INVESTIGATION OF PHEROMONE-BASED FACTORS THAT MAY REDUCE CAPTURE OF BOLL WEEVILS IN TRAPS J. K. Westbrook C. P. C. Suh USDA, ARS College Station, TX

<u>Abstract</u>

Boll weevil (*Anthonomus grandis* Boheman) eradication programs rely almost exclusively on pheromone traps to detect weevils, assess populations, and indicate the need for insecticide treatment. However, instances have been reported recently in Medina Co., TX where field infestations occur without prior detection by pheromone traps around these fields. We investigated the initial quality and release rates of pheromone from lure dispensers, and the pheromone blend produced by weevils in Medina Co. within the Winter Garden cotton production area of Texas. Mean initial content of grandlure in dispensers ranged from 7.2 mg to 12.0 mg during seven evaluation periods. Overall, the mean release rate of grandlure ranged from 1.54 mg d⁻¹ at 1 d to 0.24 mg d⁻¹ at 11-14 d of aging. Weevils collected in pheromone traps, collected off of cotton plants, and reared from infested cotton bolls produced similar ratios (44:43:2:11) of the four components of grandlure. The results suggest that initial lure quality and release rate of grandlure, but not pheromone blend produced by weevils, may have contributed to isolated cases of reduced capture of weevils in traps. This information will aid eradication program managers in developing responses to the use of pheromone lures and traps for detection of remnant weevil populations.

Introduction

Boll weevil (*Anthonomus grandis* Boheman) eradication programs have achieved eradication in greater than 94% of the cotton production areas in the U.S. The success of the eradication programs is largely attributed to the use of pheromone traps for detection of weevil populations and for triggering insecticide applications. However, there are instances where substantial weevil infestations are discovered in fields, but respective traps fail to detect weevils during preceding week(s). Failure of pheromone traps to detect incipient weevil populations in adjacent fields has hindered eradication progress in Medina Co., TX. One factor that may have contributed to these trap failures is the use of sub-standard quality lures. It also has been speculated that a proportion of the weevil population may be producing and responding to a unique pheromone "blend", and such weevils may not be responsive to the standard pheromone lure. Our objectives were to quantify the amount and composition of grandlure released from lure dispensers used by the Texas Boll Weevil Eradication Foundation, and to evaluate and compare the pheromone composition of trap-captured and field-collected weevils from Medina County.

Materials and Methods

Lure Properties

Lure dispenser evaluations were conducted during seven periods between 21 April – 23 June, 2009. Boll weevil pheromone traps (Technical Precision Plastics, Mebane, NC) and a Campbell 21XL (Campbell Scientific, Inc., Logan, UT) weather station were situated in an open environment at the USDA-ARS Southern Plains Agricultural Research Center in College Station, TX. Nine replicates of 10-mg grandlure dispensers (Hercon Environmental Corp., Emigsville, PA) were collected from traps at 1 d, 2 d, 3 d, 7 d, 10 d, and 14 d of field aging during each evaluation period. Nine un-aged (0 d) lure dispensers for each evaluation period and the field-aged lure dispensers were analyzed by gas chromatography to quantify the residual content of grandlure in each dispenser. To account for any differences in the weight of the lure dispensers, residual values as well as release rates of grandlure content were normalized by the weight of the respective dispensers. Release rates of grandlure from traps were analyzed relative to aging duration and evaluation periods.

Pheromone Production

Trap-captured weevils were collected between 14 April and 12 May, and were fed squares (6-9 mm diam with bracts intact; 1 square per weevil per day) daily for 6 d to promote pheromone production. Field-collected adults and those reared from oviposition-punctured squares were collected between 10 and 22 June, and were fed squares for 6 to 8 d prior to pheromone collection. Weevils were held at $29.4 \pm 1^{\circ}$ C and a photoperiod of 14:11 (L:D) h during the feeding periods.

