THRIPS[1] SPECIES ASSOCIATED WITH COTTON IN THE NORTHERN TEXAS ROLLING PLAINS

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ABSTRACT

Yellow and blue sticky cup traps were used to monitor thrips in wheat and cotton. Plant washing was used to determine the actual thrips species infesting cotton. Ten species of thrips were collected on sticky traps in wheat, and eight species were collected on sticky traps in cotton. However, only six species of thrips were recovered from cotton using the plant washing technique. These six species were *Frankliniella exigua* (Hood), *F. fusca* (Hinds), *F. genuina* Hood, *F. occidentalis* (Pergande), *F. tritici* (Fitch) and *Thrips tabaci* Lindeman. *Frankliniella occidentalis* was the most abundant species found on traps and in plant washing. *Frankliniella exigua* and *F. genuina* were found in plant wash samples, but neither was recovered from the sticky traps. The species capture data indicate that sticky traps in cotton and wheat did not adequately indicate the species composition of thrips that infest cotton. Additionally, thrips captures on sticky traps in wheat were not correlated with captures on sticky traps in cotton, and captures on sticky traps in cotton were not correlated with numbers of thrips recovered from plant washing.

INTRODUCTION

The thrips species complex reduced average cotton production in the U.S. and Texas by 0.62% and 1.21%, respectively, during 2001 and 2002 (Williams 2002, 2003). In Texas, the losses attributable to thrips injury are highly variable. For example, Harp and Turner (1976) reported that thrips did not reduce yield in the Texas Blacklands. However, in the Texas High Plains, Leser and Vandiver (2003) reported that yield losses averaged 21% in irrigated cotton without thrips treatment and maturity was delayed by several days. Texas Cooperative Extension guidelines for insect control suggest that treatments may be justified when there is an average of one thrips for each true leaf on the plant (Baugh et al. 2004).

In addition to actual thrips density on the cotton plant, species composition influences efficacy of insecticides and the amount of damage inflicted on seedlings. Western flower thrips, *Frankliniella occidentalis* (Pergande), is tolerant to many insecticides, and control failures have been reported (Burris et al. 2000, Kharboutli and Allen 2001). Faircloth et al. (2001) reported that damage caused by western flower thrips was greater than that caused by tobacco thrips, *Frankliniella fusca* (Hinds), on cotton seedlings, even though the reproductive potential of tobacco thrips was higher than that of western flower thrips.

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The thrips species complex infesting cotton varies across the U. S. cottonbelt. In North Carolina, Faircloth et al. (2002) found five thrips species in a three-year study, and the tobacco thrips and soybean thrips, *Neohydatothrips variabilis* (Beach), were the most common, representing an average of 62 and 22% of the population, respectively. In Oklahoma, Karner and Cole (1992) found seven thrips species in cotton prior to flowering, but the western flower thrips and flower thrips, *Frankliniella tritici* (Fitch), were the most common and were collected in 91% and 24% of the samples, respectively. In a multi-year, multi-state survey, Burris et al. (2000) indicated that western flower thrips could be considered a new pest in cotton in the south central cottonbelt as compared to tobacco thrips and flower thrips, and they indicated that soybean thrips were not found in surveys conducted in the mid-1950's. Tobacco thrips were the most common species collected from 1996 to 1999 in Alabama, Arkansas, Georgia, Louisiana, Mississippi, and Tennessee, except at two locations in Georgia in 1999 where western flower thrips was most abundant (Cook et al. 2003).

Thrips species composition varies among locations and years within a state. Cook et al. (2000) reported that tobacco thrips was the most common species in Louisiana in a three-year study, but tobacco thrips composition ranged from 39% to >90% depending on location and year. Kharboutli and Allen (2001) reported that tobacco thrips was the dominant species in 1999, but the western flower thrips was dominant in 2000 in southeast Arkansas.

Objectives of this study were to (1) determine thrips species composition collected from colored sticky traps in wheat and cotton versus species composition actually present in pre-squaring cotton, and (2) monitor thrips movement on sticky traps.

METHODS

Studies were conducted at the Texas Agricultural Experiment Stations located at Chillicothe (Hardeman Co.), Munday (Knox Co.) and Vernon (Wilbarger Co.) in the northern Texas Rolling Plains. Studies were conducted for three years, 2000-2002, at each location. At Chillicothe and Munday, thrips monitoring in wheat began each year in mid April and continued through late May. Monitoring in cotton was initiated the first week in June and continued through late June. Monitoring at Vernon began in May before cotton planting and continued through June. Wheat was planted in the fall of the preceding year at all locations, and cotton was planted about the third week of May each year of the study.

Sticky cup traps were made from blue and yellow 710-ml (24 oz) plastic drink cups (Solo Cup Co., Urbana, IL). A thin layer of the sticky compound Tanglefoot (The Tanglefoot Co., Grand Rapids, MI) was coated onto the surface of the cups with a paint brush. Six equal areas measuring 2.3 x 2.3 cm each were outlined on the cup surface prior to coating with Tanglefoot. One grid was near the bottom of the cup; three grids were in the middle of the cup, and two grids were at the top of the cup. The grids were randomly spaced around the circumference of each cup. These six grid areas represented about 10% of the surface area of the cup, and thrips were counted visually within the six grids only. The cup was mounted upside down on top of a wooden post with the cup 1 m above ground. A screw was placed in the top of the post, and a small hole was drilled into the bottom of the cup; the screw protruded through the hole to keep the cup from blowing off the post. One cup of each color, spaced 10 m apart and about 20 m into the field, was placed in each of three wheat fields or three cotton fields at Chillicothe and Munday, and thrips were counted once each week. One trap of each color was used at Vernon, and these traps were inspected daily. Traps at Vernon were maintained on cleared ground adjacent to wheat throughout the sample period. Sticky traps were monitored for thrips numbers during 2000-2002 at all three locations. Cups were replaced with fresh cups after each inspection at all locations.

A toothpick was used to remove two samples of thrips (ca. 30 total thrips) from each trap. The toothpick with thrips and Tanglefoot was placed into a small capped jar containing Bioshield Paint Thinner #24 (a mixture of isopar and orange peel oil, Eco Design Co., Santa Fe, NM), which acted to dissolve the Tanglefoot and free the thrips for later identification. Thrips samples from the three blue sticky traps were pooled into one jar on each date, and the thrips from the three yellow sticky traps were pooled into a separate jar. Thrips from Munday and Chillicothe were kept separate. At the end of the summer, the thrips were removed from the jars containing the solvent and placed into screw top vials containing a 70:30 ethanol-water solution and labeled with sample date, crop, location, and trap color. Thrips were not collected from the traps inspected daily at Vernon.

A plant washing technique (Rummel and Arnold 1989, Burris et al. 1990) was used to collect thrips from cotton plants growing in the fields. Sample size was 15 plants with five plants taken from each of three locations in a field, and samples were taken from three cotton fields at Chillicothe and at Munday. Samples were taken weekly on the same dates that sticky traps were inspected. As a plant was pulled or cut at the soil surface, it was placed immediately into a 950-ml jar containing 100 ml of a 70:30 ethanol-water solution. The jar cap was placed on the jar after every five-plant sample, and the jar was shaken vigorously. Samples were returned to the laboratory at Vernon where the jars were filled half full with water, and then 10 ml of bleach and one drop of detergent was added. The jar contents were again shaken vigorously for 30 sec and then poured into a No. 25 (707µm) sieve on top of a No. 230 (63µm) sieve. The plants in the top sieve were rinsed with tap water and the plants were discarded. The thrips and other residue in the bottom sieve were then washed with a 70:30 ethanol-water solution into a Buchner funnel lined with a coffee filter. The liquid in the funnel was suctioned off with air. The filter paper was inspected using a dissecting microscope at 10-20X, and the thrips were counted, removed and placed into a labeled vial containing a 70:30 ethanol-water solution. Thrips from each collection date from Chillicothe and Munday were kept separately, but the samples from the three fields within a location were pooled into the same vial. Vials were labeled with sample date, crop (cotton only), and location. Thrips from sticky traps were identified to species in 2000 and 2001, but thrips from plant washing were identified all three years, 2000-2002. Thrips specimens were identified by C. L. Cole.

Data were analyzed to determine species composition on blue and yellow sticky traps in wheat and cotton and on cotton plants at each location. Correlations between sticky trap captures and actual numbers of thrips on cotton plants and between last captures in wheat and first captures in cotton on sticky traps were determined also. Analysis of variance was used to compare captures of *F. occidentalis* on blue and yellow sticky traps using location (n=2), years (n=2), and trap color as main effects (Anonymous 2000). Thrips numbers were transformed (log + 1) for correlation analysis. The Shannon-Weaver diversity index (Price 1975) was calculated to compare species composition on cotton plants at Chillicothe and Munday.

RESULTS AND DISCUSSION

Ten species of thrips were captured in wheat on the blue and yellow sticky traps located in Hardeman and Knox counties (Table 1). The western flower thrips, *F. occidentalis*, was the dominant species captured on both trap colors at both locations, accounting for 54 - 99% of the total numbers captured. Numbers of tobacco thrips, *F. fusca*, were low in Hardeman Co., but this species represented 43% of the captures on yellow sticky traps in Knox Co. *Chirothrips simplex* Moulton was captured only on blue traps, while the flower thrips, *F. tritici*, was captured only on yellow traps. The soybean thrips, *N. variabilis*, was captured only in Knox Co. on both trap colors, and low numbers of the onion thrips, *Thrips tabaci* Lindeman, were captured in both

TABLE 1. Percentage of thrips species captured in wheat on blue and yellow sticky traps, 2000-2001.

Species	Hardeman Co.		Knox Co.	
	Blue Trap	Yellow Trap	Blue Trap	Yellow Trap
Aeolothrips duvali Moulton	0.00	1.01	0.05	0.73
Chirothrips simplex Hood	0.04	0.00	0.05	0.00
Frankliniella fusca (Hinds)	0.00	0.61	0.20	43.00
F. occidentalis (Pergande)	99.33	95.56	98.93	54.48
F. tritici (Fitch)	0.00	1.92	0.00	0.08
Microcephalothrips abdominalis (Crawford)	0.04	0.40	0.00	0.33
Neohydatothrips variabilis (Beach)	0.00	0.00	0.05	0.08
Plesiothrips ayarsi Stannard	0.00	0.10	0.00	0.00
Thrips tabaci Lindeman	0.58	0.40	0.66	1.30
T. winnemanae Hood	0.00	0.00	0.05	0.00
N	2245	990	1962	1228

counties on both trap colors. DuRant et al. (1994) collected thrips directly from wheat tillers in South Carolina and found *F. tritici*, *F. occidentalis*, and *F. fusca*, although *Limothrips cerealium* (Haliday) was the most abundant in their samples.

Eight species of thrips were captured in cotton on the yellow and blue sticky traps (Table 2). As in wheat, *F. occindentalis* was the most abundant and represented 89-99% of the total number of thrips captured. *Thrips tabaci* was the second most abundant species, but this thrips represented an average of only 3.4% of the captures. *Plesiothrips ayarsi* Stannard was captured on yellow traps only in both counties.

Six species of thrips were recovered from cotton plants during the three years of plant wash sampling (Table 3). The four most abundant species were *F. occidentalis* (51.56%), *F. fusca* (21.87%), *T. tabaci* (14.49%), and *Frankliniella exigua* (Hood) (11.21%). *Frankliniella genuina* (Hood) was collected at both locations in 2002 only, and *F. tritici* was collected at both locations in 2000 only. Species diversity was similar at both locations.

Three of the ten species of thrips captured on sticky traps in wheat were not found on sticky traps in cotton; these three were *Aeolothrips duvali* Moulton, *F. tritici*, and *Thrips winnemanae* Hood. *Pseudothrips inequalis* (Beach) was the only thrips captured on sticky traps in cotton that was not captured on sticky traps in wheat. *Chirothrips simplex* Hood, *Microcephalothrips abdominalis* (Crawford), *N. variabilis*, *P. ayarsi*, and *P. inequalis* were captured on sticky traps in cotton, but these five species were not recovered from cotton plant wash samples. *Frankliniella exigua* was recovered all three years from cotton plant wash samples, but this species was not captured on sticky traps in wheat or in cotton. *Frankliniella genuina* was recovered one year from plant wash samples, but this species was not captured on

TABLE 2. Percentage of thrips species captured in cotton on blue and yellow sticky traps, 2000-2001.

Species	Hardeman Co.		Knox Co.	
	Blue Trap	Yellow Trap	Blue Trap	Yellow Trap
Chirothrips simplex Hood	0.25	0.26	0.00	0.20
Frankliniella fusca (Hinds)	0.00	1.05	0.19	4.71
F. occidentalis (Pergande)	99.24	96.86	88.89	93.44
Microcephalothrips abdominalis (Crawford)	0.00	0.00	0.19	0.00
Neohydatothrips variabilis (Beach)	0.00	0.26	0.00	0.00
Plesiothrips ayarsi Stannard	0.00	0.26	0.00	0.20
Pseudothrips inequalis	0.00	0.26	0.00	0.00
Thrips tabaci Lindeman	0.51	1.05	10.73	1.43
N	393	382	522	488

TABLE 3. Percentage of thrips species recovered from cotton plant wash samples, 2000-2002.

Species	Hardeman Co.	Knox Co.	Average
Frankliniella exigua (Hood)	10.32	12.10	11.21
F. fusca (Hinds)	18.23	25.51	21.87
F. genuina Hood	0.27	0.24	0.26
F. occidentalis (Pergande)	61.80	41.32	51.56
F. tritici (Fitch)	0.54	0.72	0.63
Thrips tabaci Lindeman	8.85	20.12	14.49
N	746	835	
Species diversity (H') [a]	0.4098 a	0.4250 a	

[a] Average H' with a common letter are not significantly different (P>0.05).

sticky traps in wheat or in cotton. These species capture data indicate that sticky traps in cotton and wheat do not adequately represent the species composition of thrips that actually infest cotton plants after planting. However, sticky traps in wheat and cotton and plant wash samples in cotton all indicate that *F. occidentalis* was the dominant species of thrips in the northern Texas Rolling Plains during this study.

There was no correlation between the last captures of thrips in wheat and the first captures in cotton on either color of sticky trap ($r = 0.165$, $P = 0.754$, n = 6 for yellow; and $r = 0.337$, $P = 0.514$, n = 6 for blue). Also, there was no correlation between number of thrips

captured on sticky traps and the number of thrips collected in plant wash samples ($r = 0.018$, $P = 0.956$, n = 12 for blue sticky traps and plant wash; $r = 0.401$, $P = 0.197$, n = 12 for yellow sticky traps and plant wash). These results indicate that sticky traps were not useful for predicting thrips numbers in cotton.

The blue and yellow trap colors were selected because several of the thrips species (*F. fusca*, *F. occidentalis*, *F. tritici*, and *T. tabaci*) that we suspected were present in the northern Rolling Plains are known to respond to these two colors (Terry 1997). Because *F. occidentalis* was so dominant in numbers, the catches on sticky traps of this species was investigated. Captures of *F. occidentalis* did not vary by county ($F = 0.84$; df = 1,2; $P = 0.456$) or by trap color ($F = 1.09$; df = 1,4; $P = 0.356$). However, thrips were much easier to see and count on yellow traps.

High numbers of thrips were caught on the sticky traps during some weeks (Fig. 1). Numbers counted on the traps inspected once a week represent about 10% of the actual numbers captured. On traps inspected weekly, trends indicate that thrips numbers were high in late April and then declined after mid May. Thrips could be moving from wheat to flowering weed hosts and then to cotton, and this could explain the lack of correlation between catches in wheat and cotton. Captures on traps inspected daily indicate that there were a few days with increased flight movement and many days with reduced flight activity.

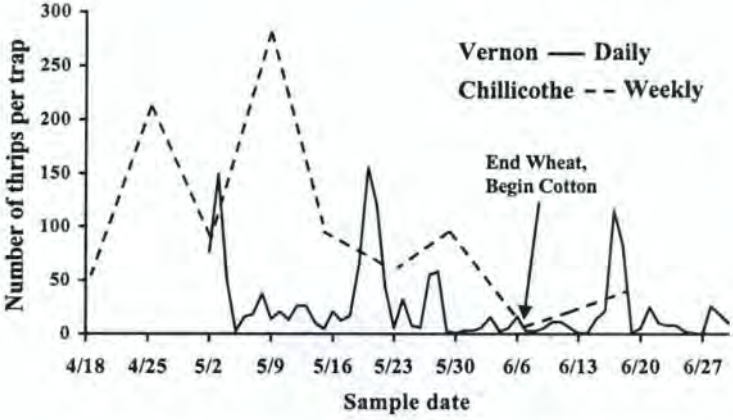

FIG. 1. Number of thrips captured daily or weekly on yellow sticky traps at Vernon and Chillicothe, Texas, 2000.

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SURVEY OF *LYGUS* SPP.[1] AND AN ASSOCIATED PARASITOID, *LEIOPHRON UNIFORMIS* (GAHAN)[2], IN COLORADO

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ABSTRACT

Surveys were conducted during 2001-2002 of nymphal *Lygus* spp. to determine incidence of the braconid parasitoid *Leiophron uniformis* (Gahan) in Colorado. The parasitoid was collected in all eastern Colorado sites, but not in sites in the San Luis Valley or west of the Continental Divide. Average combined parasitism for all sites was 5.54 and 7.62% in 2001 and 2002, respectively. Parasites were recovered from *Lygus* nymphs collected from May through October, consistent with a multivoltine life cycle. Highest percentage parasitism occurred with *Lygus* nymphs collected on alfalfa and canola. Parasitized nymphs were also collected from lambsquarters, flixweed and kochia, but not redroot pigweed. In 2002 the *Lygus* sp. found in highest percentage (58.4%) was *L. elisus* Van Duzee, which was particularly abundant during early season collections on flixweed and alfalfa/flixweed mixtures. *L. hesperus* Knight was second most abundant (29.3%) and was common on weed hosts lambsquarters and kochia. *L. lineolaris* (Palisot de Beauvois) comprised 12.3% of the 2002 collection and was most abundant on alfalfa and canola.

INTRODUCTION

The genus *Lygus* Hahn (Hemiptera: Miridae) includes important pests of cultivated and non-cultivated hosts in the United States (Schuh and Slater 1995, Wheeler 2000). Worldwide there are 43 known species of *Lygus* (Kelton 1975) with 34 present in the United States (Kelton 1975). Three *Lygus* species are important pests in cultivated crops and non-cultivated habitats in Colorado: *L. lineolaris* (Palisot de Beauvois), the tarnished plant bug; *L. hesperus* Knight, the western tarnished plant bug; and *L. elisus* Van Duzee, the pale legume bug.

Known parasites of *Lygus* include Mymaridae from eggs, several species of Braconidae, mainly from nymphs, and Tachinidae from adults (Clancy et al. 1966). Of the Braconidae, *Leiophron uniformis* (Gahan) is an important internal parasitoid of the nymphs of *L. elisus*, *L. hesperus*, and *L. lineolaris* in much of the southwestern United States (Clancy et al. 1966, Loan 1974, Marsh 1979, Graham et al. 1986, Day 1987, Debolt 1989). *L. uniformis* adults overwinter inside a cocoon among soil, under tree bark (Loan 1974, Loan 1983, Loan et al. 1987) or in other protected sites, emerging in April to May (Loan 1983). *L. uniformis* females will attempt to oviposit in all nymphal instars but prefer earliest instars (Loan 1974, Loan 1980, Debolt 1981, Loan 1983).

There have been no previous surveys of *Lygus* parasitoids present in Colorado. In order to provide better information on the natural enemy complex present, a primary purpose of this study was to determine seasonal occurrence of *L. uniformis* on *Lygus* species present in Colorado and to observe influences of cultivated hosts and non-cultivated hosts on parasitism incidence. A secondary objective was to better document the relative occurrence of *Lygus* spp. in the state and to note host associations.

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[1] Hemiptera: Lygeaidae
[2] Hymenoptera: Braconidae

MATERIALS AND METHODS

Eleven Colorado sites were sampled during 2001 in Larimer (7 sites), Weld (2 sites) and Mesa (2 sites) counties. A total of 50 samples were taking from 14 June to 15 August. Field sampling involved using a standard 15-in diameter sweep-net. The number of sweeps varied between sites, with sufficient number made to acquire a sample of *Lygus* nymphs for rearing. *Lygus* nymphs were aspirated and transferred to a cardboard ice-cream container with some host plant material for transport back to the laboratory.

A variety of vegetation types were sampled including roadsides and fields, primarily containing flixweed [*Descurainia sophia* (L.) Webb ex Prantl, alfalfa (*Medicago sativa* L.), and canola (*Brassica napus* L.). Most samples were taken from alfalfa (41 samples involving 558 *Lygus* nymphs) followed by flixweed (7 samples totaling 179 nymphs). Based on concurrent adult collections, the great majority of the *Lygus* spp. present were *L. elisus*; a small percentage of the adult insects were *L. lineolaris*.

Nymphs were reared within cardboard ice-cream containers, modified with a mesh-covered opening cut into the top. The nymphs were provided green bean pods and a small piece of cotton wick slightly moistened with water. All samples were checked daily for evidence of parasitoid emergence from nymphs. Emerged parasitoids were promptly transferred to individual glass vials to prevent predation and allow for adult emergence.

Sampling initiated 29 May and continuing through 8 October was expanded during 2002. The area of survey was also expanded to include sites in Larimer (14), Weld (32), Morgan (23), Logan (20), Mesa (5), Delta (7), Montrose (17), Pueblo (2) and Alamosa (7) counties. In total, 257 site visits were made. Samples were taken from a mixture of vegetation types. Cropland involved in the study primarily involved monocultures of alfalfa or canola. Relatively pure stands of flixweed and alfalfa stands heavily infested with flixweed were sampled, particularly early in the season when flixweed was actively growing and flowering. Other sites sampled later in the season included alfalfa and various summer annual weeds including kochia (*Kochia scoparia* (L.) Schrad.), lambsquarters (*Chenopodium album* L.), and redroot pigweed (*Amaranthus retroflexus* L.).

After transfer back to the laboratory, *Lygus* nymphs were aspirated and transferred to Petri dishes for further rearing. A piece of filter paper was placed on the bottom and a moistened cotton wick was provided to increase humidity and provide a secondary water source; fresh green bean pods were provided for food. No more than twenty nymphs were placed in each rearing dish. Cultures were checked daily and parasitoids that had emerged were immediately transferred to individual vials for rearing. Parasitoids that died were preserved in alcohol. Notes were maintained on individuals that died for unknown causes, were parasitized or which developed to the adult stage.

RESULTS AND DISCUSSION

A total of 776 nymphs from 50 different samples were collected during 2001 sampling. *Leiophron uniformis* emerged from a total of 43 nymphs for an overall parasitism of 5.54% (Table 1). In 2002, 3,046 nymphs were collected from 257 samples. There was a large percentage of mortality from unidentified causes (e.g., cannibalism, injury during collection). Of those that survived, 144 parasitoids emerged and 1,745 *Lygus* developed to the adult stage for an overall parasitism percentage of 7.62%. *L. uniformis* was recovered from all surveyed counties in northeastern Colorado (Larimer, Weld, Morgan, Logan) and in Pueblo county. However, it was never collected from any surveys west of the Contentintal Divide in Mesa, Delta, and Montrose counties nor in Alamosa county located within the San Luis Valley.

Overall levels of parasitism are similar to those reported elsewhere. Averaged monthly parasitization of *L. elisus* and *L. hesperus* by *L.* (= *Euphoriana*) *uniformis* observed by Clancy and Pierce (1966) in California alfalfa remained below 2% in May, June and July and then increased to 7.2% in August. Graham et al. (1986) reported maximum parasitism of 10.6% in July at Tucson and 11.0% at Yuma.

In this study, parasites were recovered in June and July but not in the August samples during 2001 (Table 2). More extended sampling occurred in 2002, and parasitism was observed in all months from May through October, peaking at over 34% in August.

TABLE 1. *Lygus* Spp. Nymph Collections and Subsequent Parasitism, Arranged by Sampling Location, 2001-2002.

Colorado County	No. Sites	No. Samples	No. Nymphs	Unknown Mortality	No. Adults	No. Parasitized (Percent)[a]
			2002 Sampling			
Larimer	14	84	1267	531	741	68 (8.4)
Weld	32	70	964	378	587	47 (7.4)
Morgan	23	43	346	163	183	22 (10.7)
Logan	20	22	172	61	111	4 (3.5)
Mesa	5	5	40	15	21	0 (0.0)
Delta	7	7	40	35	5	0 (0.0)
Montrose	17	17	146	95	49	0 (0.0)
Pueblo	2	2	21	12	9	3 (25.0)
Alamosa	7	7	50	11	39	0 (0.0)
			2001 Sampling			
Larimer	7	33	579	-----	-----	42 (7.3)
Weld	2	7	85	-----	-----	1 (1.2)
Mesa	2	10	112	-----	-----	0 (0.0)
Total (2002)	127	257	3,046	1,301	1,745	144 (7.6)
Total (2001)	11	50	776	-----	-----	43 (5.5)

[a] Parasitism percentage in 2002 excluded nymphs that died from unknown causes. Parasitism percentage in 2001 based on all nymphs collected.

TABLE 2. *Lygus* Spp. Nymph Collections and Subsequent Parasitism, Arranged by Collection Date, 2001-2002.

Collection Month	No. Samples	No. Nymphs	Unknown Mortality	No. Adults	No. Parasitized (Percent)[a]
		2002 Sampling			
May	6	359	59	300	2 (0.7)
June	91	746	253	493	24 (4.6)
July	71	853	444	409	75 (15.5)
August	20	153	136	17	9 (34.6)
September	60	861	378	483	31 (6.0)
October	9	74	31	43	3 (6.5)
		2001 Sampling			
June	19	434	-----	-----	22 (5.1)
July	15	195	-----	-----	21 (10.8)
August	16	147	-----	-----	0 (0.0)
Total (2002)	257	3,046	1,301	1,745	144 (7.6)
Total (2001)	50	776	-----	-----	43 (5.5)

[a] Parasitism percentage in 2002 excluded nymphs that died from unknown causes. Parasitism percentage in 2001 based on all nymphs collected.

Loan (1983) reported emergence of Euphorine (= *Leiophron*) parasites in May, early June, late June, early July and August. Clancy and Pierce (1966) observed *L. uniformis* parasitism of 7.2% in August among a mixed population of *L. elisus* and *L. hesperus*. This increased to 11.0% in September and dropped to 5.7% in October. Parasitism of 4.0 and 9.5 percent was reported by Graham et al. (1986) in September at Tucson and Yuma, respectively. In Colorado surveys during 2002, parasitism continued to exceed 6% in September and October. Such an extended period of activity strongly supports a multivoltine life cycle in Colorado, similar to that reported elsewhere (Loan 1980, Loan 1983, Day 1987, and Ruberson et al. 2000).

Sampling of different *Lygus* hosts indicated that parasitism by *L. uniformis* occurred in alfalfa, flixweed, alfalfa/flixweed mixtures, kochia, lambsquarters, and canola (Table 3). The highest percentage parasitism occurred in canola during 2002 sampling (11.8%) and *L. uniformis* was recovered from one of five *Lygus* nymphs collected during 2001. Parasitism in alfalfa, the most intensively sampled crop, was 4.1 and 10.9% in the two years, respectively. This compares closely to the 11% parasitism of *L. lineolaris* in the alfalfa-grass fields reported by Day (1999). Graham et al. (1986) sampled *Lygus* in alfalfa, cotton, grain sorghum, guayule, and a number of weed hosts in southern Arizona and found parasitism by *L. uniformis* most consistently in nymphs collected from alfalfa. Jackson and Debolt (1990) speculated parasitism might increase by 60-70% if hay cutting was delayed for seed.

