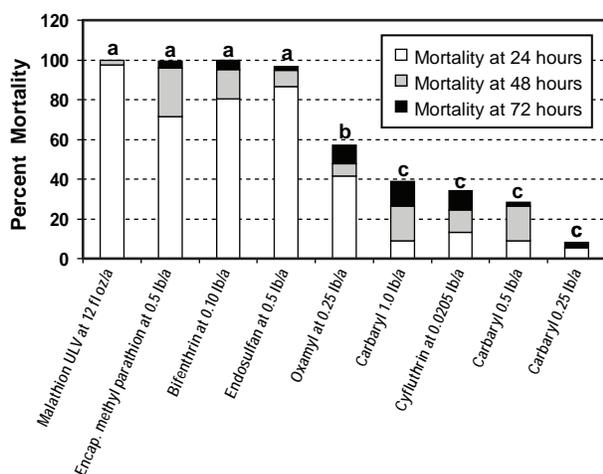


Fig. 1. Season average adult boll weevil susceptibility to selected insecticides on treated-leaf bioassays in cotton, LRGV, 2007

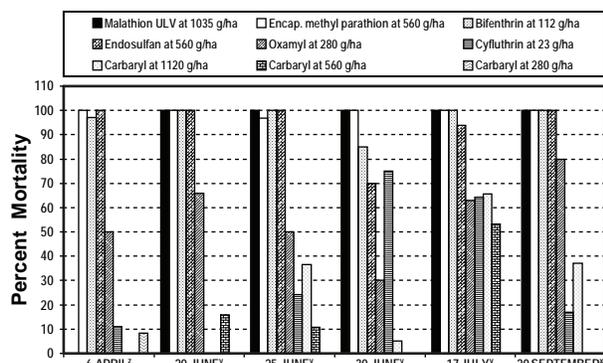


Fig. 2. Adult boll weevil mortality after 72 h of treated-leaf exposure to selected insecticides in cotton, LRGV, 2007 ²Pre-squaring season, overwintered boll weevils; ³Squaring season; ⁴Post-harvest season

Data analysis from these experiments did not reveal 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 evaluated, malathion ULV still remains highly effective for boll weevil control and that encapsulated methyl parathion, bifenthrin and endosulfan also are effective in causing high mortality of adult boll weevils. Of the top performing insecticides, bifenthrin at 112 g ha⁻¹ and endosulfan at 560 g ha⁻¹ did not appear to be as rainfast as malathion ULV at 1035 g ha⁻¹ and encapsulated methyl parathion at 560 g ha⁻¹, which could be a limiting factor during rainy cotton squaring season. However, additional studies are suggested to evaluate rainfastness characteristics in addition to efficacy, economics and environmental profile of alternative insecticides on boll weevil eradication programs.

ACKNOWLEDGEMENTS

This project was supported by Cotton Incorporated, Texas AgriLife Extension Service, Texas AgriLife Research, USDA-ARS, TBWEF and the Technical Advisory Committee for the TBWEF.

DISCLAIMER

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