Following the feeding period, weevils were transferred individually into pheromone collection vessels and pheromone was collected over a 24-h period. Pheromone contents were analyzed on a Shimadzu GC-17A equipped with dual columns and detectors. Pheromone content from each weevil was injected and analyzed on both columns. Estimates from duplicate injections were averaged before statistical analysis, and total pheromone in each sample was determined by adding the quantities of the four components. Weevils that produced <10 μ g were considered "abnormal" and were excluded from the analysis.

Results and Discussion

Lure Properties

The mean weight of the grandlure dispensers in each evaluation period ranged from 0.6608 g to 0.8566 g (Table 1), and the frequency of individual weights were represented by a distinct bimodal distribution (Fig. 1). The initial mean content of total grandlure in the lure dispensers ranged from 7.2 mg to 12.0 mg. Atmospheric conditions during the seven aging periods were described by ranges of mean air temperature (23.3 to 30.0°C), mean relative humidity (76 to 95%), and mean wind speed (2.5 to 3.9 m s⁻¹) (Table 2). Mean daily release of grandlure ranged from 1.54 mg d⁻¹ at 1 d to 0.24 mg d⁻¹ at 11-14 d (Table 3). Expressed as a cumulative percentage of the initial content, the release of grandlure increased from 15% at 1 d to 57% at 14 d (Table 4). The mean blend of grandlure components I-IV had an initial ratio of 33:41:14:13, and minor variations were evident relative to aging duration and evaluation period.

Table 1. Mean \pm SD weight (g) of 10-mg grandlure dispensers used in field aging study at the USDA-ARS Southern Plains Agricultural Research Center in College Station, TX, in 2009.

Evaluation	Dates	Ν	Weight (g)
Period			
Ι	4/21 - 5/5	63	0.6608 ± 0.0184
II	4/28 - 5/12	63	0.7462 ± 0.0982
III	5/5 - 5/19	63	0.8558 ± 0.0207
IV	5/12 - 5/26	63	0.6990 ± 0.0754
V	5/26 - 6/9	63	0.7266 ± 0.0900
VI	6/2 - 6/16	63	0.8566 ± 0.0182
VII	6/9 - 6/23	63	0.8088 ± 0.0400

Evaluation Periods I, III and VI included lure dispensers from a single package. Evaluation Periods II, IV, V, and VII included lure dispensers from two packages, which were from the same production lot.

Table 2. Minimum, mean, and maximum air temperature and wind speed at the USDA-ARS Southern Plains Agricultural Research Center in College Station, TX, in 2009.

	Air Temperature (C)		Wind S	Speed (m s ⁻¹) ^{\dagger}	Relative	Humidity (%)	
Evaluation	Mean	Min - Max	Mean	Min - Max	Mean	Min - Max	
Period							
Ι	23.3	14.2 - 32.4	3.9	0.4 - 10.3	92	38 - 100	
II	24.8	14.2 - 33.5	3.4	0.4 - 9.5	95	62 - 100	
III	24.9	11.5 - 33.5	3.4	0.4 - 9.5	90	41 - 100	
IV	23.5	11.5 - 32.5	2.5	0.4 - 9.5	85	41 - 100	
V	26.3	16.3 - 35.2	2.6	0.4 - 7.4	78	34 - 100	
VI	28.2	16.3 - 37.7	3.5	0.4 - 9.9	81	45 - 100	
VII	30.0	22.9 - 38.5	3.9	0.4 - 9.9	76	43 - 100	
$\dot{\tau}$ The start-up (minimum) wind speed value is 0.4 m s ⁻¹							



Figure 1. Distribution of weights of 10-mg grandlure dispensers used in a field aging study at College Station, TX, April 21 – June 23, 2009.

Table 3. Daily mean release rate (mg d⁻¹) of total grandlure from 10-mg grandlure dispensers aged in the field at the USDA-ARS Southern Plains Agricultural Research Center in College Station, TX, in 2009.