Substantial parasitism of *Lygus* by *L. uniformis* also occurred in many non-cultivated plantings. During 2002, parasitism in flixweed was 2.2%, kochia 6.6%, and lambsquarters 9.7% (Table 3). No parasites were recovered from *Lygus* collected in a relatively small number of samples from redroot pigweed. Clancy and Pierce (1966) noticed increasing *L. uniformis* parasitism of *Lygus elisus* on *Chenopodium* weed species from spring to midsummer. Graham et al. (1986) first recovered *L. uniformis*-parasitized nymphs from *Chenopodium* in April at Tucson, Yuma, and Gila Bend. The levels of parasitism on *Lygus* from *Chenopodium* plants were somewhat higher than on alfalfa: 28.6% in July in Yuma, 14.3% in April at Gila Bend, and 10% in August at Benson.

TABLE 3. *Lygus* Spp. Nymph Collections and Subsequent Parasitism, Arranged by Vegetation at Collection Site, 2001-2002.

Predominant Vegetation	No. Samples	No. Nymphs	Unknown Mortality	No. Adults	No. Parasitized (Percent)[a]
		2002 Sampling			
Alfalfa	141	1,466	766	700	86 (10.9)
Flixweed	40	522	126	396	9 (2.2)
Alfalfa/Flixweed	18	244	60	184	3 (1.6)
Kochia	22	263	106	157	11 (6.6)
Lambsquarters	19	278	139	139	15 (9.7)
Canola	13	243	93	150	20 (11.8)
Redroot pigweed	4	30	11	19	0 (0.0)
		2001 Sampling			
Alfalfa	41	558	-----	-----	23 (4.1)
Flixweed	7	179	-----	-----	19 (10.6)
Canola	1	5	-----	-----	1 (20.0)
Sunflower	1	34	-----	-----	0 (0.0)
Total (2002)	257	3,046	1,301	1,745	144 (7.6)
Total (2001)	50	776	-----	-----	43 (5.5)

[a] Parasitism percentage in 2002 excluded nymphs that died from unknown causes. Parasitism percentage in 2001 based on all nymphs collected.

A breakdown of the 1,745 *Lygus* species collected in 2002 and reared to the adult stage indicated three species present: *L. elisus*, *L. hesperus*, and *L. lineolaris* (Tables 4, 5). *Lygus elisus* was recovered in highest frequency at 58.4% of the total. *Lygus hesperus* was the second most abundant species in sampling (29.3%) followed by *L. lineolaris* (12.3%).

The highest population of *L. elisus* occurred during the early season and comprised 98.0 and 89.2% of all *Lygus* collected in May and June sampling, respectively. At this time, *L. elisus* was found mainly on flixweed and in alfalfa/flixweed mixtures, the plantings most commonly sampled during that period of the season. Fye (1982) reported *L. elisus* as the predominate species on weedy crucifers, chenopods, kochia, and pigweeds. Schwartz and Fottit (1992) reported *L. elisus* from 16 different families including Brassicaceae (*Brassica campestris*, *Brassica napus*, *Descurainia sophia*), Chenopodiaceae (*Amaranthus retroflexus*, *Chenopodium album*, *Kochia scoparia*), and Fabaceae, with alfalfa being the most important host. In this survey (Table 5) *L. elisus* was observed to commonly utilize both weedy hosts and the alfalfa and canola cultivated hosts.

Lygus hesperus tended to occur in higher percentage later in the season becoming the predominant *Lygus* species recovered during October and September. It was the species most commonly recovered from lambsquarters (87.8%), kochia (80.3%), canola (57.3%) and redroot pigweed (47.4%). It was also abundant in alfalfa and comprised 21% of the total *Lygus* recovered from that crop.

Lygus lineolaris was the species recovered in lowest numbers during 2002 sampling. Snodgrass et al. (1984) reported highest populations of *L. lineolaris* during May and June and again during September and October. In this study, greatest recoveries were populations occurring in July and October, with none collected in August (Table 5).

TABLE 4. *Lygus* Spp. Collected during 2002 Surveys, Arranged by Date of Collection.

Collection Month	No. Samples	No. Adults Reared	No. Identified to Species (%)		
			L. elisus	*L. hesperus*	*L. lineolaris*
May	6	300	294 (98.0)	4 (1.3)	2 (0.7)
June	91	493	440 (89.2)	36 (7.3)	17 (3.4)
July	71	409	197 (48.2)	98 (24.0)	114 (27.9)
August	20	17	11 (64.7)	6 (35.3)	0 (0.0)
September	60	483	77 (15.9)	337 (69.8)	69 (14.3)
October	9	43	0 (0.0)	31 (72.1)	12 (27.9)
Total	257	1,745	1,019 (58.4)	512 (29.3)	214 (12.3)

TABLE 5. *Lygus* Spp. Collected during 2002 Surveys, Arranged by Vegetation at Collection Site.

Dominant Vegetation	No. Samples	No. Adults Reared	No. Identified to Species (%)		
			L. elisus	*L. hesperus*	*L. lineolaris*
Alfalfa	141	700	397 (56.7)	147 (21.0)	156 (22.3)
Flixweed	40	396	383 (96.7)	9 (2.3)	4 (1.0)
Alfalfa/Flixweed	18	184	165 (89.7)	13 (7.1)	6 (3.3)
Kochia	22	157	22 (14.0)	126 (80.3)	9 (5.7)
Lambsquarters	19	139	7 (5.0)	122 (87.8)	10 (7.2)
Canola	13	150	37 (24.7)	86 (57.3)	27 (18.0)
Redroot pigweed	4	19	8 (42.1)	9 (47.4)	2 (10.5)
Total	257	1,745	1,019 (58.4)	512 (29.3)	214 (12.3)

Lygus lineolaris is an extremely polyphagous insect having over 300 recorded hosts (Schwartz et al. 1992). In this study, the highest percentage of *L. lineolaris* was found in collections of the two cultivated crops alfalfa (22.3%) and canola (18.0%) (Table 6). Leferink and Gerver (1997) reported *L. lineolaris* as the dominant species in canola, followed by *L. elisus*. In canola, (Timlick et al.1993) observed *Lygus* populations to reach a peak during the flowering of the host, decline, then peak again in the pod stages. *L. lineolaris* is well documented as a pest of alfalfa (Kelton 1975), the plant sampled most intensively late in the season during this survey.

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WATERMELON GROWTH AND YIELD REDUCTIONS CAUSED BY SQUASH BUG (HEMIPTERA: COREIDAE) FEEDING

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ABSTRACT

Adult squash bugs, *Anasa tristis* (De Geer) (Heteroptera: Coreidae), were confined at varying densities on watermelon plants (*Citrullus lanatus*, 'Mickeylee') at differing phenological stages in three field trials and were allowed to feed on the plants until plants died or fruit matured. Plant foliage and fruit were harvested and weighed to determine effects of squash bug feeding on growth and productivity. Dry foliage and fresh fruit weights were regressed on numbers of squash bugs, and results indicated that an increasing density of squash bugs feeding on plants resulted in significant reductions in plant growth and fruit yield. The earlier the stage of growth at which plants were inoculated with adult squash bugs the greater the reduction in plant growth and fruit production.

INTRODUCTION

The squash bug, *Anasa tristis* (De Geer), is a key pest of cucurbit crops grown throughout North America (Eichmann 1945, Edelson et al. 1999, Palumbo et al. 1993, Riley et al. 1998). The biology, host plant preferences, feeding behaviour and common predators and parasites have been previously described (Beard 1940, Bonjour and Fargo 1989, Bonjour et al. 1991, Neal 1993, Nechols 1987, Palumbo et al. 1991, Woodson and Fargo 1991). Although previous research indicated that watermelon, *Citrullus lanatus*, is not a preferred host (Bonjour et al. 1990), more recent research (Riley et al. 1998; Edelson et al. 1999, 2002, 2003) indicated that squash bugs feed, reproduce and negatively affect watermelon growth, productivity and survival.

Palumbo et al. (1993) and Fargo et al. (1988) found that squash bug feeding has a significant negative effect on squash growth and fruit production under field production conditions. Edelson et al. (2002, 2003) reported the negative effects of squash bug feeding on watermelon growth, productivity and survival under controlled conditions in the greenhouse and in containers under plastic hoop house covers. Research was initiated and is reported herein documenting the effect that squash bugs have on watermelon plants grown in the field using common commercial production cultural practices.

MATERIALS AND METHODS

Squash bugs used in the trials were taken from a culture as described by Edelson et al. (2002). Each year, approximately one week prior to initiating the experiments, a cohort of

fourth and fifth instars was removed from the cultures and isolated in a cage with squash fruit and plants. Adult bugs were used in all experiments and were taken from the cohort cage, examined individually to determine the sex and then placed in containers to be distributed to cages on plants in the field per treatment requirements.

All experiments were conducted in research plots at the Wes Watkins Agricultural Research and Extension Center (WWAREC) in southeastern Oklahoma, using commonly accepted commercial production practices. The fields were fertilized with 112 kg/ha nitrogen, phosphorous (P_2O_5) and potassium (K_2O) prior to planting. Drip irrigation lines were placed in the fields along each row and all plants were irrigated simultaneously as necessary. 'Mickylee' watermelon seeds were planted in June of each year. After seeds germinated, healthy individual seedlings were isolated by removing all other seedlings such that selected seedlings were separated from one another by 4m on each side (within planted rows and between rows). Plant row covers, approximately $0.5m^2$, were placed over the seedlings to prevent natural infestations by insect pests and to serve as cages to retain squash bugs placed on plants as treatments. Weeds were removed by hand from under the plant covers during the trials. Plants were monitored twice weekly; as vines lengthened, they were trained to grow under the plant cover and soil was placed around the cover to prevent squash bug movement out of the cages and to prevent other insects from moving into the cages.

A single field-based experiment was conducted each year during 2001, 2002 and 2003. The experimental design was a randomized complete block with four levels of insect density as treatments and six replicate blocks. Adult bugs were placed in the cages over plants at the seedling stage in 2002 and 2003 (1-2 true leaves), the vining stage in each year (4-5 true leaves and initiation of secondary vine growth), the flowering stage in each year (all untreated plants with one flower) and the fruiting stage in 2001 (all untreated plants with at least one fruit).

In 2001, we placed 0, 4, 8 or 16 adult bugs on plants at the vining, flowering and fruiting stages of growth. In 2002 and 2003 we placed 0, 1, 2, or 4 adult bugs on seedling stage plants and 0, 4, 8, or 16 adult bugs on vining and flowering stage plants. Therefore, we inoculated only two experiments (2002, 2003) at the seedling stage, three experiments at the vining and flowering stages (2001, 2002, 2003) and one experiment at the fruiting stage of growth (2001). For each treatment, we used equal numbers of male and female adult bugs with the exception of the treatment with '1' adult in which we used only females.

During each year we monitored plant growth at two-day intervals, noting number of leaves, production of vines, flowers and fruit. For inoculations, adult bugs were removed from the laboratory cultures containing same age cohorts, and we examined each to determine sex and placed them in plastic containers. The containers with a set number of bugs were placed in cages covering the plants and were opened to release the adult bugs. Adult bugs were counted coincident with monitoring of cages. Additional bugs from the same cohort maintained in the lab were added to cages as required based on counts of bugs present in the cages. Excess adults and/or nymphs were removed as needed to maintain set numbers for each treatment.

Plants and fruit were harvested when plants died and those plants that did not die prior to maturation of fruit were harvested on the same date. The date on which plants that grew to maturation were harvested was determined based on maturation of at least one fruit in all of the untreated (0 squash bugs) plots. All fruit on vines was removed, counted and weighed. Plant foliage was harvested by cutting the vines at the crown soil level and placing the foliage and stems in bags. The foliage was dried in open paper sacks in a greenhouse at approximately 35°C for 14 days and weighed. Results were recorded in grams dry weight for foliage and as kg fresh weight for fruit.

Data from all experiments were analysed using analysis of variance to determine whether there were interactions among treatments and years (2001, 2002, 2003) (SAS 1987). Treatments (number of adult bugs) were then regressed on dependent variables (weight of foliage and fruit) to determine and describe relationships (SAS 1987). Linear regression analyses were run using the square root transformed number of squash bug adults.

RESULTS AND DISCUSSION

Results of analysis of variance indicated that there were no significant interactions among treatments for each phenological stage of plant growth and the different years (with the exception of the fruiting stage) in which the experiments were conducted based on foliage weight with the exception of the vining stage (seedling; n= 40, F= 0.81, df= 2, P= 0.49; vining; n= 60, F= 3.61, df= 2, P= 0.02; flowering; n= 60, F= 1.05, df= 2, P= 0.38). Similarly, results of analysis of variance indicated that there were no significant interactions among treatments for each phenological stage of plant growth and the different years in which the experiments were conducted based on fruit weight (seedling; n= 40, F= 1.39, df= 2, P= 0.26; vining; n= 60, F= 0.51, df= 2, P= 0.68; flowering; n= 60, F= 0.11, df= 2, P= 0.95). Therefore, data were pooled across years for seedling, vining and flowering stages and the pooled data were used for regression analyses.

Dry weight of foliage and fresh weight of fruit for plants that were inoculated at the seedling, vining, flowering and fruiting stages varied and in general declined as the number of squash bug adults increased (Table 1). Results of regression analysis indicated that there were significant (P = 0.05) negative linear effects due to number of squash bugs feeding on plants at the seedling and vining stages but not at the flowering and fruiting stages (Table 2).

TABLE 1. Mean (± SE) Dry Weight of Foliage (Grams) and Fresh Weight of Fruit (KG) for Each Treatment by Plant Stage of Growth with Data Pooled across Experiments.

		Weight	
Plant growth stage	No. Bugs	Foliage (Grams)	Fruit (KG)
Seedling stage	0	384.8 (58.0)	2.2 (0.4)
	1	225.0 (41.1)	1.9 (0.5)
	2	216.7 (61.0)	1.4 (0.4)
	4	128.6 (47.2)	1.2 (0.5)
Vining stage	0	391.9(39.1)	4.2 (0.8)
	2	206.2 (32.6)	1.9 (0.5)
	8	183.8 (34.5)	1.7 (0.5)
	16	132.9 (29.8)	0.8 (0.5)
Flowering stage	0	410.6 (44.0)	3.7 (0.8)
	2	319.1 (38.1)	2.6 (0.5)
	8	312.8 (49.0)	2.9 (0.7)
	16	317.9 (32.3)	2.8 (0.5)
Fruiting stage	0	459.0 (45.8)	7.3 (0.9)
	2	419.0 (63.3)	6.7 (0.9)
	8	433.0 (59.2)	4.2 (1.9)
	16	423.3 (87.7)	3.8 (1.0)

Calculated values for the slope describing the relationship between number of squash bug adults and foliage weight declined from -124.9 to 8.5 as the plants increased in

size and maturation from seedling to fruiting stages (Table 2), indicating that the effect of squash bug feeding was greater when bugs first began feeding on plants at the seedling stage compared to when feeding was initiated at later stages of plant growth. Results of regression analysis indicated that there were no significant differences in slope values between the seedling stage and vining stage based on an overlap in calculated CI values for each slope.

TABLE 2. Results of Regression Analysis with Data Pooled across Experiments using Square Root Transformed Number of Squash Bugs as the Independent Variable and Foliage Weight (Grams) and Fruit Weight (Kg) as the Dependent Variables by Plant Growth Stage.

Stage x Dependent Variable	Slope (CI)[a]	Intercept (CI)[a]	R^2	P^b
Seedling x foliage	-124.9 (-170 to -80)	376.4 (317 to 435)	0.97	0.02
Vining x foliage	-65.3 (-98 to -32)	371.9 (285 to 460)	0.94	0.03
Flowering x foliage	-24.5 (-53 to 3.9)	394.2 (319 to 469)	0.76	0.13ns
Fruiting x foliage	-8.5 (-22 to 4.8)	452.3 (417 to 487)	0.63	0.20ns
Seedling x fruit	-0.5 (-0.8 to -0.2)	2.3 (1.8 to 2.7)	0.93	0.04
Vining x fruit	-0.8 (-1.2 to -0.5)	4.0 (3.1 to 4.9)	0.96	0.02
Flowering x fruit	-0.2 (-0.6 to 0.2)	3.5 (2.5 to 4.5)	0.60	0.40ns
Fruiting x fruit	-0.9 (-1.9 to -0.04)	7.6 (5.2 to 10.0)	0.82	0.09ns

[a] CI – confidence interval (90%).
[b] ns – not significant ($P = 0.05$).

Calculated values for the slope describing the relationship between number of squash bug adults and fruit weight did not vary significantly (-0.2 to -0.9) among stages of plant growth when plants were inoculated (Table 2). This indicates that there was a significant negative relationship between number of bugs and fruit weight and that the decline as described by the calculated values for the slope did not vary significantly among plant growth stages based on overlapping CI values associated with each value for the slope.

Results of this research indicated that adult squash bugs had a significant negative effect on plant growth and productivity, and that the stage of plant growth at which the bugs initiated feeding significantly affected the rate of plant response in terms of plant foliage. Fruit yield rate response is negative but the slope of the response does not vary with the stage of plant growth when inoculation occurred. Furthermore, results indicate that significant negative responses occurred only when plants at the seedling or vining stages were fed upon by squash bugs and that there is no significant negative response when plants were first infested with bugs at the flowering or fruiting stages. Results from prior research conducted by Edelson et al. (2003) indicated that response rates did differ among plant growth stages, however, the previous work was conducted under more protected conditions in hoop houses with plants grown in containers.

Clearly, these and previous results provide evidence that adult squash bug feeding has a significant effect on productivity when plants are infested at the seedling and vining growth stages but may not have an effect on plants once they reach the flowering and fruiting stages of growth. Thus, squash bug management strategies in watermelon should be focused on plants in the early stages of growth. Based on the linear relations determined with regression analysis, we estimate that fruit yield may be reduced up to 22% by the feeding of one adult squash bug when feeding is initiated at the seedling stage and up to 20% at the vining stage. Relative to importance in pest management, these reductions in yield would result in significant monetary losses and would indicate that the

threshold for adult bugs feeding on plants is less than one per plant, dependent upon the value of the fruit which varies from $0.02 to $0.14 per pound. The value varies greatly based on time of year and cultivar, with fruit values greatest prior to the 4 July holiday and greatest for seedless cultivars.

Results from this study validate previous results (Edelson et al. 2002, 2003) for watermelon as affected by adult squash bug feeding which were conducted under controlled greenhouse conditions. Threshold models will be developed using these results to estimate yield losses based on adult squash bugs and varying market values of fruit. This information combined with costs of control based on applications of insecticides will be incorporated into recommendations for watermelon producers in the region.

These studies were conducted under common commercial field production conditions; however, we used a small, 'icebox' fruited cultivar, 'Mickeylee'. Results may differ or vary with other common cultivars of watermelon especially given the differences in sizes of fruit and genetics among various open-pollinated and hybrid diploid and hybrid triploid seedless watermelons. Future research will focus on determining whether feeding effects by squash bugs vary among different cultivars.

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PASSIVE TRANSMISSION OF SORGHUM ERGOT (*CLAVICEPS AFRICANA*) BY FOUR SPECIES OF ADULT STINK BUGS

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ABSTRACT

The ability of *Oebalus pugnax* (F.) (rice stink bug), *Acrosternum hilare* (Say) (green stink bug), *Nezara viridula* (L.) (southern green stink bug), and *Euschistus servus* (Say) (brown stink bug) to passively carry and transmit *Claviceps africana* spores from diseased to non-infected plants was evaluated at the USDA-ARS, Southern Plains Agricultural Research Center (SPARC), College Station, Texas. Following exposure to ergot-infected sorghum panicles in the field for 30 minutes, stink bugs were captured and subsequently released in cages containing panicles of healthy green-house–grown sorghum plants of male–sterile line ATx623 in full bloom. The highest level of disease severity was 14.1% when greenhouse-grown ATx623 plants were exposed to ergot contaminated rice stink bugs for 30 minutes; whereas, the lowest ergot infection (2.8%) was exhibited by panicles exposed to ergot contaminated brown stink bugs. Estimates of the mean number of *C. africana* spores adhering to the external body parts of ergot contaminated stink bugs after being used as vectors on healthy sorghum panicles also was recorded. The highest mean concentration of adhering *C. africana* spores (2.2×10^5 spores/ml) was recovered from ergot–contaminated rice stink bugs, while the lowest mean concentration of 6.2×10^4 spores/ml was from contaminated green stink bugs. No significant differences in mean count of external spore concentration recovered between southern stink bugs and rice stink bugs were noted; however, these mean spore counts were significantly higher than the amounts found on green and brown stink bugs. No ergot infection was evident on ATx623, and no spore was recovered from non-contaminated stink bugs.

INTRODUCTION

Claviceps africana Frederickson, Mantle, & de Milliano, the causal agent of sorghum ergot, poses a serious threat to sorghum (*Sorghum bicolor* (L.) Moench) profitability, especially in hybrid seed production when conditions are conducive for disease development. Yield losses as high as 80% have been reported in heavily infected hybrid seed production fields in India (Sangitrao et al. 1997). In the U.S., sorghum ergot was first observed in southern Texas in 1997 (Isakeit et al. 1998).

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C. africana infects only the unfertilized ovaries, and, within a few days after infection, produces honeydew, a viscous fluid matrix containing high concentrations of sugars mixed with massive amounts of conidia (Futrell and Webster 1965, Mower and Hancock 1975, Hassan et al. 1997, Bandyopadhyay et al. 1998). Wind currents are the primary means of spreading the disease (Bandyopadhyay et al. 1998). However, the sugary matrix of the honeydew makes it attractive to insects such as flies, beetles, wasps, moths, and head-feeding bugs which in turn may act as passive vectors for many ergot pathogens (Mower and Hancock 1975, Bandyopadhyay et al. 1998, Moreno et al. 1971, Hardy 1988, Prom et al. 2003).

Prom et al. (2003) have demonstrated that adult corn earworm moths, *Helicoverpa zea* (Boddie), can act as passive vectors of *C. africana* conidia. Hardy (1988) reported the transmission of *C. paspali* F. L. Stevens & J. G. Hall spores by wasps from diseased to healthy grasses. Also, the cabbage looper moth, *Trichoplusia ni* (Hübner) moth has been shown to spread *C. purpurea* (Fr.:Fr.) spores from infected plants to non-infected male-sterile barley plants (Moreno et al. 1971).

In Texas, several head-feeding bugs have been shown to frequent sorghum panicles from flowering, a period when sorghum plants are most susceptible to ergot infection, until hard-dough stages of development (Hall and Teetes 1981, Bandyopadhyay et al. 1998, McPherson and McPherson 2000). Hall and Teetes (1981) recovered large numbers of adult rice stink bugs, *Oebalus pugnax* (F.) from sorghum panicles at anthesis to milk stages of grain development, and adult southern stink bugs, *Nezara viridula* (L.) were found to be most prevalent on sorghum from soft to hard-dough stages of development. Few brown stink bugs, *Euschistus servus* (Say) were recovered from sorghum panicles (Hall and Teetes 1981). Hall and Teetes (1981) also noted that these sorghum pests were frequent visitors to many other plant hosts, including Johnson grass (*Sorghum halepense* (L.) Pers), dallisgrass (*Paspalum dilatatum* Poir.), oats (*Avena sativa* L.), barley (*Hordeum vulgare* L.), cotton (*Gossypium hirsutum* L.), and wheat (*Triticum aestivum* L.).

In order to minimize the impact of sorghum ergot and to establish effective control strategies, it is important to determine the potential of mobile insect pests of sorghum to act as passive vectors for the pathogen. The objectives of this study were 1) to assess the ability of four species of adult head–feeding bugs (rice, southern, green, and brown stink bugs) to passively transmit *C. africana* spores from diseased sorghum plants to non-infected plants, and 2) to quantify the amounts of *C. africana* spores borne externally by the four species of stink bugs after being exposed to ergot infected panicles and allowed to feed on healthy plants.

MATERIALS AND METHODS

A male-sterile sorghum line ATx623 planted in a field near the USDA-ARS, Southern Plains Agricultural Research Center, College Station, Texas, was the source of ergot inoculum. At 100% flowering, panicles were inoculated with *C. africana* spore suspensions using hand-held bottle sprayers. At 10–14 days after inoculation, adult insects from the four species of stink bugs were exposed in a nylon cage to infected panicles that oozed profusely with ergot honeydew. Ten to fifteen insects from each species of adult stink bug were released in separate nylon cages (30 x 30 x 30cm) containing a single ergot-infected panicle. After being allowed to feed on the ergot-infected panicles for 30 minutes, insects were captured and transferred to the greenhouse, along with control insects not exposed to ergot-infected sorghum plants.

All the adult stink bugs, used in this experiment were captured in black light traps (Latheef et al. 1991) placed in various locations, 80–100m apart near fields in the Brazos

River Valley, southwest of College Station. Insects were collected daily through June and July and transported to the laboratory (USDA-ARS, SPARC, College Station, Texas). Each species was held separately in 30 x 30 x 30cm Plexiglas® cages until ready for use. Insects were starved for two days prior to being released on the ergot-infected panicles in the field.

In the greenhouse, five ergot-contaminated stink bugs per cage from each species were released for 30 minutes into clean nylon cages (30 x 30 x 30cm) containing healthy ATx623 sorghum panicles in full bloom. Each treatment was comprised of three cages per insect species with single replicate panicles per cage. Control treatments (non-contaminated insects) were set up similarly. All the panicles in each treatment were enclosed in brown paper sacks for four days to enhance infection. The experiments were repeated three times. The greenhouse-grown sorghum plants were raised from ATx623 seeds sown directly into 20cm diameter plastic pots containing Metro-Mix growing medium (Scotts-Sierra Horticultural Products Company, Maryland, OH). Seedlings were thinned to a single plant per pot, and grown at 25°C ± 2.

Fourteen days after exposure to ergot-infected or non-infected adult stink bugs, disease severity was assessed for each treated panicle. Disease severity for each treated panicle was based on the number of infected florets divided by the total number of florets per panicle multiplied by 100.

The amount of ergot spores borne externally by each species was estimated following their exposure to healthy greenhouse grown plants. Thirty minutes after the bugs were released into cages containing healthy sorghum panicles, these insects were individually transferred to vials containing 3ml reverse osmosis water. Spores adhering to the external body parts were dislodged by vortexing individual vials containing the insects for 30 seconds. The stink bugs were removed from the vials, and the number of *C. africana* spores in each vial was determined using a hemacytometer.

Data for disease severity and spore concentration were analyzed using the command Proc Univariate (Statistical Analysis 8.1, SAS Institute, Cary, NC). Mean comparisons were conducted with Tukey's studentized range test at the 5% probability level.

RESULTS AND DISCUSSION

Both the ability to transmit *C. africana* spores from diseased to non-infected male–sterile sorghum line ATx623 and estimates of external spore contamination were significantly affected by stink bug species (Table 1, 2). The highest percentage disease severity on the ATx623 sorghum line (14.1%) was caused by ergot-contaminated rice stink bugs (Table 1). This level of infection was significantly higher than values from green and brown stink bugs. The 11.1% ergot severity caused by southern stink bugs was not significantly different ($P = 0.05$) from that exhibited by the other stink bug species. No ergot infection was noted on ATx623 exposed to non-contaminated stink bugs.

TABLE 1. Mean Percent Disease Severity and Standard Error (SE) of Greenhouse-Grown Male–Sterile Sorghum Plants Exposed to Ergot Contaminated and Non-Contaminated Stink Bugs for 30 Minutes.

Insect vectors	Percentage disease severity and SE[a]		
	Mean[b]	Minimum	Maximum
Rice stink bug	14.1 ± 2.7a[c]	2	28
Southern stink bug	11.1 ± 3.9ab	1	40
Green stink bug	3.8 ± 1.1bc	1	10
Brown stink bug	2.8 ± 1.0bc	1	10
Control	0.0	0	0

[a]Percentage disease severity is based on the [(number of infected florets)/total florets] x 100.

[b]Mean percentage disease severity ± S.E., N = 9 per treatment.

[c]Mean separation by Tukey's Studentized Range Test; means within a column with the same letters are not significantly different at $P = 0.05$.

TABLE 2. Estimates of Adhering *Claviceps africana* Spores and Standard Error (SE) Recovered From Contaminated and Non-Contaminated Stink Bugs after Feeding for 30 Minutes on Non-Ergot Infected Sorghum Panicles.

Insect vectors	No. of insects	Concentration of spores and SE recovered from stink bugs[a]		
		Mean spore concentration (spores/ml)[b]	Minimum	Maximum
Rice stink bug	27	$2.2 \times 10^5 \pm 3.5 \times 10^4$ a[c]	2.5×10^4	6.4×10^5
Southern stink bug	27	$1.9 \times 10^5 \pm 3.5 \times 10^4$ a	1.5×10^3	6.1×10^5
Green stink bug	27	$6.2 \times 10^4 \pm 1.2 \times 10^4$ b	1.0×10^3	2.6×10^5
Brown stink bug	27	$8.3 \times 10^4 \pm 2.1 \times 10^4$ b	2.2×10^3	5.4×10^5
Control	45	0.0	0.0	0.0

[a]Mean spore concentration = estimates of recoverable adhering *C. africana* spores per insect.

[b]Mean spore count per stink bug ± S.E.

[c]Mean separation by Tukey's Studentized Range Test; means within a column with the same letters are not significantly different at $P = 0.05$.

This study demonstrated that rice stink bugs, southern stink bugs, green stink bugs, and brown stink bugs have the capacity to passively transmit *C. africana* spores from diseased to healthy sorghum plants. The rice stink bug was the most efficient vector of sorghum ergot spores, followed by the southern stink bug. This observation is significant because rice stink bugs have been observed to visit sorghum panicles frequently beginning at flowering to the milk stage of development (Hall and Teetes 1981), a period during which sorghum is most susceptible to ergot.

Hall and Teetes (1981) also observed that a number of plant species, including Johnson grass often found close to sorghum fields are potential sources of rice stink bugs and other head-feeding bugs. In addition, Johnson grass has been shown to invade sorghum fields, and is an alternate host to *C. africana* (Bandyopadhay et al. 1998, McPherson and McPherson 2000). In some areas in the U.S., Johnson grass flowers

continuously, and, if such fields are infected with ergot and visited by mobile pests, these contaminated insects have the capacity to passively transmit and spread the disease to flowering sorghum plants.

Other mobile insects such as, corn earworm moths, cabbage looper moths, and wasps have been shown to act as passive vectors for *C. africana* and other *Claviceps* species (Moreno et al. 1971, Hardy 1988, Lemon 1992, Prom et al. 2003).

This study has shown experimentally, for the first time, the ability of stink bugs to passively transmit ergot. As sorghum matures, it becomes less suitable as a host for the different species of stink bugs evaluated. Adults either disperse within the field with different stages of sorghum development or to other suitable hosts such as Johnson grass. Thus, the potential exists for stink bugs to be important vectors of sorghum ergot under suitable environmental conditions. Therefore, controlling these insects in sorghum fields will have the added benefit of minimizing the impact of the disease on sorghum crop.

Mention of trade names or commercial products in this article is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U. S. Department of Agriculture.

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MORPHOLOGICAL EXAMINATION OF THE EGG OF *MECIDEA MAJOR* (HETEROPTERA: PENTATOMIDAE)

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ABSTRACT

The egg of *Mecidea major* Sailer (Heteroptera: Pentatomidae) is described, including sculpturing of the chorion and number and size of the micropylar processes. Also presented is information on cluster size and arrangement of eggs within clusters and a description of the egg burster. The parasitoid *Psix tunetanus* (Mineo and Szabó) (Hymenoptera: Scelionidae) was recovered from field-collected eggs.

INTRODUCTION

The stink bug genus *Mecidea* (Pentatomidae: Pentatominae: Mecideini) occurs within the subtropical and adjacent temperate parts of the world and apparently is associated with xeric and semixeric environments (Sailer 1952). This phytophagous genus, which contains 17 species (Sailer 1952, Shuh and Slater 1995), is represented in America north of Mexico by only two species, *M. major* Sailer and *M. minor* Ruckes (Sailer 1952). *Mecidea major* and *M. minor*, collectively, range from the midwestern states to California, but only *M. minor* been reported from New Mexico (Froeschner 1988). Little is known about their biology, including their immature stages.

Mecidea major occurs most commonly from July to October (Sailer 1952) but has been collected in every month of the year (Jones 1993, Sailer 1952). It probably is a grass specialist although it has been collected from both grass and nongrass species (Sailer 1952). Host plants include side-oats grama, *Bouteloua curtipendula* (Michaux); sorghum, *Sorghum halapense* (L.); wheat, *Triticum aestivum* L.; "grasses;" spinach, *Spinacia oleracea* L.; cotton, *Gossypium hirsutum* L.; *Senecio* (Sailer 1952); wild oat, *Avena fatua* L.; Bermuda grass, *Cynodon dactylon* (L.); barnyard grass, *Echinochloa crusgalli* (L.); Lehmann lovegrass, *Eragrostis lehmanniana* Nees; bush muhly, *Muhlenbergia porteri* Beal; and *Bromus* sp. (Jones 1993).

Scattered notes have been published on the field life cycle of *M. major*. Walker (1993) found nymphs in Arizona primarily from early April to early June. He also reported that females he caged on potted *E. lehmanniana* (no date given) deposited five clusters of cream-colored eggs in two rows of 12-14 at the bases of the stems near the soil surface.

During May and June of 2003, several reproducing populations of *M. major* were found in the southern half of New Mexico on various species of range grasses but primarily on grama grasses, *Bouteloua* spp; Wright's threeawn, *Aristida purpurea* Nuttall;

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and tobosagrass, *Pleuraphis mutica* Buckley (CSB, unpublished data). The number of bugs and range of instar suggested that the populations were large enough for a life history study, including descriptions of immature stages. Presented herein is a description of the eggs.

MATERIALS AND METHODS

From April to November 2003, ≈250 adults were collected from range grasses in Doña Ana Co., including grama grasses, tobosagrass, and Wright's threeawn and used to establish a laboratory colony. From this colony, 30 males and 30 females were selected and placed in each of three ovipositional cages; dead individuals were replaced with additional field specimens as needed. Each cage consisted of a glass aquarium (61 x 32 x 41 cm) covered with a tight-fitting, metal-framed lid lined with fine cloth mesh to allow air circulation and prevent insects from escaping. Food consisted primarily of freshly cut side-oats grama and tobosagrass, depending upon availability. Stems with attached heads were placed in two, 1-pint Mason jars (≈ 0.47 liter) which were filled with distilled water and placed in the cage; plants were replaced weekly. A small plastic petri dish (≈ 10 cm diameter, 2 cm deep) filled with cotton and distilled water was placed in the cage to provide a water source for the bugs and footing if they entered the dish. Strips of cheesecloth, which served as oviposition sites, were suspended inside the cage and held in place by the lid.

The cages were examined daily for eggs. Plant material and cheesecloth, with attached egg clusters, were removed and placed on moist filter paper in the bottoms of the petri dishes; water was added as needed. Bugs and eggs were maintained in an incubator (Percival I-36 VL) at 25°C and a photoperiod of LD 14:10 h.

Clusters containing visible, mature embryos were preserved in 80% EtOH. Descriptions of the egg and egg burster is based on 217 and 49 specimens, respectively. Measurements (in mm) were made with an ocular micrometer and detailed observations of the morphology with scanning electron micrographs. Averages are expressed as means ± SE. Voucher specimens of adults and eggs were deposited in the New Mexico State Arthropod Museum in Las Cruces, NM.

RESULTS AND DISCUSSION

A total of 727 eggs were deposited in 74 egg clusters, ≈10 eggs per cluster (9.8 ± 0.44; range = 2-18). Clusters usually were deposited in regular alternating double rows on leaves (67.6%) (Fig. 1), flowers or maturing heads (5.4%), cheesecloth (24.3%), and glass walls (2.7%). As with other pentatomids, the eggs were glued to one another and the substrate. They were yellowish white when deposited, cream colored after 1-3 days, and light tan at maturity. Eyespots and mouthparts were visible in 4-5 days. Egg bursters appeared within 2-3 days of hatching.

Adults of the parasitoid *Psix tunetanus* (Mineo and Szabó) (Hymenoptera: Scelionidae) emerged in the laboratory from all eggs in a cluster of 13 that had been deposited in the field on foxtail barley, *Hordeum jubatum* L. Each operculum was shredded into thin strips which were left within the empty egg.

Egg Description: Length = 0.96 ± 0.03 (range = 0.85-1.12); width = 0.68 ± 0.003 (range = 0.58–0.83). Generally laid in clusters of 10 (range = 2–18), each cluster comprised of double alternating rows (Fig. 1); egg subcylindrical, yellowish white. Chorion strongly sculptured with irregular hexagonal reticulations (Fig. 2). Operculum present, convex and circular; diameter = 0.50 ± 0.003 (range = 0.43–0.61, n=120), surrounded by 7-18 micropylar processes. Micropylar processes clavate and smooth, each ≈0.02 mm long, opening at apex (Fig. 3). Egg burster T-shaped, heavily sclerotized,

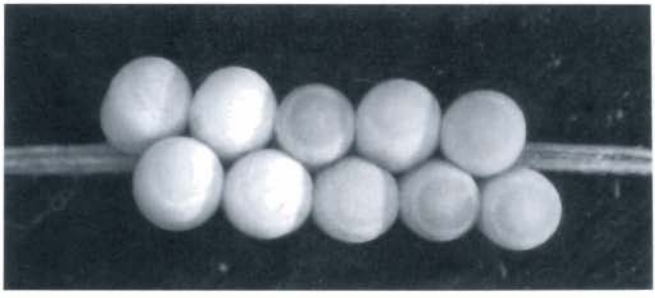

Fig. 1. Egg cluster of *Mecidea major* on leaf of side-oats grama.

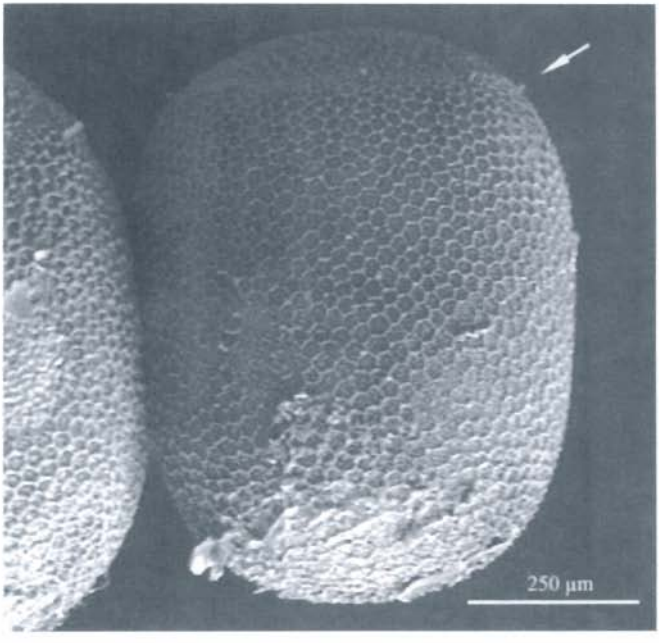

Fig. 2. Scanning electron micrograph of egg of *Mecidea major*. Note micropylar processes (arrow) along edge of operculum.

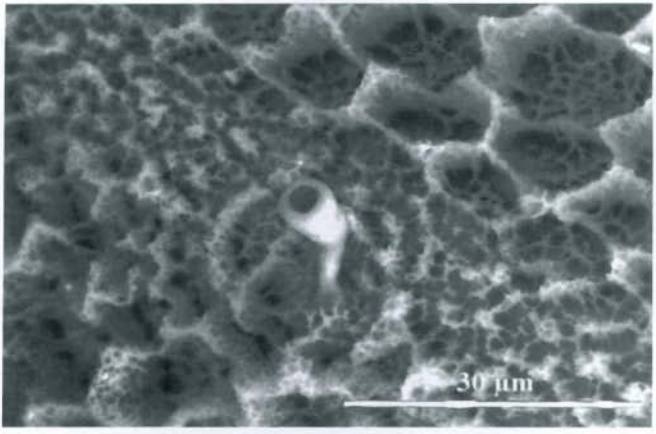

Fig. 3. Micropylar process of egg of *Mecidea major*.

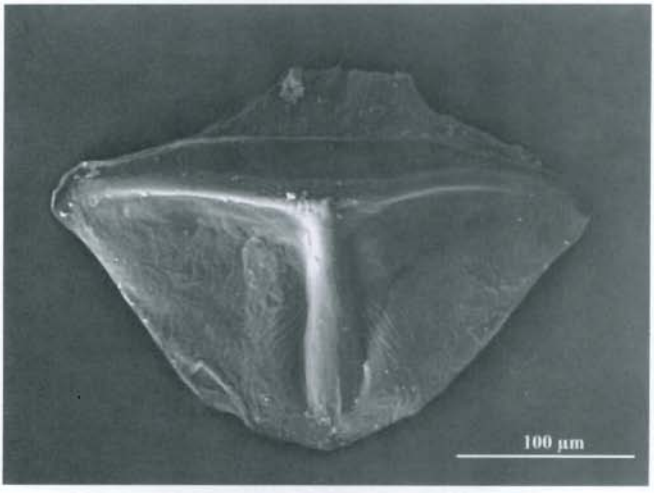

Fig. 4. Egg burster, dorsal view, of *Mecidea major*.

surrounded by membranous region, giving the entire structure a subtriangular appearance (Fig. 4).

As noted earlier, *M. major* usually deposited its egg masses in regular alternating double rows. Of the 74 clusters, 50 were laid on leaves, two on flowers, two on maturing heads, 18 on cheesecloth, and two on the glass walls. All clusters deposited on the plants and glass walls and 13 of the 18 deposited on the cheesecloth were laid in the regular double-row pattern. However, five of the clusters on cheesecloth were deposited in a loose double row (3) or in three or four irregular rows (2).

Esselbaugh (1946) discussed variation in the number of rows comprising an egg cluster within and between species of Pentatomidae. Within species, he noted that the egg masses of many were comprised of two or more rows and that the number of rows varied with size of the substrate in some species. Therefore, a species might deposit a cluster of two rows on a small stem or narrow fruit but deposit additional rows on a broad leaf blade or other broad surface.

Mecidea major appears almost inflexible in the number of rows of its egg clusters. In the present study, 72 of the 74 clusters were comprised of two rows, although they were deposited on both narrow (leaves) and broad (glass walls, cheesecloth) surfaces. As noted earlier, Jones (1993) also reported clusters of two rows. The rice stink bug, *Oebalus pugnax* (F.), another grass specialist, also characteristically deposits its eggs in regular alternating double rows and shows little variation, no matter the size of the substrate (Esselbaugh 1946; Bundy and McPherson, unpublished data). Whether or not this double-row pattern is consistent among grass-feeding species warrants further investigation.

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LIFE HISTORY OF THE PINE NEEDLE SCALE, *CHIONASPIS PINIFOLIAE* (FITCH)[1], IN NORTHEASTERN COLORADO

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ABSTRACT

Studies were conducted during 1993-1995 to determine aspects of the seasonal biology of the pine needle scale, *Chionapsis pinifoliae* (Fitch), in the Front Range of north central Colorado. The strain present is uniparental and univoltine. First emergence of crawlers occurred in mid-May in two years and late May in the third. Onset of crawler emergence was associated with several phenological indicators, including peak flowering of *Spiraea vanhouttei* Zab., *Syringa vulgaris* L., *Crataegus laevigata* Poir and several herbaceous perennials. Oviposition may begin in fall and overwintering stages include a mixture of eggs, females with visible ova but with no oviposition, and females that have not produced visible ova. Total egg production averaged between 8 and 27.6 per female, varying by site and date, with substantially lower fewer eggs produced in 1995 than in the previous year. Overwintering mortality of adult females, apparently most related to abiotic factors, averaged 44.8%.

INTRODUCTION

The pine needle scale, *Chionaspis pinifoliae* (Fitch), is an indigenous insect of North America that occurs as a serious pest of conifers in much of its range (Cumming 1953, Shour and Schuder 1987). A polyphagous species of conifers, host records include at least one species each from the genera *Pseudotsuga*, *Taxus* and *Torreya*; two species of *Abies*, *Cedrus*, and *Tsuga*; four species of *Picea*; and over twenty species of *Pinus* (Shour and Schuder 1987).

Life history of the pine needle scale can vary in several important respects among described populations in North America. Egg eclosion is reported to occur in June in many regions, with a single generation completed each year (Cumming 1953, Stimmann 1969, Luck and Dahlsten 1974); elsewhere two generations are reported with crawler eclosion in mid-May (Stimmann 1969, Shour 1986, Eliason and McCullough 1997). Both biparental (Cumming 1953, Shour 1986) and parthenogenetic (Brown 1965, Stimmann 1969) strains may occur. Luck and Dahlsten (1974) describe three major ecotypes of the pine needle scale, based on reported combinations of these life history characteristics.

In Colorado, pine needle scale is the most important armored scale infesting conifers. It is particularly damaging to various pines (e.g., mugho) and spruce. Previously published studies (Cooper and Cranshaw 1999) have reported on associated natural enemies in the region. However, *C. pinifoliae* biology in northeastern Colorado has been little studied. Improvement of information associated with life history can be essential to optimal management by allowing proper timing of sprays to coincide with crawler emergence. The purpose of this study was to better describe the life history of pine needle scale, on a range of hosts commonly planted along the northern Front Range of north central Colorado.

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[1] Homoptera: Diaspididae
[2] Currently at: 13555 N. Sandra Rd., Marana, AZ 85653.

MATERIAL AND METHODS

Field studies began April 1993 and ended June 1995. Reported study sites were all within Larimer County, Colorado, in the vicinity of the campus of Colorado State University. Within these areas, observations were made of pine needle scale on several different host plants including Austrian pine [*Pinus nigra* L.], pinyon [*Pinus edulis* (Engelman)], mugho pine/Swiss mountain pine [*Pinus mugo* Turra], ponderosa pine [*Pinus ponderosa* (Douglas)], and blue spruce [*Picea pungens* (Engelman)]. Study plantings were in widely diverse sites, including irrigated landscape/park settings, marginal areas adjacent to parking lots and roads, and natural stands. Plantings included between six and 20 individual trees, of roughly equal age. Specific attributes of the Ft. Collins study sites were as follows:

Site A: Protected area on the campus of Colorado state University in an irrigated landscape; host species present included lightly pine needle scale-infested *P. edulis*, heavily infested *P. mugo*, heavily infested *P. nigra*, and lightly infested *P. ponderosa*;
Site B: Plantings located in a commercial area surrounded by parking lots and streets; Lightly pine needle scale-infested *P. edulis* and moderately infested *P. nigra*; and
Site C: Plantings located in a non-irrigated setting of intermediate shelter compared to the above sites; light infestations of pine needle scale were present on *P. pungens* and *P. ponderosa*, moderate infestations on *P. nigra*, and heavy infestations on *P. mugo*.

Surveys were conducted at approximate one-week intervals during the periods of egg hatch, at monthly intervals during the growing season, and at two- to three-month intervals during the dormant season. All sample collections were made by removing infested needles and returning them to the laboratory for examination. During each sample, collections were made from all cardinal points of the plant and at differing heights within the canopy. Numbers of scales examined varied. Between 25 and 100 individual scales were examined at each sample site to determine scale mortality and the condition of the females before egg eclosion. Surveys following onset of eclosion typically involved examination of ten scales per site in 1993 and 25 in 1994 and during the limited 1995 sample period. Duration of eclosion and activity of first instar nymphs (crawlers) were also recorded during these observations.

Identification of blooming plants phenologically associated with first egg hatch were made during 1993 and 1994. These associations were identified by examining plant development in a botanical collection at the Plant Environmental Research Center (PERC) at Colorado State University, which was within 5km of all pine needle scale study sites.

RESULTS AND DISCUSSION

Throughout the course of this study, no male scales were ever observed. This has also been the personal observation of the second author over the course of over 20 years examining pine needle scale in northeastern Colorado. A parthenogenetic form of the pine needle scale appears to be established in north central Colorado, consistent with the uniparental ecotype that Luck and Dahlsten (1975) report as being limited to the western United States.

During 1993 studies, crawlers were first observed on 14 May. The crawlers were found beneath the protective test of overwintered females that had been collected 10 May from Site A and subsequently maintained in the laboratory at room temperature (ca 22^{0}C). The following year, at study sites A and C, crawlers were first noted in the field 17 May. Numbers of observed crawlers at Site A declined sharply (ca. 85%) between observations made 17 May and 3 June; conversely number of crawlers at Site C doubled between these two sample dates. Few crawlers were noted as being present 18 June at both locations. This extended crawler emergence may be related to the presence of gravid overwintered females (Shour 1986) which were commonly observed during 1994.

Beginning in early May 1995 an extended period of wet weather prevailed for six weeks. Crawlers were again initially observed mid-May (15 May) at Site C, but subsequent observations indicated an asynchronous emergence pattern. Crawlers were first present 31 May at Site B, while eclosion was just beginning 1 June at Site D. Newly settled crawlers were not observed at Site A until 24 June. Cool, cloudy weather may cause crawlers to

remain under the test of the mother scale for extended periods until conditions improve (Cumming 1953).

Full bloom of several flowering plants in 1993-1995 were consistently observed to be in full bloom at the approximate time of first crawler emergence (Table 1). Two of these, *Spiraea vanhouttei* Zab., bridal wreath spirea, and *Syringa vulgaris* L., common lilac, have previously been proposed as phenological indicators associated with pine needle scale crawler emergence (Gambrell 1938, Shour 1986). In addition, full bloom of three cultivars of English hawthorn and six herbaceous perennials coincided with egg hatch and first crawler activity of pine needle scale in the Ft. Collins area. These plants be used as phenological indicators of anticipated egg hatch to better time treatments directed at crawler stages.

TABLE 1. Herbaceous Perennial and Woody Plants with Peak Flowering Periods Phenologically Associated with First Crawler Emergence of Pine Needle Scale during 1993-1995, Ft. Collins, Colorado.

Scientific Name	Common Name
Woody Plants	
Spiraea vanhouttei Zab.	Bridal wreath spirea
Syringa vulgaris L.	Common lilac
Crataegus laevigata Poir cv. 'Paul's Scarlet', 'Crimson Cloud', 'Toba'	English hawthorn
Herbaceous Perennials	
Aquilegia x *hybrida* Sims	Hybrid columbine
Cheiranthus cheiri L.	Wallflower
Dianthus plumarius L.	Cottage pink
Iris hybrids	Bearded iris
Linum perenne L.	Blue flax
Ranunculus repens L.	Buttercup

This pattern of spring eclosion and crawler activity during mid-May is similar to that of biparental, bivoltine strains (Shour 1986, Stimmann 1969). However, subsequent observations throughout the growing seasons of 1993 and 1994 indicated only a single generation is produced in Colorado, more consistent with the univoltine/uniparental ecotype described by Luck and Dahlsten (1974) from western North America. With one exception, involving very small numbers of insects, crawlers were not collected, nor was there evidence of recent eclosion when examining females, following the normal spring eclosion period. However, small numbers of crawlers were observed, incidentally, during a mid-September control trial conducted at Site C in 1994 (Cooper and Cranshaw 1995). This is a full month past the last recorded crawler activity of 20 August reported by Shour (1986) with a bivoltine strain in Indiana. It is proposed that this is an anomalous event related to the extremely warm summer conditions that preceded.

Egg production was observed to occur over a very extended period. Ova were first visible within the bodies of females as early as mid-July in both 1993 and 1994. However, with a single exception (Site C on mugho pine), no oviposition was observed from late June through August.

During October sampling, a misture of forms was present: females that had begun to oviposit, females with ova but no oviposition, and females that had not visibly begun to produce ova. In observations of females collected 19 October 1993 at site A, 29% had ova visible within their bodies, but had not oviposited; 33% had already begun to lay eggs and had additional ova visibly present; and 6% had not yet produced visible ova. Thirty-two percent of the population was dead, but 14% of these had already laid eggs, averaging 9 eggs/individual. At site C during a 27 October survey 34% had both laid eggs and had additional ova visible, 14% had ova present only, and no live females were observed that had not begun egg production. Thirty-eight percent of the scales were dead, with an average of 2.3 eggs laid per individual.

TABLE 2. Percentage of Egg Production by Pine Needle Scale Produced during Fall, Comparing October and May Egg Production at Three Ft. Collins, Colorado Sites, 1993-1995.

Site	Host plant	No. scales observed	Collection date	Average no. eggs/female	Autumn-produced eggs (%)
A	*P. nigra*	100	13 October 1993	11.3	
A	*P. nigra*	100	17 May 1994	25.5	44
C	*P. ponderosa*	100	27 October 1993	8.0	
C	*P. ponderosa*	100	24 May 1994	15.0	53
A	*P. nigra*	25	30 October 1994	7.0	
A	*P. nigra*	50	15-31 May 1995	16.0	43
A	*P. mugo*	25	29 October 1994	0.0	
A	*P. mugo*	50	15-31 May 1995	18.0	0
A	*P. ponderosa*	25	30 October 1994	2.0	
A	*P. ponderosa*	50	15-31 May 1995	14.2	14
C	*P. ponderosa*	25	30 October 1994	0.0	
C	*P. ponderosa*	50	15-31 May 1994	19.4	0
C	*P. mugo*	25	28 October 1994	11.2	
C	*P. mugo*	50	15-31 May 1994	9.4	≤100

Scale survival was much lower (27%, range 4-48%) during surveys conducted 28 and 29 October, 1994, at sites A, B, and C. Fall oviposition was also greatly decreased during this season. Although 65% of the surviving females were gravid, only one had begun to oviposit (n=125). There were no visible ova or oviposition among the other living scales.

Comparing egg production during October to the subsequent May, the percentage of eggs produced prior to winter was also variable (Table 2). At the two sites surveyed in October 1993, 44-53% of the eggs had been laid. In the subsequent season, no egg production had begun at two of the six sites, while at a third site all eggs had been produced before winter. Observed fall egg production was substantially lower than the 80% reported from New York by Gambrell (1938).

Overwintering mortality was determined by comparing the percentage of surviving females in October with those the subsequent May, just prior to eclosion. Eight such comparisons were made (Table 3), with survival averaging 55.2% (range 33-100%).