Aging								
(d)	Ι	II	III	IV	V	VI	VII	Overall
1	1.00	0.90	1.00	2.70	1.30	1.90	2.00	1.54 ± 0.68
2	0.40	0.30	1.20	0.60	0.30	0.90^{b}	2.30	0.86 ± 0.72
3	0.50	0.30	0.50	0.50	0.60	0.10^{b}	0.70	0.46 ± 0.20
4 - 7	0.25	0.08	0.38	0.18	0.15 ^{<i>a</i>}	0.50	0.45	0.28 ± 0.16
8 -10	0.10	0.17	0.23	0.20	-0.53^{a}	0.40	0.30	0.12 ± 0.31
11-14	0.10	0.23	0.10	0.23	0.35 ^{<i>a</i>}	0.38	0.33	0.24 ± 0.11

^{*a*} Included lure dispensers that were taken from a second bag containing dispensers with a mean weight of 0.8564 g, compared to the mean weight (0.6665 g) of lures aged for 0-3 d and some for 7 d in Evaluation Period V.

 b^{b} The grandlure content of one lure dispenser was markedly lower than that of the other eight dispensers aged for

2 d in Evaluation Period VI.

Table 4. Mean cumulative release (%) of total grandlure per gram-weight from 10-mg grandlure dispensers aged in the field at the USDA-ARS Southern Plains Agricultural Research Center in College Station, TX, in 2009.

Aging			Εv	aluation Peri	od			_
(d)	Ι	II	III	IV	V	VI	VII	Overall
1	14	15	10	14	17	16	16	15 ± 2
2	18	19	22	14	22	23^b	30	21 ± 5
3	24	23	26	23	29	22	36	26 ± 5
7	38	42	44	32	42^{a}	40	53	42 ± 6
10	44	50	50	41	35 ^{<i>a</i>}	50	61	47 ± 8
14	50	59	55	52	49^a	63	73	57 ± 9

^{*a*} Included lure dispensers that were taken from a second bag containing dispensers with a mean weight of 0.8564 g, compared to the mean weight (0.6665 g) of lures aged for 0-3 d and some for 7 d in Cycle V.

^b The grandlure content of one lure dispenser was markedly lower than that of the other eight dispensers aged for 2 d in Cycle VI.

Pheromone Production

All weevils produced >10 μ g of pheromone with the exception of one weevil (Run 3, reared from infested square) that produced only 2.9 μ g of pheromone. Trap-captured weevils produced a range of 20.4 to 259.1 μ g of pheromone, while adults reared from infested squares produced between 2.9 to 200.2 μ g. Adults collected directly

off plants produced between 11.9 to 182.5 μ g. The component ratios (I:II:III:IV) for the three sources of boll weevils were nearly identical to one another (Table 5). The component ratio remained consistent among field sites (Table 6) and among experimental runs (Table 7), and was similar to the ratio observed in a previous study that used weevils collected from Hill, Limestone, and Hidalgo counties of Texas.

Of particular interest is the amount of pheromone released by adults (Table 5) in relation to the amount of pheromone released from lures after one week of aging (Table 3). Based on maximal values, one weevil is capable of releasing as much pheromone as a lure after 7 d of aging. Competition from pheromone-producing weevils is thought to be one of the major factors responsible for the dramatic decline in captures of weevils in traps during the mid-growing season. Given the high level of pheromone lures, subsequently reducing the effectiveness of pheromone traps.

Table 5. Overall mean \pm SD proportions of pheromone represented by the individual components and the average amount of pheromone produced from boll weevils obtained from different sources, Medina Co., TX, 2009.

Source of adults a			Pheromone			
Source of adults	n	Ι	II	III	IV	(µg)
Pheromone traps	64	0.43 ± 0.02	0.43 ± 0.02	0.03 ± 0.01	0.11 ± 0.01	87.1
Field collection	6	0.44 ± 0.02	0.43 ± 0.03	0.03 ± 0.01	0.10 ± 0.01	98.0
Infested squares	128	0.44 ± 0.02	0.43 ± 0.03	0.02 ± 0.01	0.11 ± 0.01	107.4

^{*a*} Weevils from traps collected between 14 April and 12 May; live adults collected from fields on 10 and 15 June; adults reared from oviposition-punctured squares (infested squares) collected between 10 and 22 June. ^{*b*} Summation of the four proportions within a row may not equal 1 due to rounding-off values to the nearest hundredth.