Total egg production was monitored at three Ft. Collins sites on a total of four different host plants (Table 4). The number of eggs produced per female ranged from 8 to 27.6. This was consistent with that reported by Shour (1986) with the first generation of a biparental strain in Indiana and by Luck and Dahlsten (1974) in California. Observed egg production per female was lower than reported by Herrick (1929), Cumming (1953), Brown (1959) and Eliason and McCullough (1997).

In seven of eight sites, egg production was substantially lower in spring 1995 (10.7 eggs/female) compared to the previous season (19.6 eggs/female). This may have been related to the abnormally hot, dry conditions that prevailed during the 1994 growing season.

In conclusion, pine needle scale present in northeastern Colorado has a mixture of the life history attributes described from other regions. It is uniparental and, primarily, univoltine, similar to that found in California. Overwintering can occur as eggs, as gravid females that have not yet produced eggs, and as a combination of producing an extended period of egg production. However, overall egg production is within the range described elsewhere. Also, egg eclosion and onset of crawler activity is similar to that described in other areas and involves similar phenological indicators (e.g., full bloom of common lilac, bridal wreath spirea).

TABLE 3. Overwintering Survival of Pine Needle Scale as Determined by Comparing October and May Survival Percentages at Three Ft. Collins, Colorado Sites, 1993-1995. Samples Based on Examination of 100 Scales on Each Sample Date.

Site	Host plant	Observation date	Percent live females (% overwintering survival)	
A	*P. nigra*	19 October 1993	67	
A	*P. nigra*	17 May 1994	41	(61)
C	*P. ponderosa*	27 October 1993	52	
C	*P. ponderosa*	24 May 1994	19	(36.5)
A	*P. nigra*	30 October 1994	24	
A	*P. nigra*	15-31 May 1995	8	(33)
A	*P. mugo*	29 October 1994	16	
A	*P. mugo*	15-31 May 1995	18	(≤100)
A	*P. ponderosa*	30 October 1994	24	
A	*P. ponderosa*	15-31 May 1995	16	(67)
B	*P. nigra*	30 October 1994	36	
B	*P. nigra*	15-31 May 1995	20	(55)
C	*P. ponderosa*	30 October 1994	50	
C	*P. ponderosa*	15-31 May 1995	20	(40)
C	*P. mugo*	28 October 1994	16	
C	*P. mugo*	15-31 May 1995	8	(50)

TABLE 4. Average Egg Production by Pine Needle Scale at Three Ft. Collins, Colorado Sites, 1993-1995.

Site	Host plant	No. scales observed	Collection date	Average no. viable eggs/Female
A	*P. nigra*	50	10-20 May 1993	26.0
A	*P. nigra*	100	17 May 1994	23.5
A	*P. nigra*	50	15-31 May 1995	13.5
A	*P. mugo*	100	18 May 1994	29.0
A	*P. mugo*	50	15-31 May 1995	8.0
A	*P. ponderosa*	100	3 June 1994	26.2
A	*P. ponderosa*	50	15-31 May 1995	14.2
B	*P. edulis*	25	3 May 1994	25.3
B	*P. edulis*	50	15-31 May 1995	14.7
B	*P. nigra*	50	3 May 1994	27.6
B	*P. nigra*	50	15-31 May 1995	11.6
C	*P. mugo*	100	18 May 1994	24.0
C	*P. mugo*	50	15-31 May 1995	9.4
C	*P. ponderosa*	100	24 May 1994	14.6
C	*P. ponderosa*	50	15-31 May 1995	19.4

INSECTICIDAL EFFECTS OF ALCOHOLIC EXTRACTS OF WILD PLANTS ON *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae)

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ABSTRACT

Methanol and ethanol extracts of 14 plants from the Sonora, Mexico, area were evaluated for control of the larger grain borer, *Prostephanus truncatus* (Horn). Methanolic extracts resulted in significantly greater mortality. Extracts of bursage, *Ambrosia ambrosioides* (Cav.), gave 100% mortality; no larger grain borer adults emerged, and weight loss and damaged grains were low (0.1 and 0.3%, respectively). Extracts of climbing milkweed, *Sarcostemma cynanchoides* Decne, and *Salpianthus macrodonthus* also exhibited activity, causing more than 75% mortality and adult emergence lower than 30%.

INTRODUCTION

The larger grain borer, *Prostephanus truncatus* (Horn), is a coleopteran of the family Bostrichidae. Most of the members of this family are wood and bamboo borers; however, the larger grain borer is a primary pest of maize (*Zea mays* L.) and cassava (Ramírez-Martínez et al. 1994). It has recently been detected in Sonora (Wong-Corral et al. 2001).

Fumigation is one of the most effective methods for protection of stored products, although disadvantages such as toxic residues, development of insecticide resistant, and increased costs of production often result. For these reasons, in recent years, botanical pesticides have been tested and found to provide novel modes of action against insect pests (Long et al. 2002). The present study evaluated the insecticidal potential of alcoholic extracts from wild plants of Sonora, Mexico, for control of larger grain borer.

MATERIALS AND METHODS

Plant species selected from a previous study (Cortez-Rocha et al. 1993), depending on their abundance, were creosote bush (*Larrea tridentata* DC), coyotillo (*Karwinskia humboldtiana* Schldl), castor bean (*Ricinus communis* L.), Tasmanian blue gum (*Eucaliptus globulus* Labill), canyon ragweed, bursage (*Ambrosia ambrosioides*) (Cav.), wild tobacco (*Nicotiana glauca* Graham), desert thorn apple (*Datura discolor* Bernh), slimleaf bursage (*Ambrosia confertiflora*) (DC), sticky baccharis, seepwillow (*Baccharis glutinosa* Pers.), doubleclaw (*Proboscidea parviflora*) (Woot.) Woot. & Standl., buffalo bur nightshade (*Solanum rostratum* Dunal), Arizona nettle-spurge (*Jatropha cinerea*) (Ortega), *Salpianthus*

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macrodonthus Standley, and climbing milkweed (*Sarcostemma cynanchoides* Decne). Six kilograms of leaves and stems of each plant species were collected near the Río Sonora, Río Yaqui, and the coast of Hermosillo, in Sonora, Mexico. Collected plants were sun-dried for two days and placed in the shade for three weeks until dry. Dry plant material was pulverized into particles less than 100-mesh in a Willey laboratory mill (Cortez-Rocha et al. 1993, Munro 1996), and sealed in plastic bags and stored in a dry place at room temperature.

Larger grain borer was obtained from a culture at the University of Sonora. Insects were reared at 27±2°C, 65% RH, and a 12:12 (L:D) photoperiod in glass jars with 500g of fresh corn. Newly emerged 2 to 3-day-old adults were separated by screening from the stock cultures.

Methanol (70%) and ethanol (70%) plant extracts were prepared by mixing 10g of dry plant material with 90ml of alcohol, shaking overnight, and allowing the mixture to stand for 2 days. The mixture was filtered through a cheesecloth, centrifuged at 7,000 rpm for 10 min, and filtered through Whatman No. 4 filter paper.

Filtered extracts were sprayed over the corn kernels (1ml/5g of corn). Ethanol and methanol also were sprayed on grain as controls. The kernels were agitated for 30 min and left to dry overnight at room temperature. Individual samples of 50g were placed in jars for a no election-test bioassay with three replications. Ten males and 10 females of 7-day-old larger grain borer were placed in each jar and incubated for 2 weeks at 27±2°C and 65% RH (Shires and McCarthy 1976). Live and dead insects were removed and counted after 2 weeks. The mortality was corrected using Abbot's formula (1925). Each jar was placed in a rearing chamber for 6-7 weeks until the emergence of F_1 progeny, after which the kernels were sieved three times a week for a total of 2 weeks to determine the total number of progeny.

Weight loss of the kernels was determined using the method described by Adams and Shulten (1978), which consisted of comparing the initial weight of sound grain and the final weight of infested kernels. Also, damaged and non-damaged kernels were counted and weighed to determine the percentage of damaged kernels.

A completely randomized experimental design was used with three replications. Data were analyzed using the SAS general linear model procedures (SAS 1992). Means were compared by the Tukey test to determine differences among treatments.

RESULTS AND DISCUSSION

Results of this study showed that both variables, kinds of alcohols and plant species, caused significant differences ($P \leq 0.05$) in the parameters evaluated. Methanolic extract from *A. ambrosioides* caused 100% mortality, while extracts from *S. cynanchoides, A. confertiflora,* and *S. macrodonthus* resulted in 93.0, 89.3, and 80.8% mortality, respectively, compared to the control (Table 1). Methanolic extracts from *R. communis, L. tridentata, K. humboldtiana,* and *D. discolor* caused the least mortality (6.0, 3.7, 2.0, and 2.3%, respectively).

Ethanolic extracts produced less mortality than methanolic extracts. Ethanolic extracts of *A. confertiflora* and *S. rostratum* resulted in the highest mortality (46.0, and 44.0%, respectively) compared to the control (Table 1). Lagunes and Rodríguez (1991) evaluated 130 plant species from Mexico against maize weevil, *Sitophilus zeamais* Motschulsky; larger grain borer, *Prostephanus truncatus;* bean weevil, *Acanthoscelides obtectus* (Say); and Mexican bean weevil, *Zabrotes subfasciatus* (Boheman). They reported that only 36 plants caused greater than 20.0% mortality and less than 50% emergence. Because methanol extracts of *A. ambrosioides, S. cynanchoides,* and *A. confertiflora*

resulted in the highest insect mortality, future work on isolation and characterization of bioactive compounds are necessary.

TABLE 1. Percentage Mortality and Adult Emergence of the Larger Grain Borer on Maize Treated with Methanolic and Ethanolic Extracts of Plants.[a]

	Mortality (%)		Adult Emergence (%)	
	Methanol	Ethanol	Methanol	Ethanol
A. ambrosioides	100.0a	1.8 e	0.0 d	35.7 de
S. cynanchoides	93.3a	4.5 cde	0.0 d	46.0 de
A. confertiflora	88.3a	46.0a	35.6 b	21.4 ef
S. macrodonthus	80.8a	1.7 e	0.7 d	58.6 cd
P. parviflora	40.3 b	0.0 e	2.6 d	57.3 cd
N. glauca	32.0 bc	13.3 c	0.0 d	41.3 de
B. glutinosa	27.0 bcd	0.0 e	2.3 d	28.7 ef
S. rostratum	17.0 cde	44.0a	1.0 d	10.0 f
E. globulus	11.0 de	12.3 cd	1.3 d	38.3 de
J. cinerea	10.0 de	0.7 e	0.0 d	60.0 cd
R. communis	6.0 e	3.7 cde	16.3 c	21.3 ef
L. tridentata	3.7 cde	3.7 cde	2.0 d	72.0 bc
K. humboldtiana	2.3 e	2.3 de	1.3 d	87.0ab
D. discolor	2.0 e	23.6 b	29.0 b	26.0 ef
Control	0.0 e	0.0 e	100.0a	100.0a

[a]Values based on the average of three replications. Values followed by the same letter in a column are not significantly different at 5% level (Tukey's multiple range test).

Both variables, kinds of alcohols and plants, also resulted in significant differences ($P \leq 0.05$) in the percentages of emergence of larger grain borer adults (Table 1). Fewer adults emerged from grain treated with methanolic extracts than from those of ethanolic extracts. No larger grain borer adults emerged from maize kernels treated with methanolic extracts of *A. ambrosioides, S. cynanchoides,* and *J. cinerea*. Most larger grain borer adults emerged from maize kernels treated with methanolic extracts of *A. confertiflora, D. discolor,* and *R. communis* (35.6, 29.0, and 16.3%, respectively). Methanolic extracts of *A. confertiflora* resulted in the highest mortality; however, it allowed most larger grain borer adults to emerge from the grain.

Kind of alcohol, but not plant species, resulted in significant differences ($P \leq 0.05$) in percentage of weight loss of maize kernels (Table 2). Maize kernels treated with methanolic extracts of plants had less weight loss than did those treated with ethanolic extracts. However, weight loss of the maize kernels did not differ significantly among the different kinds of plants.

Data from damaged grain were statistically different between alcohols and also among plant species ($P \leq 0.05$). Maize treated with ethanolic extracts resulted in the greatest percentage of damaged grain (Table 2). Eighty-seven percent of larger grain borer adults emerged, and 18.1% of grain was damaged from maize treated with ethanolic extracts of *K. humboldtiana*. Methanolic extracts resulted in the lowest percentage of damaged grains. Methanolic extracts from *A. ambrosioides, S. cynanchoides,* and *S. macrodonthus* resulted in 0.3, 1.2, and 2.3% of damaged grains, respectively. Maize kernels treated with *D. discolor* and *A. confertiflora* extracted in either kind of alcohol resulted in similar percentages of damaged grain.

TABLE 2. Percentage of Damaged Grain and Weight Loss by Larger Grain Borer in Maize Treated with Methanolic and Ethanolic Extracts of Wild Plants.[a]

	Weight loss (%)		Damaged grain (%)	
	Methanol	Ethanol	Methanol	Ethanol
S. rostratum	3.5a	3.5a	7.3abc	8.8 bc
D. discolor	2.9a	4.8a	9.2ab	9.2 bc
A. confertiflora	1.9a	6.7a	8.1abc	7.5 c
P. parviflora	1.5a	9.0a	5.7abc	13.8abc
E. globulus	1.4a	6.2a	3.6abc	13.3abc
K. humboldtiana	1.4a	6.9a	8.1abc	18.1a
S. cynanchoides	1.4a	7.2a	1.2 bc	12.2abc
R. communis	1.3a	5.3a	4.4abc	9.1 bc
B. glutinosa	1.2a	5.0a	6.8abc	9.1 bc
N. glauca	0.7a	5.2a	4.0abc	8.3 bc
S. macrodonthus	0.5a	7.1a	2.3 bc	11.7abc
J. cinerea	0.4a	7.2a	5.1abc	13.6abc
A. ambrosioides	0.1a	7.4a	0.3 c	11.4abc
Control	2.4a	7.8a	11.2a	16.3ab

[a] Values based on the average of three replications. Values followed by the same letter in a column are not significantly different at 5% level (Tukey's multiple range test).

Methanolic extracts were better than ethanolic extracts of plants in controlling larger grain borer. The greatest control was achieved when a methanolic extract from *A. ambrosioides* was used. Maize kernels treated with this extract resulted in the least amount of damaged grain or weight loss, and no larger grain borer adults emerged. Methanolic extracts of *S. cynanchoides*, and *S. macrodonthus* offer an alternative control for *P. truncatus* because of their toxic potential observed in this study. *N. glauca, S. rostratum*, and *J. cinerea* are other species whose methanolic extracts resulted in little or no emergence of larger grain borer adults from treated kernels of maize.

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SOUTHERN CORN STALK BORER[1], *DIATRAEA CRAMBIDOIDES* (Grote), FEEDING DAMAGE ON EASTERN GAMAGRASS[2] IN OKLAHOMA.

D. L. Maas[3] and T. L. Springer[3]

Eastern gamagrass, *Tripsacum dactyloides* (L.) L., is a perennial warm-season bunchgrass native to the eastern, central, and southern United States and is propagated vegetatively or by seed (Dewald and Louthan 1979). During a vegetative increase of 'Verl' eastern gamagrass germplasm in the late winter of 2002, an infestation by insects was detected. We found that several shoots of the proaxis were hollowed out from feeding and occupied by overwintering larvae (Fig. 1.). Similar amounts of infestation and damage were found during random sampling of plants from other nurseries, grazed pastures and seed production of eastern gamagrass blocks in Woodward and Harper Counties, Oklahoma. Adult insects were collected after emergence from the proaxis (Fig. 1.) and were identified by M. Alma Solis, USDA Systematic Entomology Laboratory, Beltsville, Maryland, as the southern corn stalk borer, *Diatraea crambidoides* (Grote). Voucher specimens were retained by the USDA Systematic Entomology Laboratory, Beltsville, Maryland.

During the summer of 2003, larvae ranging in size from 5 to 20 mm and pupae were observed in both reproductive and vegetative shoots of eastern gamagrass. A survey within a 6-year-old plot of 'FGT-I' germplasm showed an average of 11 insects per crown with a 2:1 preference for reproductive versus vegetative shoots (Springer and Maas, unpublished data). Eastern gamagrass has been previously noted as a food source for the southern corn stalk borer (Howard 1891, Phillips et al. 1921), although this is the first documentation in Oklahoma of eastern gamagrass being a host to all life stages of the southern corn stalk borer. The observed feeding damage caused by southern corn stalk borer to the proaxis is similar to that of maize billbug, *Sphenophorus maidis* Chittenden, (Maas et al. 2003) in Oklahoma. Southern corn stalk borer was identified as a contributing agent to severe stand damage of 'Pete' eastern gamagrass in Beltsville, Maryland, where random crowns of a 1996 established plot died in 2001 (Krizek et al. 2003). Plant vigor and number of inflorescences were reduced but few plants died in established plantings of 'Pete', 'Verl', and 'FGT-I' suffering from severe infestation in Oklahoma. Removal of shoot reserves by feeding may reduce the organic reserves needed for reproductive and vegetative shoot growth the following season. Loss of reproductive tillers in stands of eastern gamagrass stands will have a significant economic impact on the seed and forage industries.

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[1]Lepidoptera: Pyralidae

[2]Contribution of the USDA-ARS Southern Plains Range Research Station, Woodward, Oklahoma. All programs and services of the USDA are offered on a nondiscriminatory basis without regard to race, color, national origin, religion, sex, age, marital status, or handicap.

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FIG. 1. Larvae of *D. crambidoides* in a vegetative shoot of eastern gamagrass. Inset. Adult *D. crambidoides*. Scale: bar equals 5 mm.

Springer et al. (2004) estimated forage yield losses of as much as 1,100 kg ha^{-1} or economic losses of $45.00 per hectare in severely infested areas. Severe infestation by southern corn stalk borer may also contribute to center 'die out'. Additional research is needed to determine the life of southern corn stalk borer in *Tripsacum* sp. and to develop effective control procedures for a perennial species.

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SPRING EMERGENCE AND DEVELOPMENT OF THE COTTON FLEAHOPPER (HETEROPTERA: MIRIDAE) IN HOST PLANTS OF THE TEXAS ROLLING PLAINS

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ABSTRACT

The role of four spring weeds as hosts for the cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter), and the spring emergence periods of fleahopper nymphs from cotton, *Gossypium hirsutum* (L.), stalks and silverleaf nightshade, *Solanum elaeagnifolium* (Cav.), stems were studied for five years. Vacuum samples from horsemint, *Monarda punctata* (L.), contained the largest number of fleahoppers in all but one year of the study. Sampling two species of primrose, *Oenothera laciniata* Hill and *O. grandis* (Britt.) Smyth, yielded more fleahoppers than two species of gaura, *Gaura sinuata* (Ser.) and *Gaura villosa*, (Torr.). Redstem stork's-bill, *Erodium cicutarium* (L.) L'Hér., samples contained the least number of fleahoppers. The interval between the last fleahoppers found in spring weed hosts and the first fleahoppers found in cotton was short enough to allow infestation of a new cotton crop in three of five years studied. Eggs survived the winter and nymphs emerged from cotton stalks and nightshade stems beginning in March and continued to emerge until the first week of July.

INTRODUCTION

By 1926 the cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter), was found in all of the principal southern cotton states, including Alabama, Georgia, South Carolina, North Carolina, Tennessee, southern Oklahoma, and a few Gulf Coast counties of Texas (Ewing 1926). The fleahopper causes shedding of squares and abnormal growth of the plant, which results in excessive growth of the main stem and suppression of the fruiting branches. The result can be a loss of much of the early crop and varying amounts of the middle crop. Feeding injury during the seedling stage and on terminals results in retarded growth, abortion of pinhead squares, development of an excessive number of branches, the shortening of internodes, and the formation of irregular leaves (Eddy 1927). Damage thresholds are 25-30 fleahoppers per 100 cotton terminals coupled with reduced square set during the first three weeks of squaring (Baugh et al. 2005).

Cotton fleahopper eggs that survive the winter hatch the following spring, then nymphs infest various host plants (Ewing 1926). The fleahoppers leave weed hosts as the nymphs mature, and adults move to young cotton plants (Schuster et al. 1969). Research has shown that some of the most important spring host plants for the cotton fleahopper are croton (*Croton spp.*), evening primroses (*Oenothera spp.*), horsemint (*Monarda spp.*) (Fletcher 1940), and silverleaf nightshade, *Solanum elaeagnifolium* (Cav.) (Hixon 1941). Croton plants were not abundant in the general area of this study. There appears to be an increase in silverleaf nightshade associated with conservation tillage systems in winter wheat. Fleahoppers emerging from shredded cotton stalks and stems of nightshade in conservation tillage systems may infest the following year's cotton crop.

Primroses are a source of fleahoppers early in the growing season (Almand 1976). Since primrose plants grow in the rosette form with leaves nearly parallel to the ground, the plants may afford some protection for the fleahoppers from heavy spring rains. Almand (1974) noted that as the primrose plants produce more and more flowers, fleahopper numbers increase. Horsemint has been shown to be one of the preferred hosts of the cotton fleahopper (Holtzer and Sterling 1980), but this plant is generally not abundant adjacent to cotton fields in the Texas Rolling Plains. It is mainly a late spring and early summer plant in this area. Redstem stork's-bill, *Erodium cicutarium* (L.) L'Hér., is a common spring weed distributed throughout the Rolling Plains of Texas (Niehaus et al. 1984). Silverleaf nightshade grows in and adjacent to cotton and wheat fields.

A survey of some of the common host plants for fleahoppers in the Rolling Plains of Texas was undertaken to determine (1) utilization of spring host plants, and (2) the emergence period of fleahoppers from cotton stalks and silverleaf nightshade.

MATERIALS AND METHODS

This study was conducted in Knox Co., northern Texas Rolling Plains, during 2000-2004. Plants sampled in this study were mixtures of two species of primroses, *Oenothera laciniata* (Hill) and *O. grandis* (Britt.) Smyth; two species of gaura, *Gaura sinuata* (Ser.) and *Gaura villosa*, (Torr.); stork's-bill, *Erodium cicutarium* (L.) L'Hér.; horsemint, *Monarda punctata* (L.); silverleaf nightshade, *Solanum elaeagnifolium* (Cav.); and cotton, *Gossypium hirsutum* (L.). Croton, a common host for fleahoppers, was not available near the sites of this study; however, primroses, gaura, stork's-bill, horsemint, silverleaf nightshade and cotton were present in isolated locations. Nuessly and Sterling (1984) found that a vacuum sampler (D-vac) was preferred for sampling arthropods over a modified drop cloth method of sampling. A Model 1612 backpack aspirator with a 2-cycle single cylinder gasoline engine (John W. Hock Co., Gainesville, Florida) was used in this study. Fleahoppers vacuumed from host plants were trapped in nylon organza collection bags, approximately 50 cm long by 33 cm wide at the mouth and 7.6 cm wide at the tip. Sampling sites were selected along county roads and borders of fields where plant density was sufficient to allow a 30-second sample within each plant species. However, no data were collected concerning plant density. Sampling of spring weeds was initiated in mid to late April. The vacuum samples were refrigerated overnight at 0-2° C and examined for fleahoppers the next day.

According to Hixon (1941) good growing sites for the hosts of the cotton fleahopper are disturbed soils such as abandoned fields, road sides and fence rows. Vacuum samples of primrose, gaura and horsemint were taken along Farm Road 1292 approximately three miles northwest of Knox City, Texas in Knox Co. and along the edges of fields near county roads two miles northwest of Knox City. Samples of stork's-bill were taken at the Smith Farm leased by the Texas Agricultural Experiment Station at Munday, Texas. Soil types at all of the collection sites were sandy and sandy-loam. In most years, when there were multiple areas of the weeds available, two 30-second samples from each spring host were taken each week beginning in early April.

In addition to the vacuum sampling, cotton and nightshade stems were collected in the late fall of each year in Knox County and put into closed PVC tubes to monitor emergence of fleahopper nymphs the following spring. Samples for emergence study were taken by cutting 15-20 cm lengths of the stems, with leaves removed, of each kind of plant and inserting them into tubes made from 30 cm long PVC pipe with a diameter of 10 cm. Only one species of plant was placed into each tube. A PVC cap was installed on one end of the pipe. A hole was drilled into the center of the cap to hold a 28 x 70 mm, 18.5-ml capacity vial to capture the emerged fleahopper nymphs. On the other end of the pipe, black cloth, to allow ventilation yet exclude excessive light, was held in place by hose

clamps to prevent the fleahoppers from escaping. The tubes were placed in an outdoor, screened insectary at Vernon, Wilbarger Co., Texas. Water was sprayed into the tubes following rain to simulate natural conditions. Two PVC tubes each were filled with 75-100 cotton stalks and nightshade stems. The emergence tubes were checked for fleahopper nymphs two times per week beginning in late February each year. Emerged fleahopper nymphs were identified using a binocular microscope.

Additionally, cotton stems were collected from fields in late March of 2002-2004. Each year approximately 50-75 stems were inserted into each of four PVC tubes as described above. The tubes were checked twice per week for emergence of fleahopper nymphs beginning the first week after collection. The purpose of this collection was to study emergence from stems that had over wintered under natural conditions.

RESULTS AND DISCUSSION

Small numbers of both adults and nymphs of the cotton fleahopper were found in primrose in late April of 2000 (Fig. 1a.). Numbers of nymphs averaged between 1.5 and 3.0 per 30-second sample until 10 May, when they increased to 23.0. Numbers of adults steadily rose from a low of 1.0 to a high of 34.0 on 17 May. After peaks of adults and nymphs were reached, numbers dropped until sampling was discontinued after 24 May. These data indicate that nymphs emerged during late April-early May, so more nymphs than adults would be detected during this time period. Adult fleahoppers were not found in the vacuum samples from gaura until 10 May. The peak was ten fleahoppers per 30-second sample, much lower than in primrose. The capture of nymphs in gaura averaged from one to two fleahoppers per week for the 2000 sampling season. Horsemint was not sampled in 2000.

Sampling began in mid-April in primrose and gaura in 2001 (Fig. 1a). Adults were not found until 26 April in primrose, and their numbers peaked a week later at 49.0 fleahoppers per 30-second sample. Numbers decreased until only two were found on 17 May. Numbers of nymphs in primrose were almost non-existent for the sampling season of 2001. Only a small number of adults and nymphs were found in gaura. Sampling in primrose and gaura was discontinued after 17 May. Sampling in horsemint began on 17 May when 91.0 adult fleahoppers were collected, and numbers peaked at 174.0 on 14 June. That horsemint may support large numbers of fleahoppers agrees with the findings of other researchers (Fletcher 1940, Almand 1974). Nymphs were found only in small numbers in horsemint with a peak of 9.5 on 24 May. On all other sample dates, numbers of nymphs were near zero. Almand et al. (1976) found a similar trend although their peak numbers were higher, which may have been a result of a different sampling technique.

Sampling in 2002 (Fig. 1b) began in primrose on 16 April, and the highest number of adult fleahoppers was found on 14 May which was similar to the year 2000. Few nymphs appeared in primrose in 2002 with a high of 6.5 recorded per 30-second sample on 23 April. From initial samples taken 17 April, the number of adult fleahoppers in gaura increased to a peak of 44 on 14 May. Nymphs in gaura peaked on 23 April. Samples yielded no fleahopper nymphs from 7 May to 28 May, but a small number appeared in the first and second week of June. Very few fleahopper adults and nymphs were found in stork's-bill during the 2002 spring season. The largest number of adults (150.0) was found in horsemint on the first day of sampling (14 May). Nymphs appeared for two weeks with 44 collected on 28 May and 21 on 4 June.

In 2003 (Fig. 1b) adult fleahoppers in primrose increased for approximately a month from 9 April to 7 May, then decreased for two weeks. A resurgence of numbers was observed the last two weeks of the sampling season when the peak in numbers was observed. Numbers of fleahopper nymphs in primrose increased to a peak about three-quarters of the way through the season on 21 May. Adults in gaura reached their highest

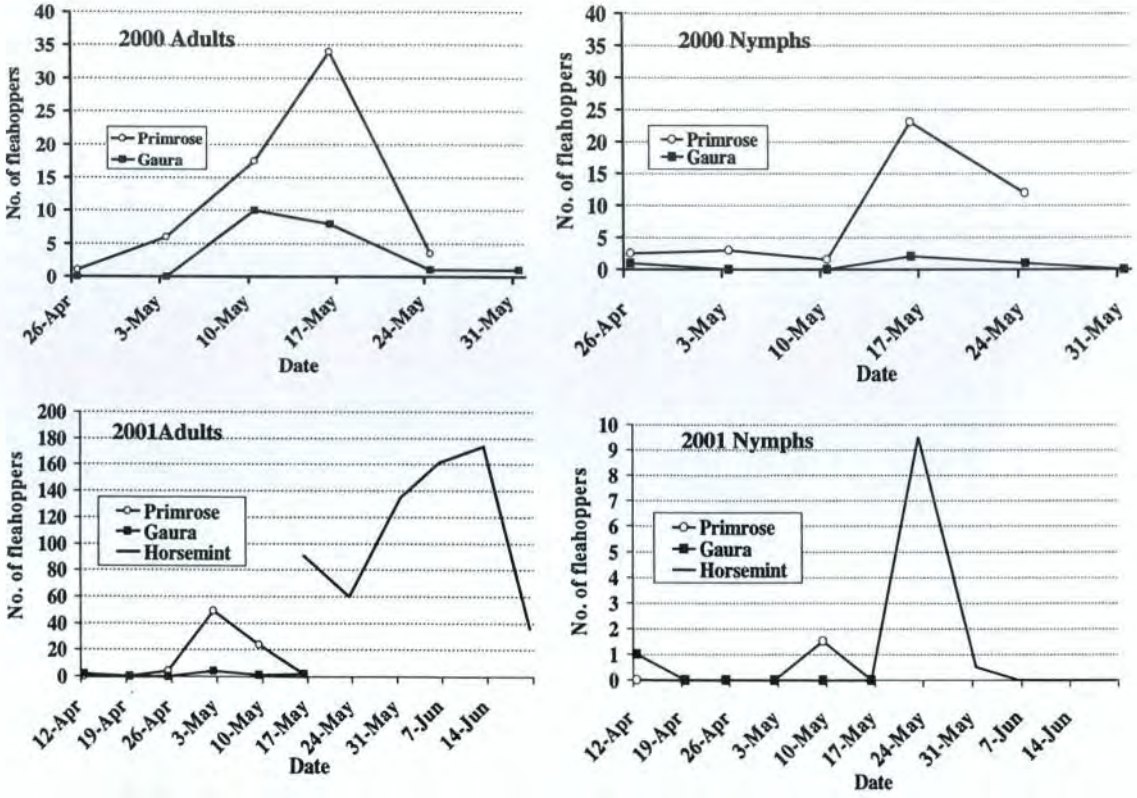

FIG. 1a. Cotton fleahoppers in vacuum samples of spring weeds 2000-01.

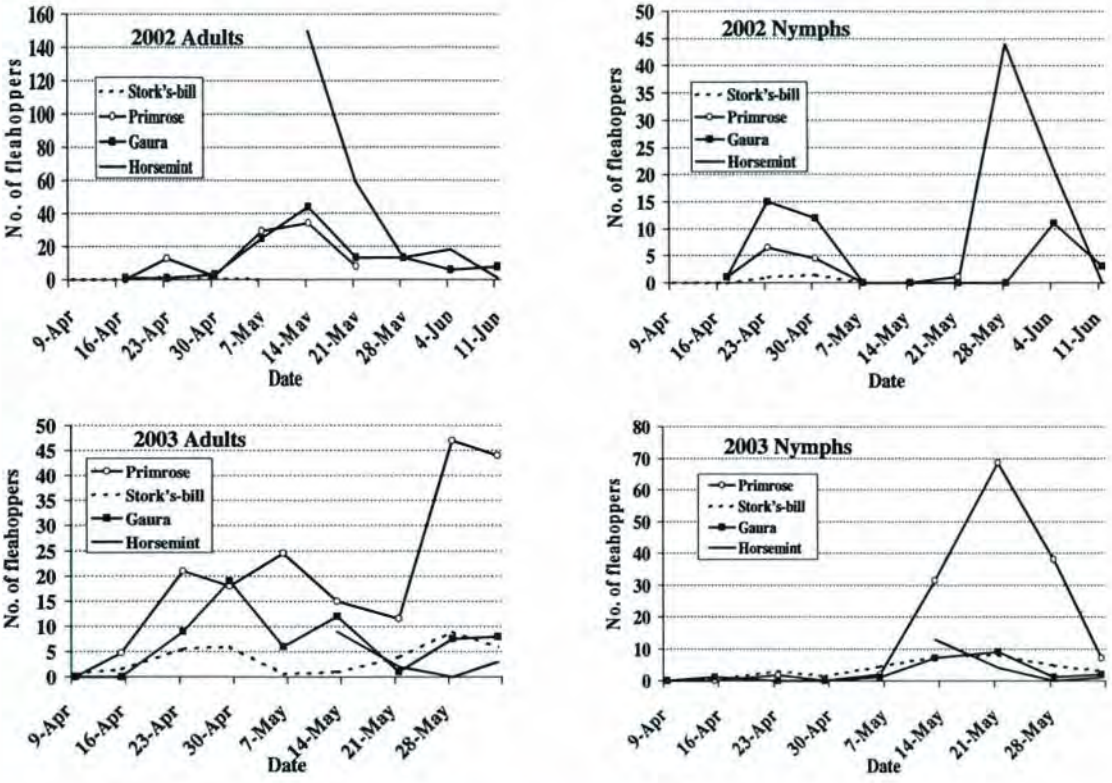

FIG. 1b. Cotton fleahoppers in vacuum samples of spring weeds 2002-03.

numbers on 30 April while nymphs did not appear until 14 May. Overall numbers of fleahopper nymphs were low relative to primrose, the highest being nine on 21 May in the 2003 samples. Fleahopper adults and nymphs were more prevalent in stork's-bill in 2003 than in 2002. The two highest numbers of adults appeared a month apart on 30 April and 28 May, the largest number of nymphs occurred on 21 May. However, the total number of adults and nymphs found in the vacuum samples was much smaller for stork's-bill than for primrose and gaura. The number of adults and nymphs caught in horsemint during 2003 was much smaller than in the previous two years of sampling. The largest number found in the vacuum samples was on 14 May, the first day of sampling in horsemint.

The active growing period for spring weeds was longer in 2004 than in previous years of this study (Fig. 1c). Primrose sampling began on 5 April and ended on 30 June. The peak number of adult cotton fleahoppers (240.0) was found on 1 June. There were more nymphs in the samples than in previous years, and the largest number was found on 26 May. Again, the number of fleahoppers in gaura was lower than for primroses. Fleahopper adults in gaura peaked on 1 June with an average of 66.5 per 30-second sample. The largest number of nymphs caught was 25.5 on 26 May. Many of the nymphs in stork's-bill may have died, given that a high number of 28.5 was caught on 5 April and only a very small number appeared in subsequent weeks as adults. The plants appeared to be suitable hosts through mid-May, but the reason for absence of adults was not determined. The numbers of fleahoppers in horsemint followed the trend of 2001 and 2002 with more collected than in samples of other host plants studied. However, the peak numbers of adults occurred at the same time as in primrose and gaura.

The timing of the appearance and disappearance of cotton fleahoppers in spring host (Fig. 2) is important because this insect ultimately infest cotton. In 2000 and 2001, fleahoppers were found in primrose the third week of April and remained for about a month until the third week of May. In 2002 and 2003, they appeared in primrose beginning in the second week of April and remained until the third week of May and the first few days of June, respectively. During 2004, fleahoppers were found for a much longer period of time in primrose, from 5 April until 30 June. The spring of 2004 was cool and wet, and these conditions apparently allowed the spring host plants to persist longer than in previous years.

Fleahopper persistence in gaura was similar to that in primrose. In 2001, a small number of fleahoppers appeared earlier in gaura than in primrose. Stork's-bill was also a host for fleahoppers beginning in April. In 2002, the stork's-bill plants matured and sampling was discontinued by 11 May, and the fleahoppers dispersed to other hosts or died. The numbers of fleahoppers caught in stork's-bill were generally very low. Horsemint stands yielded fleahoppers in early to mid-May in the four years that they were sampled and contained relatively large numbers of adults compared to the other spring weeds.

The time period from the last appearance of fleahoppers in spring weed hosts to their first appearance in cotton depended on weather conditions, which affected suitability of the spring host and the planting date of cotton. Ewing and Johnson (1925) found that the average longevity of the cotton fleahopper male was approximately 17.5 days and that of females was 15.5 days. In 2001, 2002, and 2004 the number of days from the last fleahoppers found in the spring weeds until the first fleahoppers found in cotton were 1, 10, and 12, respectively (Fig. 2). These periods would allow fleahopper adults ample time to migrate and infest cotton.

The number of fleahoppers that emerged per 100 cotton stalks collected in the fall (Fig. 3) declined from 2001 (3.3) through 2003 (1.5) and increased in 2004 (4.0). However, only 4.0 fleahopper nymphs emerged from a total of 888.0 cotton stems collected during late March 2002-2004. Over-winter survival was very low in the field (stems collected in the spring) compared to our survival records in cotton stems maintained in the insectary (fall collected stems). These results indicated, nevertheless, that fleahoppers do

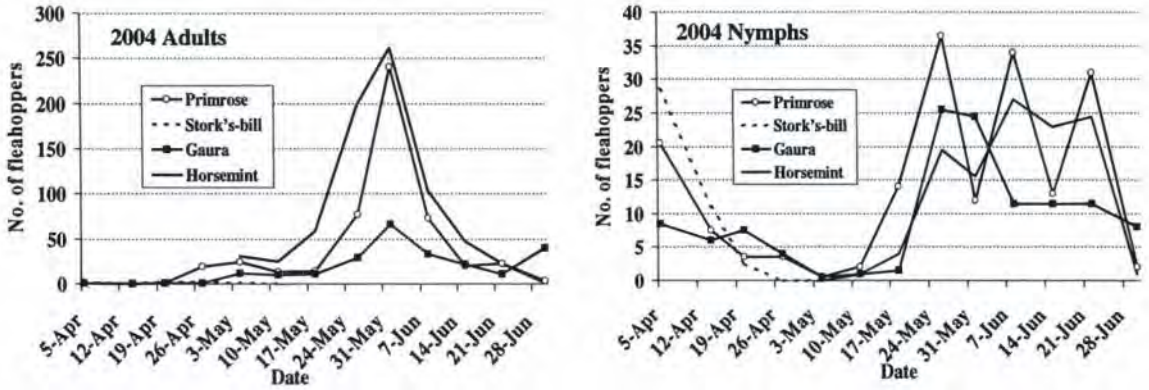

FIG. 1c. Cotton fleahoppers in vacuum samples of spring weeds 2004.

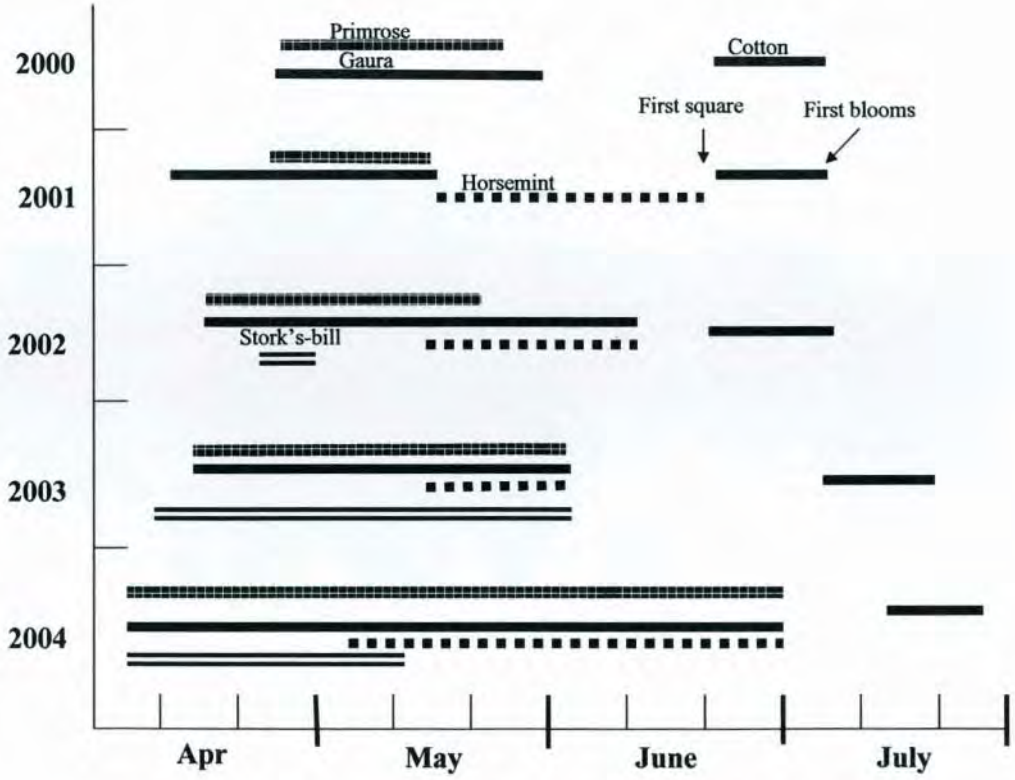

FIG. 2. Cotton fleahopper presence in vacuum samples in spring and early summer weed hosts and cotton

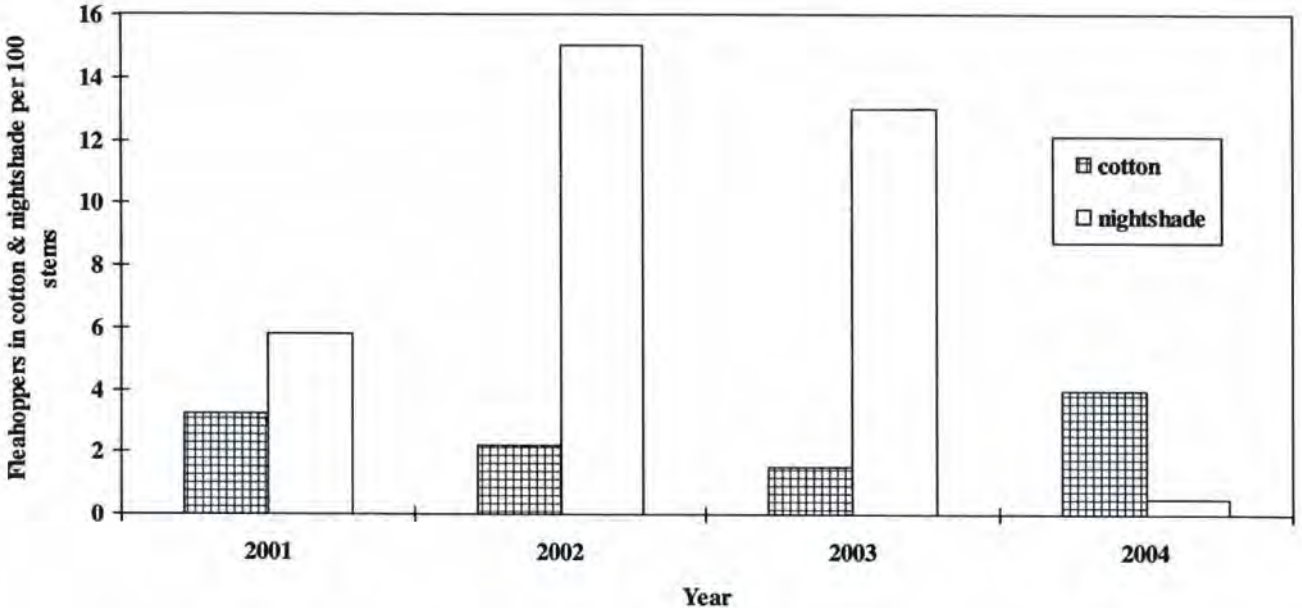

FIG. 3. Spring emergence of cotton fleahoppers from nightshade and cotton stems collected in the fall. Wilbarger Co., Texas.

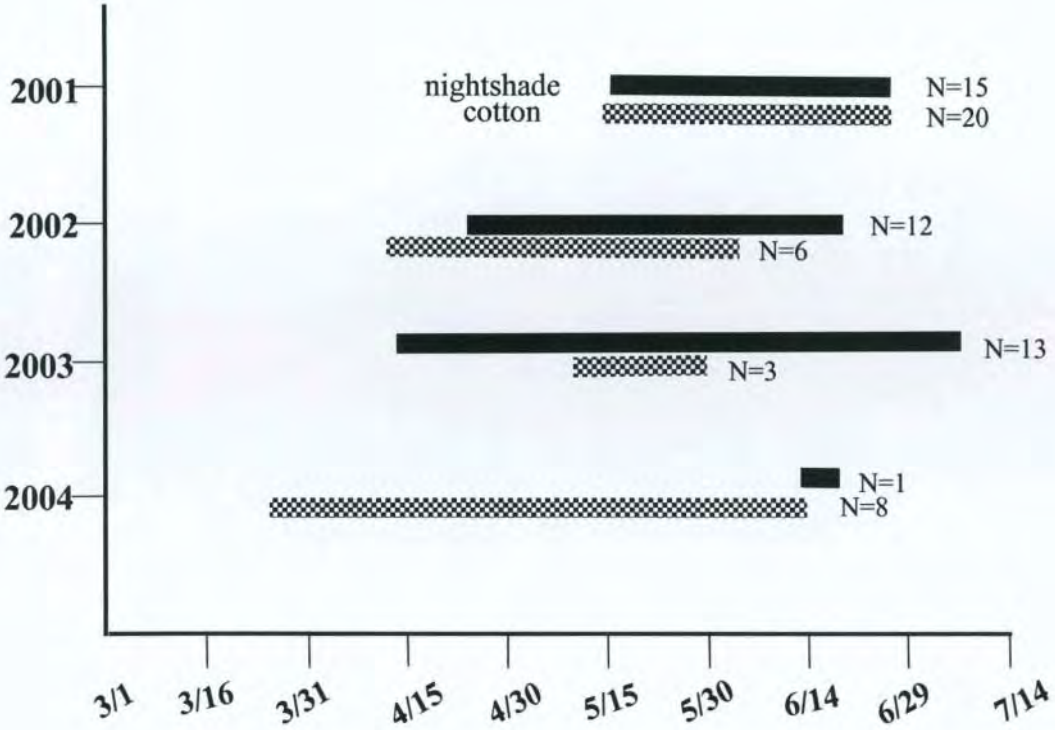

FIG. 4. Emergence periods of cotton fleahoppers from cotton and nightshade stems collected in the fall. Wilbarger Co., Texas.

survive in shredded cotton stems in the field. Earlier research indicates similar results (Reinhard 1928).

Fleahopper numbers coming from nightshade stems were 5.8 in 2001, increasing to 15.0 in 2002, dropping slightly to 13.0 in 2003, and then to 0.5 in 2004. The results of this study indicate that nightshade stems are a preferred overwintering host, but fleahoppers do survive in cotton stems.

Reinhard (1928) found that fleahopper eggs began to hatch in the spring when the temperatures reached 14-15°C. He observed that there was a relation between the time of maximum emergence and the extent of injury to cotton. He stated that if emergence of fleahoppers from hibernation occurs before cotton is up in the field, then little injury to the crop results. When climatic conditions delay emergence of the insects from hibernation in the spring and cotton is planted at the average date, conditions are favorable for injury to the crop by the fleahopper. The latter may have been the case in this study. Fleahoppers began to emerge from cotton stalks (Fig. 4) as early as 26 March in 2004 and, continued to emerge as late as 26 June in 2001. They began to emerge from nightshade stems on 14 April 2003, and the last ones emerged on 7 July in that same year. This was the longest period of emergence recorded in this study. Because the emergence period from these two sources can extend into the early weeks of the new cotton crop, it is apparent that both cotton stalks and nightshade stems can be a source of fleahopper infestation for a new cotton crop.

In summary, based on our sampling technique, horsemint vacuum samples contained the highest number of fleahoppers, except for the year 2003. Primrose was favored over gaura as a host by the cotton fleahopper. Stork's-bill was the least favored host for the three years that it was sampled. In three of the five years that the interval between the last fleahoppers found in the spring weeds and the first fleahoppers found in cotton was studied, the interval was short enough to allow infestation of a new cotton crop. In conservation tillage systems, cotton stalks are generally shredded after mid-November, and the shredded stalks are left on the soil surface. This practice occurs in fields planted to cotton following cotton and in wheat fields planted into cotton stubble. Since overwintering fleahopper eggs are oviposited between early October and the occurance of frost (Reinhard 1928), shredding probably destroys many of these eggs. However, our study shows that fleahoppers can survive the winter as eggs in the cotton stalks left after harvest, and nymphs can emerge late enough in the spring to infest the following year's cotton crop. Overwinter survival of fleahoppers in cotton stalks and nightshade stems in conservation tillage systems is especially important when spring host plants are allowed to grow within the fields. In such a case, emerged nymphs have a readily available host plant. The nightshade, primrose, gaura, stork's-bill and horsemint plants that grow in spring near cotton fields or in wheat and other nearby land are hosts for fleahoppers which can ultimately infest the cotton plants and cause damage.

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SURVEY RESULTS FOR THE SUGARCANE PEST, *BLASTOBASIS GRAMINEA* (LEPIDOPTERA: COLEOPHORIDAE), IN TEXAS AND LOUISIANA IN 2002

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ABSTRACT

Recent discoveries of *Blastobasis graminea* Adamski (Lepidoptera: Coleophoridae) in Mexico have prompted interests that this insect pest, originally discovered in South America, may be moving northward. A survey in Texas and Louisiana was conducted in 2002 to determine if *B. graminea* has extended its range into the U.S. Surveys included five nights of blacklight trapping in Texas and three nights of blacklight trapping plus diurnal surveys of 23 fields in Louisiana. Field surveys in Louisiana included examination not only of sugarcane (interspecific hybrids of *Saccharum* spp.) but also of maize, *Zea mays* L., and sorghum, *Sorghum bicolor* (L.) Moench, as well as non-cultivated gramineous species. We did not collect *B. graminea* during blacklight trappings nor did we detect it in association with cultivated host species (i.e., sugarcane, maize, and sorghum). We did, however, discover *B. graminea* in smooth cordgrass, *Spartina alterniflora* Loisel., in Louisiana in a non-agricultural environment. Finding *B. graminea* only in cordgrass suggests the possibility that rather than expanding its geographic range, *B. graminea* may be a widespread tropical species that is expanding its host range from native grasses to sugarcane. Finding *B. graminea* in Louisiana represents a new U.S. record for the species. Also, from this survey, new U.S. hosts records for *Pyroderces badia* (Hodges) (Lepidoptera: Cosmopterigidae) and *Dicymolomia julianalis* (Walker) (Lepidoptera: Crambidae), moths reared from grain sorghum seedheads, are documented.

INTRODUCTION

Introductions of new insect pests are a major concern for domestic agribusinesses. The mainland U.S. cane sugar industries have experienced several significant insect invasions within the last 20 years. The Mexican rice borer, *Eoreuma loftini* (Dyar) (Lepidoptera: Crambidae), which originated from Mexico, was first detected in the U.S. in 1980 (Johnson 1981) and is now the key pest of sugarcane (interspecific hybrids of *Saccharum* spp.) in the Lower Rio Grande

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Valley (LRGV) of Texas and has also become a serious pest of rice, *Oryza sativa* L., in the Texas rice belt (Way and Reagan 2001). The Mexican rice borer has not been reported in other sugarcane producing states. The sugarcane delphacid, *Perkinsiella saccharicida* Kirdaldy (Homoptera: Delphacidae), is a recent introduction to North America having been reported in Florida in 1982 (Sosa 1985), Texas in 1989 (Meagher et al. 1991), and Louisiana in 1994 (White et al. 1995). The principal concern for this insect is its ability to be a vector of *Fijivirus* sp., the causal agent of Fiji disease - a devastating disease of sugarcane not yet reported from the western hemisphere. Meagher et al. (1991) also reported the sugarcane lacebug, *Leptodictya tabida* (Herrich-Schaeffer) (Hemiptera: Tingidae), in Texas after earlier discoveries in 1910 (Drake 1925). It was reported for the first time in Florida sugarcane in 1990 (Hall 1991). The sugarcane aphid, *Melanaphis sacchari* (Zehntner) (Homoptera: Aphididae), is a widely distributed insect reported in the continental U.S.; it was reported on sugarcane in Florida in 1977 (Mead 1978) and in Louisiana in 1999 (White et al. 2001). The sugarcane aphid is also a vector of several important sugarcane viruses. Since the early 1950s, *Blastobasis graminea* Adamski (Lepidoptera: Coleophoridae) has been known as a pest of sugarcane in Colombia and Venezuela (e.g., Box 1953, Cárdenas et al. 1985, Guagliumi 1962, Adamski 1999). In recent years it has been documented from sugarcane fields in Costa Rica and from Veracruz and Jalisco, Mexico (Adamski et al. 2002). Because the species appears to be a general feeder on many monocotyledonous plants, it is uncertain whether *B. graminea* is actually an invasive South America species that is slowly moving northward, as suggested by collection data, or an extremely widespread tropical species that is slowly switching from native grass hosts to sugarcane throughout portions of its range where sugarcane is available. Because of its possible economic impact to sugarcane production in the southeastern United States and its apparent northward range expansion, we conducted survey work in Texas and Louisiana during 2002 to determine its presence/absence there. The purpose of this paper is to present the results of that survey.

MATERIALS AND METHODS

We conducted surveys for *B. graminea* at 31 sites in Texas and Louisiana from 22 July - 3 August 2002 (Fig 1). Only blacklight sampling was conducted in Texas. These surveys were conducted in two distinct geographical areas. The first was the Lower Rio Grande Valley (LRGV), an area with approximately 14,000 ha of sugarcane. The second was near Beaumont, TX, an area of traditional rice culture. We chose to survey in east Texas because approximately 400 ha of sugarcane have been under cultivation there since 2000. A portion of this cane is shipped to Louisiana for grinding. Survey sites in East Texas included the Texas A&M University Agricultural Research and Extension Center near Beaumont, TX.

Diurnal survey sites in Louisiana were sugarcane fields in active production, but one abandoned sugarcane field also was surveyed. Blacklight survey sites in Louisiana included a commercial cane field; the Louisiana State University, Louisiana Agricultural Center, St. Gabriel Research Farm, St. Gabriel, LA; and the USDA, National Resources Conservation Service, Plant Material Center, Golden Meadow, LA. The sites were selected arbitrarily, but are believed to constitute a representative sample of the Louisiana sugarcane industry.