Table 6. Mean \pm SD proportions of total pheromone represented by the individual components and average amount of pheromone produced by boll weevils reared from oviposition-punctured squares collected from different fields, Medina Co., TX, 2009.

Field ID ^{<i>a</i>}	14		Pheromone component ^b					
	n	Ι	II	III	IV	(µg)		
Carpe Diem	21	0.43 ± 0.02	0.44 ± 0.02	0.02 ± 0.01	0.11 ± 0.01	80.9		
CR 482	42	0.43 ± 0.02	0.44 ± 0.03	0.02 ± 0.01	0.11 ± 0.01	103.5		
Lamons	53	0.44 ± 0.02	0.42 ± 0.02	0.02 ± 0.01	0.11 ± 0.01	120.8		
Stella	10	0.44 ± 0.01	0.43 ± 0.01	0.03 ± 0.01	0.11 ± 0.01	104.3		
Weiblens	2	0.45 ± 0.01	0.40 ± 0.01	0.04 ± 0.01	0.11 ± 0.01	72.4		

^{*a*} Approx. GPS coordinates for each field: Carpe Diem, 29.453822°N, -98.900973°W; CR 482, 29.359789°N, -98.813854°W; Lamons, 29.278558°N, -98.855780°W; Stella, 29.298377°N, -98.836230°W; and Weiblens, 29.390598°N, -98.964862°W.

^b Summation of the four proportions within a row may not equal 1 due to rounding-off values to the nearest hundredth.

<u>Summary</u>

Minor changes in the blend of grandlure components (grandlure I:II:III:IV) among aged lure dispensers may not have significantly impacted boll weevil response. However, large differences between individual lure dispenser weights may have created situations where an adequate dose to attract weevils was not released from a baited trap, especially after lures had been aged for several days. Because the weights of individual grandlure dispensers were not normally distributed, the mean dose of total grandlure for un-aged dispensers in each aging period varied from 72% to 120% of the target value of 10 mg. Although the data are not presented here, we observed microscale differences in air temperature and wind speed in Medina County that may contribute to local differences in release rates and diffusion of grandlure from dispensers. Given the similarities in component ratios among the three sources of weevils, we found no evidence of weevils producing a unique pheromone blend. In light of these findings, our results suggest research efforts should focus on other factors that can influence captures of weevils in pheromone traps (e.g., weather conditions, lure quality, trap placement, etc.).

Table 7. Mean \pm SD proportions of total pheromone represented by the individual components and average amount of pheromone produced during each experimental run for trap- and field-collected boll weevils, Medina Co., TX, 2009.

Source of adults a	Dun	74		Pheromone			
Source of adults	Kull	n	Ι	II	III	IV	(µg)
Pheromone traps	1	16	0.44	0.43	0.03	0.10	68.7
	2	16	0.43	0.43	0.03	0.11	72.3
	3	16	0.44	0.42	0.03	0.11	92.3
	4	16	0.42	0.43	0.03	0.12	115.2
Field-collected	1*	5	0.43	0.44	0.02	0.10	96.7
	2*	4	0.45	0.41	0.04	0.10	128.8
	3	5	0.45	0.42	0.03	0.10	80.6
	4	16	0.44	0.44	0.02	0.11	129.0
	5	16	0.45	0.42	0.02	0.11	125.5
	6	16	0.44	0.42	0.02	0.11	84.4
	7	16	0.44	0.43	0.02	0.11	110.2
	8	16	0.44	0.42	0.02	0.12	102.1
	9	15	0.44	0.43	0.02	0.11	123.6
	10	16	0.44	0.43	0.03	0.11	102.3
	11	16	0.43	0.43	0.02	0.11	85.9

^{*a*} Field-collected adults consist of adults collected directly off plants as well as those reared from infested squares. ^{*b*} Summation of the four proportions within a row may not equal 1 due to rounding-off values to the nearest hundredth.

* All weevils in Run 1 and 1 weevil in Run 2 were live adults collected directly off plants. The remaining runs consisted entirely of adults reared from oviposition-punctured squares.

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