During diurnal surveys we chose a strategy of surveying as much geographical area as possible during the time frame available to us. At each site we physically searched for Lepidoptera larvae and larval damage not only in sugarcane but in adjacent stands of monocots during the daytime hours. We remained at each site approximately 30 min (five surveyors x 30 min = 2.5 h searching per site). Larvae found were placed in 30-ml plastic cups with cuttings of the plant upon which they were discovered. Larvae were taken to the laboratory within 24 hr of collection where they were dissected from plant cuttings and reared to adulthood in a second, 30-

FIG. 1. Location of survey sites and black light sites within the Texas and Louisiana sugarcane industries.

ml plastic cup with 10 ml of sugarcane borer rearing media (Southland Products™, Lake Village, AR). Cups were held in a growth chamber at 26 ° C and 14/10 light/dark cycle. Blacklight surveys were conducted using a 15-watt ultra-violet light hung in front of a white sheet at night (ca. 2100-2300 hr) at five locations in Texas and three locations in Louisiana.

Adult and larval specimens were identified by systematists from the Systematic Entomology Laboratory, National Museum of Natural History, Smithsonian Institution, Washington, D.C. Vouchers of all species are deposited in the National Museum of Natural History and at the USDA-ARS Sugarcane Research Unit, Houma, LA.

RESULTS

Table 1 summarizes the results of the survey. Specimens collected from the same host or sampling method are reported together. Locations of where specimens were collected can be crossed referenced to Fig. 1.

Seventy-five larvae were collected from direct field dissections of infested sugarcane stalks. All of the larvae successfully completing development were identified as sugarcane borer, *Diatraea saccharalis* (F.).

One larva was collected from infested sorghum, *Sorghum bicolor* (L.) Moench, stalks and 12 were collected from infested maize, *Zea mays* L. stalks. All larvae that completed development were identified as sugarcane borer. Over 90 larvae and pupae were collected from grain sorghum seed-heads. Two species were identified from these: *Pyroderces badia* (Hodges) (Lepidoptera: Cosmopterigidae) and *Dicymolomia julianalis* (Walker) (Lepidoptera: Crambidae: Glaphyriinae). Sorghum is a new host record for both species in the U.S. The *P. badia* larvae appeared to be causing extensive injury to the seed-heads.

Four larvae were collected from vaseygrass, *Paspalum urvillei* Steud. Larvae reared to adults were identified as *Diatraea evanescens* Dyar. Five larvae were collected from smooth cordgrass, *Spartina alterniflora* Loisel., at the USDA, NRCS Plant Materials Center, Golden Meadow, LA. We were unable to rear any of these larvae to adults; however, larvae were identified as crambids (Crambidae: Schoenbiine). Two of these larvae were parasitized. Additional larval specimens collected at a later date (27 August) also failed to complete development. However, at this later date we collected an additional 13 larvae of a species that were not collected during the first survey of the cordgrass. Nine of these larvae were reared successfully to adults on sugarcane borer diet and identified as *Blastobasis graminea* Adamski (Lepidoptera: Coleophoridae). Finding *B. graminea* in Louisiana represents a new U.S. record for the species.

A number of microlepidoptera specimens were collected during light trapping in both Texas and Louisiana. Although there were several Blastobasini, none were *B. graminea*. We collected one specimen of the Southwestern corn borer, *Diatraea grandiosella* Dyar, while blacklighting near New Iberia, LA.

DISCUSSION

Although we did find *B. graminea* during the Louisiana phase of our survey, we did not detect the insect in the sugarcane agroecosystem. Further sampling will be necessary to determine if *B. graminea* has made the host shift into cane. Two significant examples of successful host shifts from native plants to agricultural plants were by the stemborers *Eldana saccharina* Walker (Lepidoptera: Crambidae) in Africa and the sugarcane borer in the Americas. Both moved from a riparian habitat to cultivated sugarcane (Wiedenmann and Smith 1999). Local climatic conditionsmay play some role in this ecological phenomenon. In Mexico, *B. graminea* was found in sugarcane in the arid state of Jalisco, but only from blacklight samples in the tropical state of

TABLE 1. List of Specimens Collected during Survey of Texas and Louisiana for the Sugarcane Pest *B. graminea*, 29 July - 3 August 2002.

Sample method	Host or Blacklight site	Taxon	Number	Location	Pest status	Comments
Dissections	Sugarcane	*D. saccharalis*	75	2-4, 6-8,10,11, 13,14, 17,19 22,23	Key pest	Only LA cane borer, Secondary pest of cane in LRGV of TX, but occasional pest of rice
"	Sorghum (stalk)	*D. saccharalis*	1	16	Occasional pest	
"	Sorghum (seed head)	*Pyroderces badia*	96	16	Unknown	New host record
"	"	*Dicymolomia julianalis*	1	16	Unknown	New host record
"	Maize	*D. saccharalis*	12	21	Key/occasional	
"	smooth cordgrass	Schoenbiinae	5	blt2[a]	Unknown	Cordgrass
"	"	*B. graminea*	13	blt2[a]	Unknown	New U.S. record
"	Vaseygrass[b]	*D. evanescens*	4	1	Non-pest	Sugarcane in FL
Blacklight	Texas	Blastobasini	57	blt1, blt3	Unknown	Species unknown
"	"	Torticoidea	17	blt5	Unknown	Species unknown
"	Louisiana	Blastobasini	1	blt 2	Unknown	Species unknown
"	"	Gelechiidae	11	blt 2	Unknown	Species unknown
"	"	*D. grandiosella*	1	blt 1	Maize pest	Sugarcane in Texas

[a]Both blacklight and diurnal samples were made at this location.
[b]One specimen of *Eoreuma densella* identified in the personal collection of V.A. Brou, Louisiana Lepidoptera Survey, Abita Springs, LA.

Veracruz (Villanueva-Jiménez et al. 2002). We plan to do further survey work in the coastal areas of Louisiana, particularly areas where native, coastal grasses impinge into cane culture. It also will be important to conduct diurnal sampling in Texas, where only black light sampling was conducted.

Although pests surveys are routinely conducted in both the Louisiana and Texas sugarcane industries, this work represents the most inclusive stemborer survey of the Louisiana sugarcane agroecosystem conducted since 1962 (Agarwal 1963). Results of our survey in Louisiana indicate that the sugarcane borer continues to be the only stemborer occurring on sugarcane in Louisiana. *Diatraea evanescens* has been collected from sugarcane in Florida, but we found it only in vaseygrass in Louisiana. Agarwal (1963) also reported *D. evenescens* only from vaseygrass. The Southwestern corn borer, also reported from sugarcane in Mexico and Texas (Agarwal 1963), was not found in sugarcane, maize, or grain sorghum during our diurnal surveys. The one specimen we collected was from light trapping in the proximity of a sugarcane field near New Iberia, LA. None of the specimens of southwestern corn borer in the Louisiana State Arthropod Museum, Department of Entomology, Louisiana State University was collected from sugarcane or this far south in the state.

No specimens of the Mexican rice borer were collected in Louisiana. A related species, *Eoreuma densella* Zeller, has been reported from vaseygrass and broomsedge, *Andropogon virginicus* L., in Louisiana (Agarwal 1963). We did not collect this species, but did identify a specimen from the personal collection of V.A. Brou, Louisiana Lepidoptera Survey, Abita Springs, LA. The Mexican rice borer may become a greater economic pest than the sugarcane borer in Louisiana. It has been found approximately 100 km from a small commercial cane operation in east Texas that is shipping cane to Louisiana for milling.

The potential impact of *B. graminea*, if it is found on cane in Texas and/or Louisiana, remains unknown. Currently it is regarded as a pest of minor importance in both Colombia and Venezuela. *Blastobasis graminea* generally is found in association with the sugarcane borer and therefore it is difficult to separate the damage caused by the two species. The immediate concern for *B. graminea* may be its impact on smooth cordgrass, which is being propagated for distribution in wetland stablization projects. Although *B. graminea* may be of limited importance as a pest of smooth cordgrass, the movement of infested cordgrass may serve as a important source for *B. graminea*, facilitating a possible host shift into sugarcane.

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PLANT GROWTH PARAMETERS OF MUSK THISTLE *CARDUUS NUTANS* [1] AND EGG DISTRIBUTION PATTERNS OF *RHINOCYLLUS CONICUS* [2] ON THEIR BLOOMS

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ABSTRACT

Plant growth phenology of musk thistle, *Carduus nutans*, L., in Oklahoma was three weeks ahead of patterns previously reported from Virginia. For example, plant dormancy ended in late February to early March, bolting initiated in early April, anthesis began in middle to late May and plant senescence occurred in early July. The musk thistle head weevil, *Rhinocyllus conicus* Froelich, was present when bolting occurred and was active for five and one-half weeks, ovipositing for up to four weeks. In a two-year study, preference sites for ovipostion by musk thistle head weevils were heads on the outside and top of plants. In 2001, surveyed locations had been infested with weevils for six or more years with larger weevil populations, and greater numbers of eggs present. Checking the top, outside heads for egg numbers will allow growers a focused, more decisive method in determining levels of weevil infestation. Irrespective of ovipositional preference, redistribution of adult weevils can occur when 50% of thistle heads contain at least six eggs.

INTRODUCTION

In Oklahoma, musk thistle *Carduus nutans* L., is an exotic invasive plant causing serious problems for landowners, managers, and environmental specialists. Presence of musk thistle in Oklahoma was first recorded in Payne County in the 1940s. By 1960, it had spread through 29 counties in northeast and central Oklahoma (Stritzke et al. 1999) and by 2001, musk thistle was reported in 61 of 77 counties (Medlin et al. 2003).

Musk thistle competes well with desirable plants, reducing economic values of fields and pastures and the crops they produce. One *Carduus* plant on 1.49 m^2 (6711 plants per ha) can reduce pasture yields by 23% (Trumble and Kok 1982) by competing for space, light, and nutrients. Musk thistle easily invades poor soils, overgrazed pasture areas, roadside ditches, abandoned areas, and crops (Medd and Lovett 1978).

A survey of Oklahoma growers revealed that the average amount of improved pasture for each producer farm ranges from 16 to 64 ha (New 1997). The average cost of

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controlling musk thistles for one year, using 2,4-D herbicide, is estimated at a minimum of $1.85/ha for 0.39 l of material per ha (not including the cost of application) (Medlin et al. 2003). In Oklahoma, there are approximately 2.9 million ha of improved pastures. Therefore, the statewide cost of controlling musk thistle with this herbicide for 10 yr, if all improved pastures were infested, would exceed $131 million (Roduner et al. 2003).

In 1994, the Oklahoma state legislature declared musk thistle a locally noxious weed, upgrading that status to statewide in 2000. The noxious weed statute provides regulations on allowable densities and control timing. According to these regulations, a landowner must prevent musk thistle from producing seeds when thistle densities exceed 25 plants/ha (10 plants/acre). Once a landowner is determined to be out of compliance, fines are set at $1,000 per day. The law also provides counties with authority to apply herbicides on private property and assess those costs to a landowner's property taxes (Oklahoma Noxious Weed Law Rules 2000).

Studies done concurrently at the same location and with similar densities of musk thistle showed that plant populations were many times the legal limit. Plant densities as high as 50,000 plants/ha (20,243 plants/acre) were recorded in several counties (Roduner et al. 2003). Densities of this level excluded all other plants and rendered the land useless.

In light of the Noxious Weed Law, control of musk thistle has been a priority for many farmers and Extension Educators. During the summers of 2000 and 2001, landowners and producers attending meetings that provided information on musk thistle expressed frustration with the available data on plant growth and musk thistle head weevil, *Rhinocyllus conicus* Froelich, activity. As part of informal conversations, these landowners commented that thistle growth and head weevil activity timelines in current fact sheets were at least two (and possibly four) weeks later than what they actually observed in Oklahoma fields. Therefore, early growth and development of weeds and early controls (herbicides or weevil releases) were significantly later than desired. Growers requested an updated plant growth timeline and an easy method to determine if head weevils were present in their fields. They felt this information would help them accurately time thistle control and monitor head weevil activity.

Extensive research has been conducted on musk thistle and its predators in Virginia, Missouri, Montana, and Nebraska. Each of these states has a different climate than Oklahoma (National Climatic Data Survey 1988). Table 1 compares the weather differences between these states, highlighting Oklahoma's unique climate. Comparisons of field observations and anecdotal reports to published phenological data on musk thistle from Virginia shows a different plant growth timeline in Oklahoma. Currently, data do not exist on soil temperature requirements, growth initiation, or growing degree day (GDD) calculations for musk thistle in Oklahoma. Patterns observed in Oklahoma vary enough from the published norm to justify studying plant growth parameters.

TABLE 1. Climatic Differences between Oklahoma and Other States Conducting Musk Thistle Research and Education Programs.

State	$\bar{x}$ days @ Temp.		$\bar{x}$ wind speed (KmPH)	$\bar{x}$ annual rainfall (cm)	Month of highest rainfall
	< 0° C	> 32° C			
Missouri	103.5	37.5	16.25	104.9	evenly through year
Montana	166.5	22	16.9	33.5	Apr.-Sept.
Nebraska	176.5	37	15.6	47.7	May-July
Oklahoma	75.5	70	18.1	93.7	Apr.-June & Oct.-Nov.
Virginia	89.5	25.5	12.2	104.1	evenly through year

30 year averages (1961 – 1990) from the National Climatic Data Survey (1988).

MATERIALS AND METHODS

Two areas with dense musk thistle infestations were chosen to follow musk thistle growth from germination to senescence, to determine how the musk thistle reacts to Oklahoma climate, focusing on the time of plant bolting and weevil infestations. To avoid inaccurate data from transplant shock, only plants naturally germinating in the field were used. Local conditions such as intra- and interspecific competition, availability of water, and weather conditions were allowed to take their natural course in these areas. Plants behaving as true biennials and winter annuals were followed separately.

One musk thistle infested field site in Major County and two sites in Payne County were chosen for this study. Payne and Major counties were chosen to compare widely different growing conditions. Both areas in Payne County had good pasture or grass cover while the Major County site consisted of primarily bare soil. At each location, 50 or 100 plants were selected and marked. Seedlings selected were less than 75mm in diameter and possessed fewer than five leaves. Only vigorous plants were chosen, rejecting those appearing weak, damaged, or otherwise unfit. This selection process helped to insure long-term survival of enough plants to complete the study. Crowding or unique local conditions were noted. During the growing season, rosette diameter and numbers of leaves on each plant were recorded weekly. Measurements were discontinued during winter dormancy and restarted when growth resumed in the spring.

For each plant used in this study, the following data were recorded: date of bolt initiation, rate of stem elongation, first bloom, number of heads, musk thistle head weevil presence in spring, dates of oviposition by the weevil, and senescence of the plant. Whenever possible, the cause of plant death was recorded. When the plants had attained normal senescence, plant height, stem diameter, number of heads, and head location were recorded. Heads were then removed and taken to the laboratory where measurements were taken on head diameter, number of head weevil pupal chambers, and the presence or absence of seed.

Major County. An unused area between two fields was chosen for observation. This site had a very dense (> 4 plants/m^2) infestation of musk thistles and no head weevils, and was located 8.0 km west and 4.8 km north of Lahoma, OK. The owner allowed unlimited access to the land from May 2000 to July 2001. He actively participated by voluntarily canceling all planned herbicide treatments on his property to prevent damage to the experiments and fenced the area (0.21 ha) to prevent cattle damage once thistle plants began to bolt.

In May 2000, 100 spring-germinated seedlings were marked. Lack of rain delayed fall thistle germination until the first week in November 2000. On 10 November 10, 100 fall germinating plants were selected.

Payne County. The second study area (Sangre Bend Road) was a pasture, heavily infested with musk thistle. This site was owned by the First United Methodist Church of Stillwater, OK. This pasture consisted of various grasses, weeds, blackberries, and native trees. Cattle were present part of the year. Pasture management was conducted by the renter to maintain good feed for his cattle, but he expressed frustration with the density of musk thistle. All thistle plants were infested with head weevils. Thistle density varied throughout the pasture, with a mixture of clear areas and dense patches of thistles.

Initially, spring-germinated plants were marked in 2000 but were lost to a herbicide application by the renter. The site was unused for study until November of 2000 when 100 fall-germinated plants were marked and measured as described for spring-germinated plants. Fall-germinated plants were followed through August of 2001.

In April 2001, a second location owned by the City of Stillwater, at the junction of Airport and Jardot Roads, on the northeast edge of Stillwater, was chosen to replace the

spring germination site at Sangre Bend . The roadside ditch, measuring 6x800 meters, was heavily infested with musk thistles and head weevils. Plant density was 4.13/ m^2. Fifty plants, approximately the same diameter as spring-germinated plants in Major Co., were selected and marked on 12 April 2001.

Growing Degree Days (GDDs). Soil and air temperature data from the Oklahoma Mesonet (Brock et al. 1995) was used to compare weather conditions at all sites to determine when plants broke dormancy, bolted and developed heads. These data were then used to establish preliminary GDDs for each growth stage.

The following information was used to determine which factor(s) had the greatest influence on flower head numbers: numbers of leaves at bolting, rosette diameter at bolting, final plant height, number of heads, and final stem diameter. These data were analyzed using SAS correlation functions (SAS Institute 2000) to determine which had the most effect on head numbers per plant.

Head Weevil Activity. Two different factors were studied to determine head weevil activity. The first factor was the dates of weevil activity in the fields and areas surrounding the growth data plots. The second factor studied was egg distribution by female weevils during oviposition.

Based on field collection of thistle heads in infested areas, dates for the emergence of head weevils from winter dormancy, initiation and length of oviposition, and end of adult activity were recorded. These dates were then compared to dates from studies conducted in Virginia.

Egg Distribution Preference. Since eggs are oviposited on the undersides of flower bracts, they are visible and make verification of head weevil presence, after adult activity ends, very easy. Plants were studied to determine if weevils showed a preference for taller plants or plants with more blooms.

During the summers of 2000 and 2001, mature musk thistle plants were collected from heavily infested pastures. A total of 188 plants were removed from 12 sites in 9 counties (Adair, Cherokee, Craig, Delaware, Noble [3 sites], Nowata, Osage, Payne [2 sites], and Washington). All sites had a history of weevil releases in 1995 or 1996 (Bill Stacey, Area Extension Entomologist – personal communication). Weevils were present in moderate to large numbers (10-75 eggs per head) at all sites. At each site, large and small plants were selected to represent the range of sizes present (3-30 heads per plant). To avoid late-developing, uninfested heads, plants were collected within two weeks of the cessation of oviposition by head weevils. Plants missing multiple heads or lateral branches were rejected. Thistle plants were removed at soil level, measured, and numbers of heads per plant recorded.

Each head was removed and bagged separately, its position on the plant numbered according to McCarty (1982) (Fig. 1), and then frozen until processed. The diameter of each head was measured in mm at the receptacle base. Eggs on the underside of flower bracts were counted and recorded.

PROC MIXED and PROC REG (SAS Institute 2000) were used to determine numbers of eggs per mm of head diameter and ovipositional site on the plant, numbers of heads per plant, and plant size. To determine if head weevils showed a preference for tall or short plants or plants with more or fewer heads, plant height, head number, and eggs per head were analyzed using analysis of variance (ANOVA) and multiple regression with backward elimination.

RESULTS AND DISCUSSION

While many factors were monitored in this study, for the purposes of concise reporting, only plant growth and head weevil data will be presented in this paper.

General Plant Growth (Major County). Growth of individual plants varied, depending on the degree of intra-specific competition. Spring-germinated plants

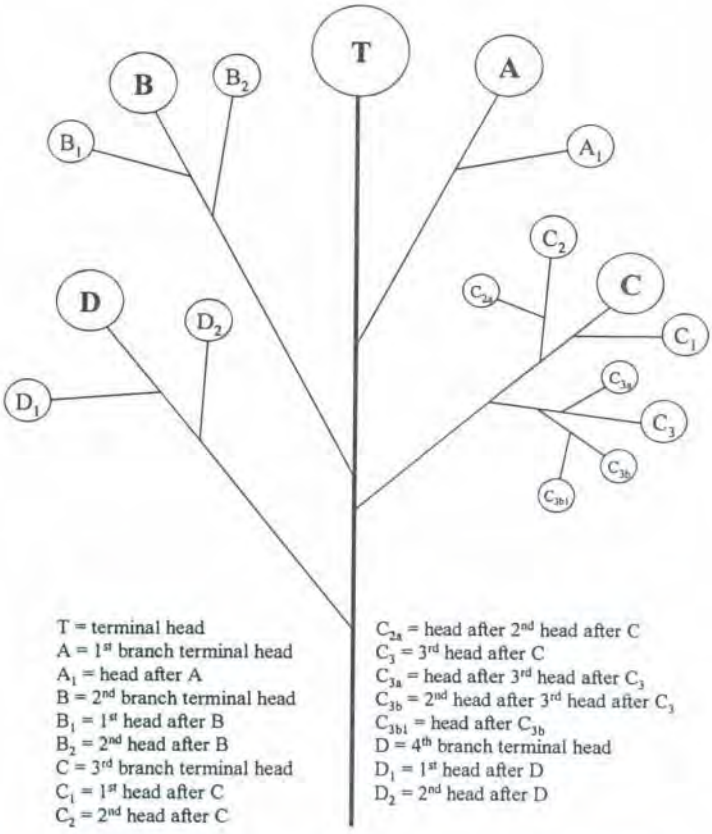

FIG. 1. Thistle head positions on plants. Adpated from McCarty (1982).

consistently added new leaves at a rate of one to four per month throughout the summer and fall. During the hot dry weeks of summer, rosette growth was temporarily halted. New leaves emerged from the rosettes forming a dense center, but did not elongate until after rain fell in October. In December 2000, plants entered dormancy

Bolting for both spring- and fall-germinated seedlings began during the second to fourth weeks in April, 2001. Maximum plant height was attained within seven to nine weeks after initiation of bolting (fall v/s spring). Fig. 2 shows the weekly and daily bolting rates for each location and germination time. Rosette growth ended once bolting began. As the flower stalk grew, rosette leaves dried, leaving a thick mat of leaf litter covering the ground. As plants began to senesce, leaves began to dry from the bottom upward. Seeds from individual heads were released quickly, over two to three days, with the majority falling near the base of the plants.

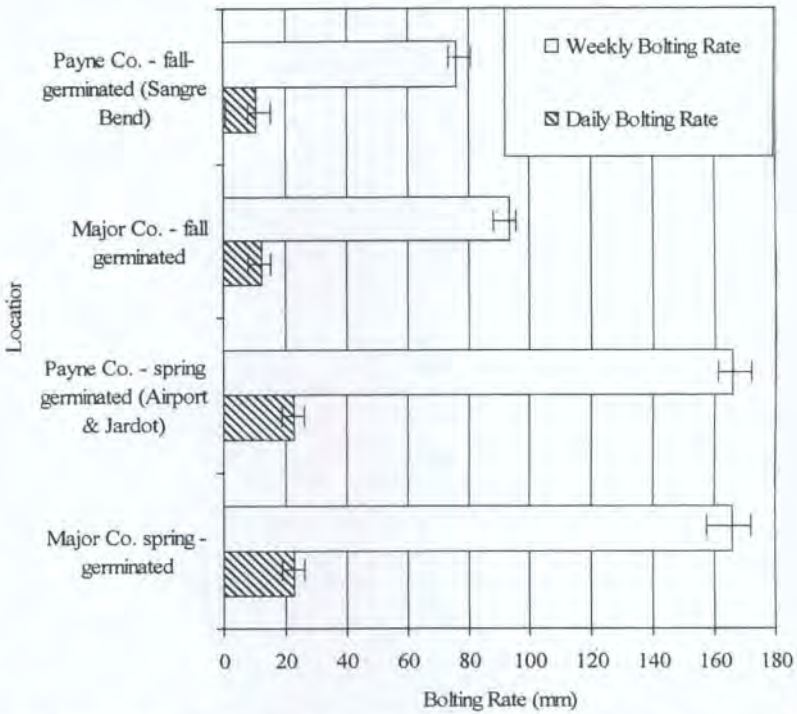

FIG. 2. Daily and weekly bolting rates (mean ± SE) of spring and fall germinated musk thistle plants in Major and Payne Counties, Oklahoma.

Spring-germinated plants. Germination occurred from late April to early June 2000. Selected plants germinated during the last two weeks of May 2000. After an ice and snow storm on 10 December 2000, plants entered dormancy. Plant monitoring resumed on 3 March 2001. Musk thistle plants initiated bolting throughout the month of April and maximum height was reached seven to ten weeks later. The first buds appeared between four and six weeks after initiation of bolting and bloomed two to three weeks later. Senescence began in late June with most plants dead or dying by July 2001.

Fall-germinated plants. Germination was delayed by dry weather in September and October, 2000. Heavy rains in late October, totaling 16cm, triggered germination. Plants were large enough to mark and measure by 10 November 2000, in densities ranging from 155-400 plants per m^2. Seedlings grew for five weeks before entering dormancy. Plants resumed growth and bolted on a parallel with the schedule of spring-germinated seedlings. Although bolting dates were similar (Table 2), fall seedlings bolted at a slower rate than spring seedlings (Fig. 2).

Fall-germinated plants that were crowded appeared to be unable to withstand hot, dry conditions compared with the more robust spring-germinated plants. Plants that germinated in the fall reached senescence a minimum of two weeks earlier than those that germinated in the spring.

Payne County (Sangre Bend). Fall-germinated seedlings emerged after rains in October and grew approximately five weeks before entering dormancy. Growth resumed in

TABLE 2. Growing Degree Days (GDD) and Soil Temperatures in 2000 and 2001.

Location	Plant Phenology	Date	GDD (° C)*			Soil Temp.	
Temp.			-1.1	1.1	4.4	@ 5 cm (°C)[a]	Diff.[b]
Lahoma	end of dormancy	2/22/00	503	308	160	11.1	
(Major Co.)		3/1/01	423	209	74	1.9	9.2
	bolting	4/7/00	1478	1049	682	13.3	
		4/11/01	1264	846	514	15.6	2.3
	bud initiation	5/2/00	2149	1600	1112	16.3	
		5/9/01	2164	1606	1134	21.0	4.7
	heading	5/16/00	2667	2048	1490	21.4	
		5/23/01	2732	2104	1563	20.1	1.3
Stillwater	end of dormancy	2/22/00	622	410	234	11.0	
		3/1/01	497	268	118	5.2	5.8
(Payne Co.)	bolting	4/10/00	1766	1314	902	14.3	
		4/17/01	1591	1127	746	13.8	0.5
	bud initiation	4/24/00	2182	1661	1179	17.0	
		5/1/01	2078	1544	1094	18.7	1.7
	heading	5/8/00	2676	2085	1533	21.8	
		5/15/01	2651	2047	1526	22.2	0.4

[a] Data obtained from the Oklahoma Mesonet.

[b] Soil Temperature difference between 2000 and 2001.

March 2001. Interspecific competition caused plants to develop very slowly and with a fragile structure. Dense pasture cover shaded the seedlings preventing more robust growth. Initiation of bolting did not begin until mid-April (Table 2) and continued through June. On 15 June 2001, the renter sprayed the pasture with a 2,4-D herbicide (formulation and concentration unknown). Plants inside the spray area were weakened already from competition and died within three weeks. Although marked plants outside the primary spray area were unaffected by the herbicide, intense interspecific competition caused very few plants to survive to bolting and even fewer produced viable seed.

Airport and Jardot. Marked plants initiated bolting the second week of April. The rate of bolting (166.2 mm per week or 23.7 mm per day) was similar to spring-germinated plants from Major Co. (Fig. 2). Bud initiation began three weeks after bolting, and continued until senescence. Grass and low-growing annual weeds were present but the thistles were able to compete with these plants. Senescence began the last week of June 2001 and progressed rapidly.

Growing Degree Days. Weather data from the Lahoma (Major Co.) and Stillwater (Payne Co.) monitoring stations of the Oklahoma Mesonet (Brock et al. 1995) were used to calculate GDDs from January 1 using base temperatures of -1.1° C, 1.6° C, and 4.4° C. The year 2000 was a warmer season than 2001; therefore, thistles bloomed approximately one week later in 2001. Dates were compared between 2001 and 2000 and GDDs were calculated for the end of dormancy, bolt, bud, and bloom times for 2000 and 2001 in both Major and Payne Counties. Using a base temperature of -1.1° C appears impractical, since air temperatures are below freezing. A base temperature of 1.1° C or 4.4° C seems more practical, because plants will continue growth at low temperatures and the ambient temperature is above freezing. In 2001, musk thistle broke dormancy after accumulating 209 and 74 GDDs, using 1.1° C and 4.4° C base temperatures, respectively. Either method illustrates the ability of plants to grow at low ambient temperatures.

Additional studies need to be conducted to confirm the number of GDDs needed to break dormancy in Oklahoma. The number of GDDs for bolting was quite variable (up to

200 GDDs). In contrast, the blooming period was more consistent, with less than 50 GDDs difference. Bud initiation using a base temperature of 1.1° C and 4.4° C was nearly identical at Lahoma in 2000 and 2001 and varied less than 120 GDDs at Stillwater.

Smaller differences in soil temperature (Table 2) compared to differences in GDD at the end of dormancy suggest that, at this time, plants appear to respond to air temperature rather than soil temperature. The initiation of bolting and bloom times occurred at similar soil temperatures for both years. In each year, bud initiation also did not appear to be related to soil temperature. Based on these observations, it appears that musk thistle plants appeared to respond to air and soil temperatures differently during distinct life stages.

Head Weevil Activity. Head weevil adults were present at all sites as soon as flower stems emerged. Head weevil eggs were present on flower buds within 10 days of adult emergence from hibernation. Adult weevils were present for approximately five and one-half weeks. Head weevils infested all terminal and most secondary heads. Tertiary and quaternary heads were infested if they developed while weevil adults were present.

Egg Distribution Preference. The analysis of egg deposition patterns provided a more accurate model of weevil-plant interactions. In both analyses (plant height and number of heads), the number of heads per plant was a more important factor than plant height. Table 3 shows the number of heads per plant significantly ($P<0.0001$) affected oviposition at each elimination step. When all factors were included in the analysis ($r^2 = 0.803$), for every additional head, 20 eggs were added to the total recovered. After performing the backward elimination, the number of heads per plant was the only important factor ($r^2 = 0.776$), contributing 9.27 eggs for each additional head.

TABLE 3. Results of ANOVA and Backward Elimination in Multiple Regression Analysis. Parameter Estimates ± SEM.

ANOVA	r^2	Factor[a]	Parameter Est.	p-value
All factors	0.803	HGT	-0.201 ± 0.129	0.1262
		HEADS	20.852 ± 4.492	<0.0001
		HGT2	0.0001 ± 0.00006	0.084
		HEADS2	-0.058 ± 0.035	0.103
		HGTHEADS	-0.007 ± 0.002	0.021
Elimination step 1	0.794	HEADS	16.54 ± 3.581	<0.0001
		HGT2	0.00 ± 0.00001	0.2865
		HEADS2	-0.044 ± 0.035	0.2006
		HGTHEADS	-0.004 ± 0.002	0.0802
Elimination step 2	0.789	HEADS	15.279 ± 3.387	<0.0001
		HEADS2	-0.056 ± 0.033	0.0918
		HGTHEADS	-0.002 ± 0.002	0.1548
Elimination step 3	0.781	HEADS	10.954 ± 1.589	<0.0001
		HEADS2	-0.034 ± 0.029	0.2492
Elimination step 4	0.776	HEADS	9.269 ± 0.659	<0.0001

[a] HGT - height in mm of individual plants; HEADS - number of heads on each plant; HGT2 and HEADS2 - give location and degree of angle for changes in regression line; HGTHEADS - interaction between height and heads

The interaction between numbers of heads per plant and plant height was significant only when all factors remained in the analysis. Plant height alone was the least significant factor ($P=0.1262$) and eliminating this parameter in step one removed the significance of

any interaction. Female head weevils deposited more eggs on thistle plants with the greatest numbers of heads. Therefore, concentrating control efforts on plants with greater numbers of thistle heads would help eliminate those contributing more head weevils to the overall population. Likewise, selecting collection sites and possible release sites could be centered on those areas with a greater concentration of thistle heads.

In Oklahoma, musk thistle head weevils have established successfully and occasionally experience population fluctuations due to weather or grazing pressure; however, these changes have been temporary. In Oklahoma, the synchrony between musk thistle head weevils and their host has been a major reason for the success of biological control release programs for this noxious weed. Combining biological control of musk thistle with properly-timed herbicide treatment, while increasing plant competition for healthful forage have all contributed to reducing noxious weeds and enhancing production of grazing livestock and the forages on which they rely. Results from these studies help in focusing sampling and redistribution efforts on thistle-infested areas that can support a greater number of head weevils.

SUMMARY

Following the development of musk thistle plants in Oklahoma for a full generation confirms a growth timeline different from Virginia (Surles et al. 1974). Bolting and blooming were approximately three weeks earlier and hot summer weather induced senescence six to eight weeks earlier. Head weevil adults emerged from the soil two weeks earlier than Virginia, but remained active approximately five weeks in both Virginia (Surles and Kok 1977) and Oklahoma. These differences verified grower observations and justified the need for rewriting thistle control and weevil capture and release recommendations. Accurate timing of thistle growth and weevil activity will allow growers to modify herbicide spray programs, mowing, and weevil collection and release programs.

Giving growers a reliable time frame to determine appropriate times for herbicide application, mowing, and weevil releases has been an important goal in this study. GDD verified the ability of musk thistle to resume growth as early as the end of February. Bolting, an important life stage in control measures, is more closely tied to soil temperatures than the resumption of growth. Growers who monitor meteorological data will be able to predict bolting and time herbicide applications.

Musk thistle survival depended, to some extent, on the site. Plants were able to thrive through Oklahoma's hot dry summers, insect damage, and moderate competition. Extreme crowding caused by thistles themselves (Major County) reduced both plant vitality and mature seed production. Competition from pasture grasses at the Payne County (Sangre Bend) site prevented fall-germinated seedling transition from juvenile to mature plants and seed production. Lower plant density at the Payne County (Airport and Jardot) site, appeared to give thistles a competitive edge, as they grew larger and produced more heads than the Major County and Sangre Bend sites.

Head weevils were present in both Payne County sites but not in Major County, but were present in surrounding areas. The ability to accurately predict the dates when head weevil activity should peak, by knowing when oviposition will occur, helps determine the proper time to collect weevils for redistribution to other areas of the state. Egg distribution showed a distinct preference by females for plants with larger numbers of blooms. Growers and producers, knowing that they can quickly check plants with large numbers of blooms for egg presence, are provided with a quick method to verify weevil presence after adult activity has ended.

These results are from one plant cycle only, but following musk thistle plants through additional generations will verify our finding, providing a more accurate model of

growth patterns in Oklahoma. Verifying GDDs, head weevil activity times and egg distribution preference will facilitate the construction of new and more precise thistle management plans for landowners.

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IMPACT OF CHLORPYRIFOS FOR PINK BOLLWORM[1] CONTROL ON SECONDARY PESTS AND BENEFICIALS

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ABSTRACT

Non-target effects of the insecticide chlorpyrifos for control of pink bollworm, *Pectinophora gossypiella* (Saunders), were assessed for populations of beneficial arthropods and secondary pests on Acala cotton in New Mexico for 2001 and 2002. Treatments consisted of an untreated check, early-, mid-, late-season, and season-long applications of chlorpyrifos applied at 10-day intervals. Populations of cotton aphids (*Aphis gossypii* Glover), pink bollworms, beneficials, and phytophagous plant bugs were estimated. Aphid populations were low both seasons, while late-season pink bollworm infestations reached economic levels. Chlorpyrifos applications did not appear to affect the seasonal abundance of the majority of predaceous Heteroptera, plant bugs, or lacewings. Only populations of *Geocoris* and spiders were significantly less in the season-long chlorpyrifos treated plots than the control. Lady beetle numbers were significantly lower in the early- and mid-season treatments than the late-season, season-long, and untreated check. This is likely credited to low aphid populations building just prior to initiation of the late-season treatment. Results indicate that repeated use of chlorpyrifos in southern New Mexico during typical light aphid infestations should not result in an increase of this pest. Possible reasons for these results are discussed.

INTRODUCTION

In recent years, New Mexico cotton has been greatly impacted by the boll weevil, *Anthonomus grandis grandis* Boheman, and the pink bollworm, *Pectinophora gossypiella* (Saunders). Part of the success of these non-native insects may be attributed to their feeding habits, larvae of both species feed within developing squares and bolls where they are protected from insecticides and most beneficials. These pests are targets of separate ongoing eradication projects in the state.

The boll weevil eradication project (BWEP) was initiated for south central New Mexico in 1998 and eastern New Mexico in 2000 and 2001, and the pink bollworm eradication project (PBWEP) was initiated in 2002 for south central New Mexico. The BWEP has utilized malathion for control of adult weevils. The use of this insecticide often results in increased populations of cotton aphids, *Aphis gossypii* Glover, and whiteflies (Layton et al. 1999). These pests, generally maintained at subeconomic levels

[1]Lepidoptera: Gelechiidae

in New Mexico, often develop extremely large populations with the sustained insecticide use required in BWEP and implicated in a PBWEP (Leonard et al. 1999), becoming increasingly important as late season pests because of the resulting "sticky" cotton (Henneberry et al. 2000). Sticky cotton is difficult to gin, and in many areas is either docked significantly or not accepted. In contrast, the PBWEP utilizes chlorpyrifos, a broad spectrum organophosphate insecticide, to target adult mating flights.

Chlorpyrifos has longer residual activity than malathion. Its efficacy on cotton aphids appears to be variable, depending upon the host plant and environment (Godfrey and Fuson 2001). Although populations of beneficials can be reduced by chlorpyrifos applications, seasonal abundance of certain beneficials may not be significantly affected (Braman and Pendley 1993). The goal of this study, begun one year prior to initiation of the PBWEP, was to evaluate the impact of repeated applications of chlorpyrifos, similar to those possible for an eradication program, on beneficial and secondary pest populations in southern New Mexico cotton.

MATERIALS AND METHODS

This study was conducted during 2001 and 2002 at the Leyendecker Plant Science Research Center (LPSRC) near Las Cruces, New Mexico. Acala cotton (1517-99) was planted on 40-in bed spacings, furrow irrigated, and grown using local agronomic practices. Each plot was 10 rows by ≈7.6 m (0.01 ha). Four untreated buffer rows and six-foot alleys separated adjacent plots.

The experimental design was a randomized complete block with four replications. Treatments, in addition to an untreated check (UNT), consisted of chlorpyrifos applied at 1,120g ai/ha during four periods of the growing season: season-long (ALL), early season only (EAP), mid-season only (MAP), and late-season only (LAP) applications. Except for season-long, all other treatment periods consisted of three sequential chlorpyrifos applications made at ≈10-day intervals. Chlorpyrifos was applied by a commercial agricultural ground rig through a 10-row boom with three nozzles per row. Sampling was initiated at squaring and continued until defoliation. Data for seasonal arthropod abundances were analyzed using a split plot in time model using an ANOVA and means separated with Duncan's Multiple Range Test, with $\alpha = 0.05$ (SAS Institute 1999).

In 2001, EAP was sprayed on 20 and 29 June, and 9 July; MAP on 2, 10, and 18 August; and LAP on 28 August, and 9 and 17 September. Nine chlorpyrifos applications were made to the ALL treatment starting on 20 June at ≈10-day intervals, with the last application on 17 September.

In 2002, EAP was sprayed on 4, 16, and 31 July, and MAP on 10, 18, and 28 August. On 14 and 21 September, all cotton at the LPSRC was sprayed with leverage (cyfluthrin and imidacloprid) to control building pink bollworm and whitefly populations. Therefore, treatments LAP, ALL, and UNT were discontinued at that time.

Cotton aphid populations were evaluated by counting the number of aphids on each of 20 randomly selected upper and lower canopy leaves. Aphid evaluations were initiated prior to first bloom and continued until plant senescence.

Pink bollworm (PBW) moth populations were monitored throughout the season using delta traps baited with gossyplure (Trécé® pherocon). Pink bollworm damage was evaluated at the end of the season by cutting 20 randomly collected bolls from each plot during two weeks in 2001 and 2002 (20 and 27 September and 12 and 30 September, respectively).

Other arthropod populations, including beneficials, were evaluated weekly with a vacuum sampler placed over 10 plants (≈4 sec. each) of one randomly selected row in each plot. Each sample was bagged, taken to the laboratory, and frozen until it could be counted.

RESULTS

Aphids were not detected until mid-August in 2001, and no treatment reached an economic level this season. Populations peaked at 4.2 per leaf in the EAP treatment. The ALL plots averaged 1.7 aphids per leaf at the end of the season, significantly lower than the UNT and the EAP treatments at 3.2 and 4.2 respectively (Fig. 1A). Aphid herbivory did not result in noticeable accumulation (by visual observations) of honeydew in any plots.

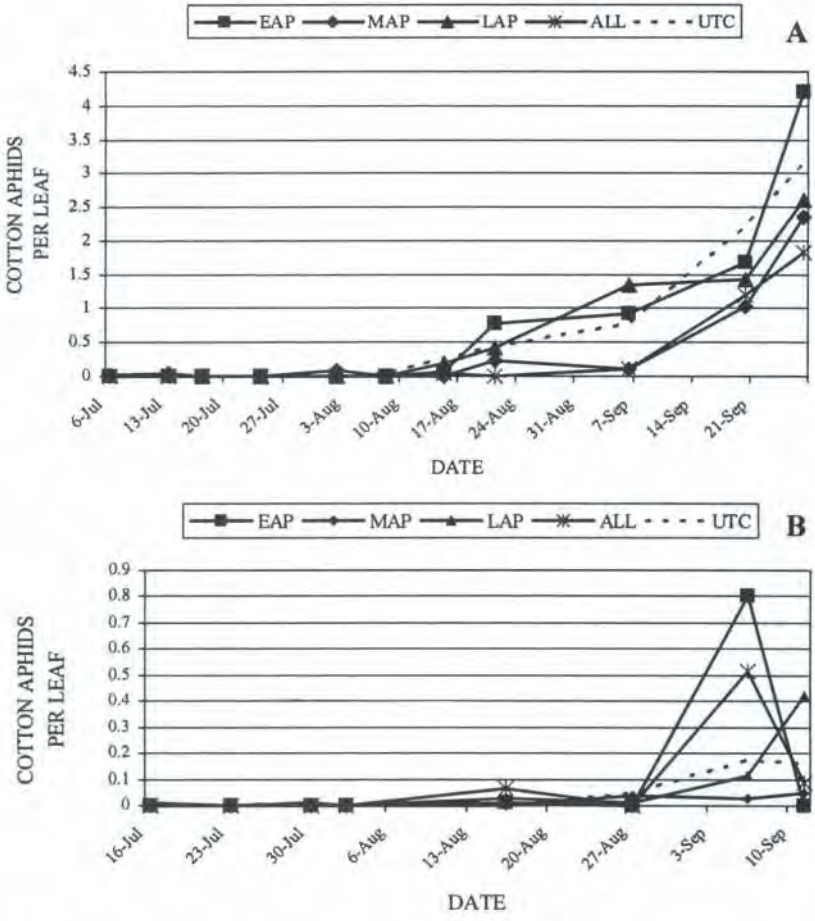

FIG. 1. Seasonal mean number of aphids per leaf at Leyendecker Plant Science Research Center. A) 2001. B) 2002. EAP = early-season only; MAP = mid-season only; LAP = late=season only; ALL = season-long; UNT = untreated check.

No significant differences in PBW-infested green bolls were observed among chlorpyrifos treatments at the end of the 2001 season (Fig. 2). The percentage of damaged bolls ranged from 27.5% in the ALL to 38.8% in the UNT treatments for the 20 September evaluation. Seven days later, percent PBW-damaged bolls increased in all plots, ranging from 35.0% in the MAP to 56.3% in the LAP treatments.

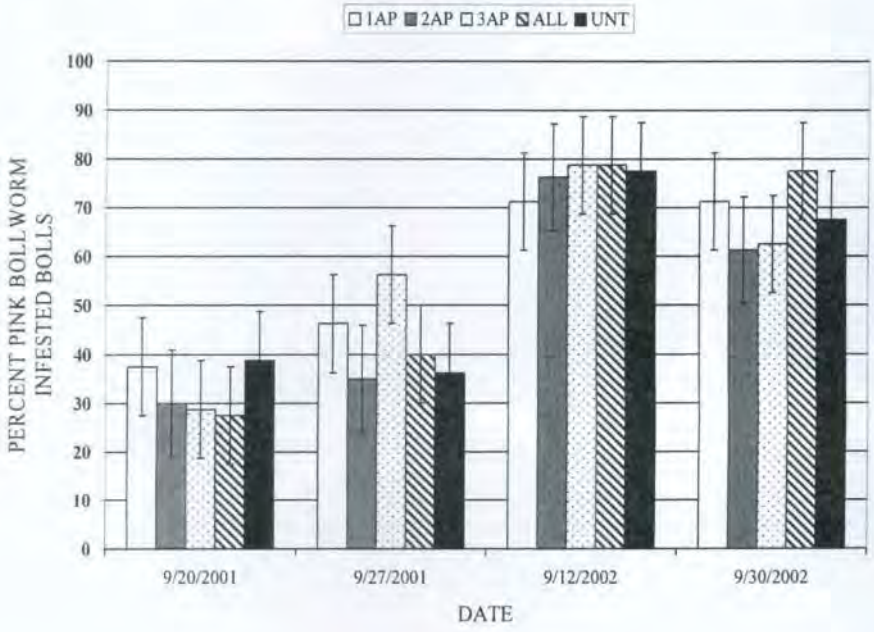

FIG. 2. Percentage of pink bollworm-infested bolls at Leyendecker Plant Science Research Center for 2001 and 2002. EAP = early-season only; MAP = mid-season only; LAP = late=season only; ALL = season-long.

Predator populations remained consistent in cotton for 2001. Heteropterans were the most numerous predators in this system. *Orius* spp. was seasonally most abundant (42.3%); followed by *Nabis* spp., primarily *N. alternatus* Parshley (33.1%); *Geocoris punctipes* Say and *G. pallens* Stål (12.7%); *Deraeocoris* (4.9%); reduviids, primarily *Zelus* sp. and *Sinea* sp., (3.8%); and miscellaneous predatory bugs (3.2%). As a group, predaceous Heteroptera were significantly less abundant in the ALL and EAP treatments of chlorpyrifos than the MAP and LAP, but not significantly different from the UNT (Table 1). Individually, only *Geocoris* spp. was found to be significantly less abundant in the ALL than the UNT; this also was true of spiders. Lady beetle populations were significantly lower in the EAP and MAP treatments than the other treatments. Lacewings (99% of which were chrysopids) were not significantly different among any treatment.

Phytophagous plant bugs were seasonally the most common pest group in cotton. *Lygus*, primarily *L. hesperus* Knight and *L. elisus* Van Duzee, composed only a small portion of these numbers (12.0%); most were other mirids (88.0%), including whitemarked fleahoppers, *Spanagonicus albofasciatus* (Reuter); western plant bug,

Rhinocloa forticornis Reuter; and cotton fleahoppers, *Pseudatomoscelis seriatus* (Reuter). No significant differences among treatments were observed for these insects.

TABLE 1. Comparative Abundance of Arthropods Among Chlorpyrifos Treatments at New Mexico's Leyendecker Plant Science Research Center for 2001 and 2002.[a]

	Chlorpyrifos Treatments[b]				
Arthropod	EAP	MAP	LAP	ALL	UNT
		2001			
Geocoris	0.55bc	0.87ab	0.69abc	0.45c	1.04a
Nabis	1.82a	1.98a	2.04a	1.66a	2.09a
Orius	2.14b	2.46b	3.29a	2.07b	2.29b
Heteroptera (total)[c]	5.04b	5.89ab	6.38a	4.66b	5.82ab
Lacewings	0.73ab	0.82a	0.47ab	0.59ab	0.45b
Coccinellids	0.27b	0.32b	0.65a	0.39ab	0.45ab
Spiders	0.98a	1.16a	1.40a	0.48b	1.37a
Plant bugs	2.39b	3.05ab	3.40a	2.43b	3.30ab
		2002			
Geocoris	0.23a	0.25a	----	----	0.10a
Nabis	0.17a	0.40a	----	----	0.29a
Orius	3.71a	3.83a	----	----	3.36a
Heteroptera (total)[c]	4.56a	4.71a	----	----	4.15a
Lacewings	0.21a	0.25a	----	----	0.23a
Coccinellids	0.00a	0.02a	----	----	0.00a
Spiders	0.33a	0.25a	----	----	0.33a
Plant bugs	2.40a	2.71a	----	----	2.79a

[a] Means within rows followed by the same letter are not significantly different ($P \leq 0.05$; Duncan's Multiple Range Test).
[b] EAP = early-season only; MAP = mid-season only; LAP = late-season only; ALL = season-long; UNT = untreated check.
[c] Predatory Heteroptera as a group.

Aphid populations remained low during the 2002 season, peaking at less than one per leaf (Fig. 1B) for all treatments. Since sampling was terminated early, aphid populations could not be effectively assessed late in the season. As in the previous year, there was no significant accumulation of honeydew.

PBW populations were extremely high late season 2002 in all plots (Fig. 2), and no significant differences were observed among treatments for either date. The percentages of damaged bolls on 12 September ranged from 71.25% in the EAP to 78.7% in the LAP and ALL treatments, and from 61.25% in the MAP to 77.5% in the ALL treatments on 30 September.

Heteropteran bugs were the most numerous predators during 2002. *Orius* was seasonally most abundant (84.3 %), followed by *Nabis* spp. (6.2 %), *Geocoris* spp. (4.2%), *Deraeocoris* (2.5%), reduviids (2.4%), and miscellaneous predatory bugs (0.4%). After PBW, phytophagous plant bugs were seasonally the most common pests. Lygus was again a small portion (10%) of total plant bugs. As in the previous season, nabids were *N. alternatus*, big-eyed bugs were *G. punctipes* and *G. pallens*, reduviids were

primarily *Zelus* sp. and *Sinea* sp., and the most common mirids were *S. albofasciatus*, *R. forticornis*, and *P. seriatus*. There were no significant differences among treatments for any arthropod sampled this season.

DISCUSSION

Chlorpyrifos treatments were initiated to study possible effects of the insecticide on non-target pests and beneficials during distinct periods of the growing season; therefore, timing of application did not necessarily correspond to ambient flights of PBW. This explains why PBW populations were not significantly more abundant in the UTC than the pesticide treatments for our observations.

Aphid populations found during both years of this study were typical of southern New Mexico cotton. Aphid populations reached low to moderate levels, and no treatment reached an economic population. In 2001, season-long applications of chlorpyrifos did not result in an increase in aphid populations above that of the untreated check. No chlorpyrifos applications appeared to affect the seasonal abundance of the majority of predaceous Heteroptera, lacewings, or phytophagous plant bugs. It is important to note that the "phytophagous" plant bugs discussed here also are facultatively predaceous, particularly the whitemarked fleahopper (Coll 1998).

Only populations of *Geocoris* and spiders were significantly less in the season-long chlorpyrifos-treated plots than the control. The significantly lower numbers of lady beetles in the EAP and MAP treatments is likely credited to low aphid populations that did not begin to build until just prior to the beginning of the LAP treatment. No chlorpyrifos treatment differences were detected for any arthropod population (EAP and MAP only) in 2002.

Possible reasons for chlorpyrifos' failure to affect the populations of the majority of beneficial arthropods examined in this study include tolerance to the insecticide and continued reinfestation from surrounding fields. Agriculture in southern New Mexico includes significant alfalfa acreage interspersed among small fields of a wide variety of crops. Alfalfa is a well-known harborage for beneficials that readily move into surrounding fields when hay is cut. In New Mexico, alfalfa supports two dozen predators and ≈120 species of parasitoids (J. J. Ellington, unpublished data), and at least three alfalfa fields were within 400 meters of this study each year. Immigration of beneficials (and plant bugs) could affect seasonal abundance of these insects in cotton following chlorpyrifos applications. Spiders generally do not move over long distances as readily as most other beneficials evaluated during this study, possibly explaining the seasonal reduction in their numbers in the season-long treated plots compared to the check.

Results indicate that repeated use of chlorpyrifos during typical light aphid years in southern New Mexico should not result in a population increase of cotton aphids above that of an untreated field. Findings indicate that chlorpyrifos may still be somewhat efficacious on cotton aphids in southern New Mexico and/or the impact on many of the principal beneficials is minimal with respect to aphid herbivory. More research is needed to determine if the reduction in *Geocoris* spp. and spider populations occurred as a result of decreased levels of immigration relative to other arthropods, or due to a lack of tolerance to chlorpyrifos.

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SURVIVAL, DEVELOPMENT, AND GROWTH OF *COCCINELLA SEPTEMPUNCTATA*[1] FED *SCHIZAPHIS GRAMINUM*[2] FROM RESISTANT AND SUSCEPTIBLE WINTER WHEAT

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ABSTRACT

Coccinella septempunctata L. (Coleoptera: Coccinellidae) larvae were supplied daily with 4 or 16mg of biotype-E *Schizaphis graminum* Rondani (Homoptera: Aphididae) reared on either resistant TAM-110 (TAM 105*4/Amigo*5//Largo), TAM-107 (TAM 105*4/Amigo), or the susceptible isoline TAM-105 winter wheat cultivars. No significant differences in survival ratios or developmental times were observed among *C. septempunctata* larvae supplied with increasing daily levels of greenbugs from each winter wheat cultivar. However, among cultivars small but significant differences in adult dry weight were observed. At the 16 mg daily prey level individuals supplied with greenbugs reared on TAM-105 were ≥2.5 mg heavier than those supplied with greenbugs reared on TAM-107 or TAM-110. Results from our study indicated that winter wheat cultivars with Amigo and Largo resistance genes would have little effect on the nutritional value of *S. graminum* for *C. septempunctata*.

INTRODUCTION

The greenbug, *Schizaphis graminum* (Rondani), is frequently the most important insect pest of wheat, *Triticum aestivum* L., in the U.S. (Burton et al. 1985, Pike and Schaffner 1985, Webster 1995). In the Southern Plains, injury from greenbug feeding can severely inhibit growth of winter wheat, resulting in reduced grain yields and net returns (Royer et al. 1998, Kindler et al. 2002, Elliott et al. 2004). In many areas where small grains are grown, cultivars resistant to greenbugs have been important components of profitable production (Curvetto and Webster 1998, Kindler et al. 2002). According to Porter et al. (1997), six genes conferring resistance to a range of greenbug biotypes have been identified in small grains. Of those resistance genes, *Gb2* (Amigo) and *Gb3* (Largo) are present in the genetic makeup of wheat cultivars widely grown in the Southern Plains. The cultivar TAM-107 with *Gb2* (TAM 105*4/Amigo) confers resistance to greenbug biotypes B and C, whereas TAM-110 with *Gb3* (TAM 105*4/Amigo*5//Largo) confers resistance to biotypes C and E (Porter et al. 1997, Lazar et al. 1998). The indirect effects of crop plants on entomophagous insects have been well documented in tritrophic

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interaction studies. Biological control can be significantly enhanced or reduced when host plants alter prey abundance, influence detection or access to prey, or alter the suitability of prey (Kareiva and Sahakian 1990; van Emden and Wratten 1990; Bottrell et al. 1998; Biswas and Singh 1998; Giles et al. 2002 a,b). Understanding the interactions among resistant crops, insect prey, and natural enemies is essential for the development of sustainable pest management approaches (Bergman and Tingey 1979, van Emden and Wratten 1990).

Several researchers have investigated the effects of greenbug-resistant wheat, sorghum, barley, and oat cultivars on Hymenopteran parasitoids and Coccinellidae predators (Starks et al. 1972, Schuster and Starks 1975, Salto et al. 1983, Rice and Wilde 1989, Campbell et al. 1990, Fuentes-Granados et al. 2001). Results of these studies suggest that resistant cultivars can enhance or reduce the suitability of greenbugs for parasitoids and Coccinellidae, and subsequently enhance or reduce biological control efforts. These previous studies provide valuable information for comparing tritrophic interactions among several greenbug-resistant and susceptible cultivars; however, none have quantitatively isolated the indirect effects of greenbug resistance genes in wheat on Coccinellidae development, survival, and growth.

The objective of this study was to determine the suitability of *S. graminum* (biotype-E), reared on TAM-110, TAM-107, or the susceptible isoline TAM-105, for an important greenbug predator *Coccinella septempunctata* L. To quantitatively assess whether greenbug resistance genes in wheat alter the suitability of greenbugs for *C. septempunctata*, we evaluated two limiting daily prey levels (4 and 16mg) reared on each cultivar. By supplying immature *C. septempunctata* with a range of sub-optimal daily levels of *S. graminum*, and therefore controlling daily consumption, we were able to determine the effects of greenbug resistance genes in wheat on the suitability of greenbugs for *C. septempunctata*.

MATERIALS AND METHODS

Colonies of *S. graminum* (biotype-E) were established in an environmental growth chamber on winter wheat cultivars TAM-105, TAM-107, or TAM-110 and maintained at 22°C with a 16:8 (L:D) photoperiod. To isolate greenbug colonies, wheat was grown in 15-cm-diameter pots covered with a vented (nylon mesh) plexiglass cylinder (33-cm tall). Plants were infested with greenbugs after reaching a height of approximately 20cm. Greenbugs were collected from plants daily as a food source for Coccinellidae feeding studies; plants in colonies were disposed of prior to severe injury (significant necrosis) from greenbug feeding.

Adult *C. septempunctata* were collected from north-central Oklahoma alfalfa fields. Twelve mating pairs of *C. septempunctata* were maintained in half-pint (236.6ml) cardboard containers covered with nylon mesh in an environmental chamber at 24°C and a 16:8 (L:D) photoperiod. Mating pairs were provided each day with an unlimited supply of *Acyrthosiphon pisum* Harris reared on 'Windsor' faba beans (*Vicia faba* L.), a cotton ball moistened with water, and a wheat-yeast-honey mixture.

Eggs from *C. septempunctata* mating pairs were collected each day, placed in 5ml glass vials stopped with cotton and maintained in an environmental chamber at 24°C and a photoperiod of 16:8 (L:D) h. Upon eclosion, larvae were placed individually in vials stopped with cotton and supplied each day with either 4 or 16mg of live apterous greenbugs from one of the three previously mentioned winter wheat cultivars. A digital microbalance was used to carefully weigh live greenbugs from each cultivar colony.

Daily prey levels were selected to represent a range of sub-optimal levels of aphids likely to be consumed in agricultural systems (Obrycki et al. 1998). While both daily prey

levels (4 and 16mg) represented an unlimited supply of greenbugs for first and second instars (98% survival and similar developmental times), these quantities were a limiting supply of greenbugs for third and fourth instars. The upper level of 16mg was chosen as the highest daily prey level because it represents an adequate diet for near maximal developmental rates, but is below the maximum daily consumption capabilities of third and fourth instar *C. septempunctata* (Michels and Behle 1991; Michels and Flanders 1992; K. L. Giles unpublished data). Because all greenbugs supplied daily were consumed when *C. septempunctata* individuals were in the third or fourth stadium, this upper daily prey level significantly reduced the confounding effects of preference and satiation and allowed the effect of daily prey levels among cultivars to be compared quantitatively.

Individuals were selected at random from among the twelve parental lines of *C. septempunctata*, and 34 to 43 were assigned to each daily prey level / cultivar treatment. During the period of development, individual larvae were systematically checked each day to record mortality, molting, pupation, and adult emergence. One day after emergence, adults were dried in a laboratory oven for 3 days at 60°C, then weighed using a digital microbalance.

All analyses were performed using SAS version 6.12 for windows (SAS Institute 1996). A 0.05 significance level was used for all statistical analyses. Alive to dead ratios were compared among treatments using χ^2 analysis (PROC FREQ) followed by Fisher's exact test. Ratios (alive to dead) for fourth-instar survival, larval survival, and preimaginal survival were compared among cultivars and daily prey levels (factorial analyses) using χ^2 analysis (PROC CATMOD). Developmental times (days) and adult dry weights (g) were compared among cultivars and daily prey levels (factorial analyses) by analysis of variance (PROC MIXED), followed by LSMEANS comparisons among cultivars at each daily prey level (SLICE option) when the fixed effect of cultivar was significant. The Mixed Procedure was used because it does not assume equal variances among treatments and because it supplies ANOVA with both random and fixed effects. Parental line may be a source of experimental error and was included as a random factor, whereas cultivar and daily prey level were included as fixed factors.

Voucher specimens (*C. septempunctata* adults) were deposited at the Department of Entomology and Plant Pathology Museum at Oklahoma State University, Stillwater.

RESULTS

Before the fourth stadium, survival of *C. septempunctata* was not affected by daily greenbug level; however, survival of fourth instars, larval survival, and preimaginal survival ratios increased significantly with increasing daily levels of greenbugs (Table 1, 2). Despite an observed higher survival of developing *C. septempunctata* supplied with 4 mg/day of greenbugs reared on TAM-110, the cultivar from which greenbugs were reared did not significantly affect survivorship (Table 1, 2).

Third instar, fourth instar, larval and preimaginal developmental times for *C. septempunctata* declined significantly with increasing daily levels of greenbugs (Table 3, 4). However, the cultivar from which greenbugs were reared did not significantly affect developmental times of *C. septempunctata* at any stage (Tables 3 and 4).Adult dry weight for *C. septempunctata* increased significantly with increasing daily levels of greenbugs (Tables 3 and 4).

Additionally, the cultivar from which greenbugs were reared had a significant influence on adult weights at the 16mg daily greenbug level (Table 3, 4). At the 16 mg daily greenbug level, average adult weights were highest for *C. septempunctata* supplied with greenbugs reared on TAM-105 (Table 4).

TABLE 1. ANOVA Results (CATMOD Procedure, SAS) for Survival Ratios of *C. septempunctata* Supplied with Increasing Daily Prey Levels of *S. graminum* (Biotype E) from Susceptible and Resistant Winter Wheat Cultivars[a].

Response Variable	Source of Variation	df	χ^2	*P*
Survival				
Fourth Instar	Cultivar	2	1.7	0.423
	Prey Level	1	10.2	0.001
	Cultivar x Prey Level	2	2.8	0.247
Larval	Cultivar	2	1.6	0.453
	Prey level	1	11.2	<0.001
	Cultivar x Prey Level	2	1.5	0.466
Preimaginal	Cultivar	2	1.3	0.533
	Prey level	1	15.4	<0.001
	Cultivar x Prey Level	2	3.8	0.148

[a]Cultivars were TAM-105, TAM-107, and TAM-110; daily prey levels were 4 or 16 mg/day of *S. graminum*

TABLE 2. Survival Ratios at 24°C for *C. septempunctata* Supplied with Increasing Daily Prey Levels of *S. graminum* (Biotype E) from Susceptible (TAM-105 and TAM-107) and Resistant (TAM-110) Winter Wheat Cultivars.

		Proportion Surviving			
S. graminum (mg / day)	Cultivar	3rd Instar	4th Instar	Larval	Preimaginal
4	TAM-105	1.000	0.600	0.600	0.543
	TAM-107	0.971	0.606	0.588	0.441
	TAM-110	0.943	0.818	0.771	0.714
16	TAM-105	0.971	0.853	0.829	0.800
	TAM-107	0.977	0.905	0.884	0.860
	TAM-110	1.000	0.861	0.861	0.806

DISCUSSION

Integration of resistant wheat cultivars and biological control as components of sustainable pest management approaches requires detailed knowledge of the effects of resistant cultivars at the second and third trophic levels. Several researchers have documented that wheat cultivars containing the Amigo or Largo resistance genes can significantly influence biotype-E greenbug reproductive parameters, (Campbell et al. 1990, Lazar et al. 1995, and Michels et al. 1997); however, tritrophic interactions among resistant wheat cultivars, biotype-E greenbugs, and natural enemies have not been studied adequately. Campbell et al. (1990) and Fuentes-Granados et al. (2001) reported that

resistant wheat cultivars (TAM-107 and TAM-110) have little influence on parasitism of greenbugs by *Lysiphlebus testaceipes* Cresson. In fact, Campbell et al. (1990) suggested that resistant wheat and *L. testaceipes* worked synergistically to reduce greenbug populations. Similarly, the results from our study suggested that resistance genes (*Gb2* and *Gb3*) that are incorporated into winter wheat cultivars (TAM-107 and TAM-110) had little effect on the suitability of greenbugs for the survival and development of *C. septempunctata*.

TABLE 3. ANOVA Results (Mixed Procedure, SAS) for Developmental Times (Days) and Adult Dry Weight (g) of *C. septempunctata* Supplied with Increasing Daily Levels of *S. graminum* (Biotype E) from Susceptible and Resistant Winter Wheat Cultivars[a].

	Tests of Fixed Effects			
Response Variable	Source of Variation	df	*F*	*P*
Developmental Time				
Third Instar	Cultivar	2, 196	0.37	0.694
	Prey level	1, 196	55.92	<0.001
	Cultivar × Prey level	2, 196	0.01	0.986
Fourth Instar	Cultivar	2, 149	0.28	0.755
	Prey level	1, 149	257.27	<0.001
	Cultivar × Prey level	2, 149	0.29	0.749
Larval	Cultivar	2, 149	0.35	0.702
	Prey level	1, 149	290.24	<0.001
	Cultivar × Prey level	2, 149	0.19	0.827
Pupal	Cultivar	2, 136	0.36	0.700
	Prey level	1, 136	0.09	0.764
	Cultivar × Prey level	2, 136	0.59	0.556
Preimaginal	Cultivar	2, 136	0.87	0.422
	Prey level	1, 136	230.91	<0.001
	Cultivar × Prey level	2, 136	0.54	0.585
Adult Dry Weight (g)	Cultivar	2, 133	6.73	0.002
	Prey level	1, 133	298.60	<0.001
	Cultivar × Prey level	2, 133	1.62	0.202

[a]Cultivars were TAM-105, TAM-107, and TAM-110; Daily prey levels were 4 or 16 mg/day of *S. graminum*.

However, adult dry weights were lower for *C. septempunctata* supplied with greenbugs from TAM-107 and TAM-110. These observed differences in average adult dry weights could potentially influence the numerical response of *C. septempunctata*. Indeed, as demonstrated by Sundby (1968), smaller *C. septempunctata* females tended to be less fecund. Additional studies examining the influence of resistant winter wheat cultivars on the suitability of greenbugs for *C. septempunctata* reproduction are clearly needed.

Based on our results, it appears that TAM-107 and TAM-110 had little negative effect on *C. septempunctata*. Future field studies are needed, and should be designed to

TABLE 4. Average (LSMeans ± SE) Developmental Times (days) at 24°C and Adult Dry Weight (mg) of *C. septempunctata* Supplied with Increasing Daily Prey Levels of *S. graminum* (Biotype E) from Susceptible (TAM-105 and TAM-107) and Resistant (TAM-110) Winter Wheat Cultivars.

		Development Time (days)					
S. graminum (mg / day)	Cultivar	3rd Instar	4th Instar	Larval	Pupal	Preimaginal	Adult Dry Weight (mg)
4	TAM-105	3.0 ± 0.2	8.4 ± 0.4	15.8 ± 0.5	5.5 ± 0.2	24.4 ± 0.5	15.2 ± 0.8
	TAM-107	3.1 ± 0.2	8.1 ± 0.4	15.4 ± 0.5	5.5 ± 0.2	23.9 ± 0.6	14.5 ± 0.9
	TAM-110	2.9 ± 0.2	8.2 ± 0.3	15.5 ± 0.5	5.6 ± 0.1	24.1 ± 0.5	14.0 ± 0.7
16	TAM-105	2.1 ± 0.2	4.6 ± 0.3	11.2 ± 0.5	5.6 ± 0.1	20.1 ± 0.5	25.8 ± 0.7 a
	TAM-107	2.1 ± 0.2	4.6 ± 0.3	11.2 ± 0.4	5.6 ± 0.1	20.1 ± 0.5	23.3 ± 0.6 b
	TAM-110	2.0 ± 0.2	4.8 ± 0.3	11.1 ± 0.5	5.3 ± 0.1	19.6 ± 0.5	22.5 ± 0.7 b

Means followed by different letters in a column are significantly different ($P = 0.05$).

investigate the potential interactive effect between plant resistance and predation by Coccinellidae for population suppression of *S. graminum*.

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COMPOSITION OF *LYGUS*[1] SPECIES FOUND IN SELECTED AGRONOMIC CROPS AND WEEDS IN THE SAN JOAQUIN VALLEY, CALIFORNIA

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ABSTRACT

The species composition of lygus bugs in seed and forage alfalfa, *Medicago sativa* L.; cotton, *Gossypium hirsutum* (L.); cowpeas, *Vigna unguiculata* (L.) Walp.; and overwintering weed species was determined during 2002-2003. *Lygus hesperus* (Knight) and *L. elisus* (van Duzee) were the only species recovered. Alfalfa supported the highest number of *Lygus* spp. Overall, *L. hesperus* was the predominant species recovered from all crops. The highest proportion of *L. elisus* was found in cowpeas. Among the weeds sampled, *L. elisus* appeared to favor shortpod mustard, *B. geniculata* (Desf.) J. Ball., and Kellogg's tarweed, *Hemizonia Kelloggii* Green. There was a 1:1 male:female sex ratio in all crops except cotton where females predominated in 2002, but not in 2003.

INTRODUCTION

There are approximately 43 species of *Lygus* worldwide, of which over 30 Neartic species are known to occur in North America (Kelton 1975, Schwartz and Foottit 1998). In most agricultural settings, however, the number of species encountered is relatively few. Armstrong and Camelo (2003b) found *L. hesperus* (Knight), *L. elisus* (van Duzee) and *L. lineolaris* (Palisot de Beauvois) to be the most common species associated with cotton, *Gossypium hirsutum* (L.); alfalfa, *Medicago sativa* L.; and weeds in the Texas High Plains. In Canada, Carcamo et al. (2002), found a "northern" assemblage dominated by *L. lineolaris*, with a minor representation by *L. borealis* (Kelton), *L. elisus* and *L. keltoni* (Schwartz); the "southern" assemblage (grassland) was dominated by *L. keltoni, L. elisus,* and *L. borealis* with a few *L. lineolaris*. In Oklahoma, *L. lineolaris* was the most abundant species recovered (Karner et al. 2001). In California, *L. hesperus* is the most common lygus bug associated with cotton (Leigh et al. 1988).

Three species, *L. hesperus*, *L. elisus* and *L. lineolaris*, are believed to be associated with several agronomic crops in the San Joaquin Valley (Mueller et al. 2003). These crops are generally grown adjacent to another agronomic crop, forage alfalfa, which is a preferred

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host for *Lygus* spp. although it is not damaged by lygus bug feeding and control is unnecessary (Stern 1969, van den Bosch and Stern 1969). Following the alfalfa harvest, however, lygus bugs are driven out of the forage alfalfa and move into adjacent crops of cotton, seed alfalfa and cowpeas, *Vigna unguiculata* (L.) Walp., where they can cause considerable damage (van den Bosch and Stern 1969). These three crops are grown extensively in the San Joaquin Valley and many of the insecticide applications made on these crops are for lygus bug control.

Lygus spp. are generally grouped together and referred to collectively as "lygus bugs." However, there are some differences between the species relative to their behavior, impact on host plants, reaction to pheromones, and response to biological control agents. Mueller and Stern (1973) found differences in the selection of oviposition sites in safflower, *Carthamus tinctorius* L., between *L. hesperus* and *L. elisus* as well as differences in female longevity. Hills (1943) determined that *L. hesperus* and *L. oblineatus* Knight (now called *L. lineolaris*) caused more damage to sugarbeets grown for seed than did *L. elisus*. Armstrong and Camelo (2003a) found that *L. elisus* caused similar or greater damage to cotton anthers compared to *L. hesperus* and caused significantly more damage to first position 11th-node squares in laboratory studies than did *L. hesperus*. Graham (1987) and Ridgway and Inscoe (1996) noted that there is cross attraction of males by females between *L. hesperus* and *L. elisus*; however, *L. hesperus* males are attracted only by conspecific females. DeBolt (1989) found that *Leiophron uniformis* (Gahan) reared on *L. lineolaris* accepted that host 2.5 times as often as did wasps reared on *L. hesperus*. Knowing the species composition in various crops may lead to an improved interpretation of research results and enhance research findings in areas of biological control, assessment of economic thresholds, monitoring, management strategies, and insecticide resistance.

Previously, *Lygus* spp. keys were very detailed and complicated to use since they were written to identify all known species. We recently developed a simplified field key that can be used to easily separate the three major species associated with agronomic crops in the San Joaquin Valley (Mueller et al. 2003). This paper reports the results of a lygus survey conducted to determine the relative proportion and abundance of the common *Lygus* spp. found in alfalfa (seed and forage), cotton, cowpea, and overwintering weed hosts in the San Joaquin Valley.

MATERIALS AND METHODS

Three sample sites in Madera, Fresno, and Tulare counties were identified. Using the most recent cropping inventory data from the State of California Department of Water Resources (2000), crop abundance and diversity were summarized within the township in which the sample sites were located using ArcGIS I (Murad 2003). Three distinct crops (cotton, seed alfalfa, and cowpeas), which are all highly susceptible to *Lygus* spp. injury, were sampled on a weekly basis. We also evaluated *Lygus* spp. in forage alfalfa, a preferred host, but one not susceptible to damage. Each sample site had the commonality of being grown near forage alfalfa, which is a frequent source of lygus bugs, but other adjacent crops characterized them as distance landscapes (Fig. 1).

In April and May of 2002 and 2003, lygus bugs were also collected on weeds from foothill areas of the Coast Range in Merced, Madera, Fresno, Kings, Tulare, and Kern counties by taking fifty 180° sweeps from each weed species using a standard 15-inch sweep net. Weed species sampled in each year depended on their prevalence and density. During 2002, weed species sampled included black mustard, *Brassica nigra* (L.) Koch.; Kellogg's tarweed, *Hemizonia Kelloggii* Green; and yarrow (*Achillea* spp.). In 2003, black mustard, tarweed, shortpod mustard, *B. geniculata* (Desf.) J. Ball.; and London Rocket, *Sisymbrium*

Irio L., were sampled. Common and scientific names of weed species are according to Munz (1959).

In the agronomic crops, the study was conducted during the spring and summer of 2002 and 2003. Sampling began in early April and ended in mid-September. Adult lygus bugs were collected by taking one hundred 180° sweeps every two to three weeks at each location and in each crop using a standard 15-inch sweep net. Lygus were aspirated from the net, placed in plastic vials and returned to the laboratory where they were placed in petri dishes and frozen (-80 °F) until they could be evaluated. The lygus bugs were separated by sex (Mueller et al. 2003) and the males identified to species. The key developed by Mueller et al. (2003) was used for all identifications. Only males were identified to species since the species identification of females is unreliable (Mueller et al. 2003). Specimens were routinely compared to museum material from the Bohart Museum of Entomology at the University of California, Davis. Voucher specimens were deposited in the Bohart Museum of Entomology at the University of California, Davis, Lot No. UCD204011. Sex ratios were evaluated by goodness of fit χ^2 (Zar 1974).

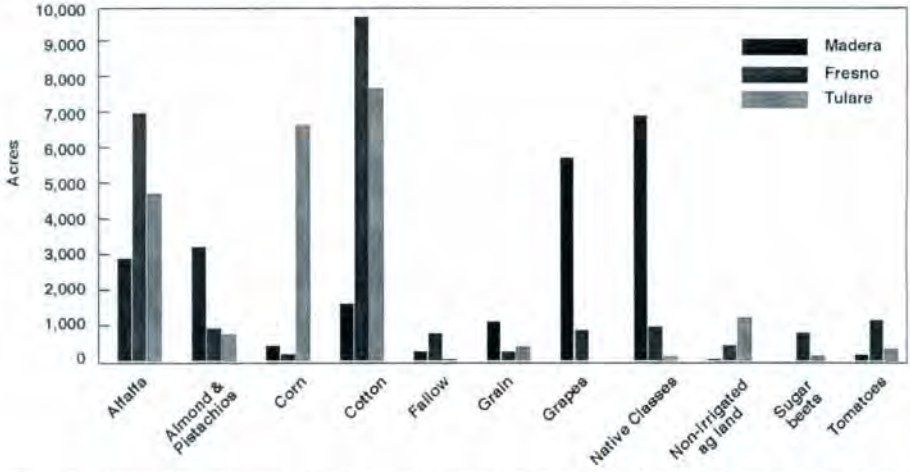

FIG. 1. Crop diversity and abundance in three townships in Madera, Fresno and Tulare counties, California.

RESULTS AND DISCUSSION

On a township scale (36 mi^2), the diversity of crops was similar between the sites, but the abundance varied (Fig. 1). The Madera County site was characterized by native classes (rangeland), grapes, (*Vitis vinifera* L.), nut crops, alfalfa, and cotton. The Fresno County location was characterized by cotton, alfalfa, both forage and seed, tomatoes, *Lycopersicon esculentum* Mill., and native classes. In Tulare County, the most abundant crops were cotton, corn, *Zea mays* L., alfalfa, and winter forage (*Triticale hexaploide* Lart.).

Only two lygus species, *L. hesperus* and *L. elisus*, were recovered during the two year study. No *L. lineolaris* were recovered from either crop or weed hosts. *Lygus hesperus* clearly dominated in forage and seed alfalfa in both 2002 and 2003 (Table 1). Slightly more *L. elisus* were found associated with forage alfalfa than with seed. Our results agree with those of Pickett (personal communication) who surveyed forage and seed alfalfa in Kern, Fresno, Yolo, and Sacramento counties and found that *L. hesperus* comprised the majority

of the adult *Lygus* spp. collected. In the Texas High Plains, Armstrong and Camelo (2003b) found a higher proportion of *L. hesperus* in forage alfalfa in 2000; however, *L. elisus* predominated in 2001. In the current study, alfalfa, both forage and seed, was clearly the preferred hosts for *Lygus* spp. compared to the other crops sampled. Collections from these two crops comprised 91 and 82% of the total *Lygus* spp. collected in 2002 and 2003, respectively. The number of lygus captured in cotton was higher than from cowpeas. *Lygus hesperus* was the predominant species recovered from cotton (Table 1), but the proportion of *L. elisus* was higher in this crop than in either alfalfa forage or seed. Armstrong and Camelo (2003b) also found a higher proportion of *L. elisus* in cotton compared to forage alfalfa. The proportion of *L. elisus* recovered from dry beans was considerably higher than that recovered from any of the other crops (Table 1).

TABLE 1. Percentage *L. hesperus* and *L. elisus* Males Recovered from Targeted Agronomic Crops in Madera, Fresno, and Tulare Counties and from Selected Weed Species on the West Side of the San Joaquin Valley in 2002 and 2003.

Crop	n [a]	2002		n [a]	2003	
		L. hesperus	*L. elisus*		*L. hesperus*	*L. elisus*
Alfalfa seed	309	97.4	2.6	201	100.0	0.0
Alfalfa hay	251	96.4	3.6	315	98.7	1.3
Cotton	28	89.3	10.7	90	98.9	1.1
Cowpeas	13	61.5	38.5	35	62.9	37.1
Weeds	36	55.6	44.4	650	51.6	48.4
Yarrow	7	100.0	0.0	–	–	–
Tarweed	4	0.0	100.0	97	69.1	30.9
Black mustard	25	52.0	48.0	133	66.9	33.1
Shortpod mustard	–	–	–	344	39.5	60.5
London Rocket	–	–	–	76	48.7	51.3

[a] Number of male specimens recovered.

Many weed species in the western coastal range serve as overwintering hosts of *Lygus* spp. As the winter annuals dry in the spring, the lygus bugs leave, migrating to the San Joaquin Valley floor where they attack a variety of crops (Stern 1969, Goodell et al. 2000). While there were differences in *Lygus* species composition from various weed species (Table 1), it is difficult to draw conclusions due to differences in the availability of the various hosts for sampling (e.g., shortpod mustard was present in much higher densities in 2003 than the other species and thus more samples were taken from it than from black mustard or tarweed). There were also inconsistencies between years in the number of individuals recovered on the various weed species. Across all weed species, *L. hesperus* and *L. elisus* were recovered in a 1:1 ratio in both years ($\chi^2 = 0.22$ in 2002, $\chi^2 = 0.02$ in 2003). The equal proportion of *L. hesperus* to *L. elisus* collected on weeds during April and May did not translate into equal proportions of the two species found on the various crop plants early in the season. *L. hesperus* predominated from the beginning of sampling and *L. elisus* was not found in significant numbers until later June and July which coincided with the beginning of sampling in cowpeas.

Lygus spp. exhibited a 1:1 male:female ratio in alfalfa hay, alfalfa seed, and cowpeas (Table 2). In cotton the ratio was 1:1 in 2003; however, in 2002, the females outnumbered males by approximately 2 to 1. Zink and Rosenheim (2004) reported that male *Lygus* spp.

spend less time feeding on squares than do females, and that some cotton fields have *Lygus* spp. adult populations more heavily dominated by females than do other cotton fields. Although they did not report on male:female ratios, they did note that *Lygus* spp. sex ratio does not appear to be large enough to generate a large amount of variation in crop damage. Based on the approximate 1:2 male:female ratios we observed in 2002, this concept may need to be re-evaluated, and the sex ratio taken into account when determining economic threshold levels. When all crops were considered together, the ratio remained at 1:1 (Table 2).

TABLE 2. Percentage *Lygus* spp. Males and Females Recovered from Representative Agronomic Crops in 2002 and 2003.

Crop	n	2002		$\chi^{2\,a}$	n	2003		$\chi^{2\,a}$
		Male	Female			Male	Female	
Alfalfa seed	592	54.2	45.8	0.16	424	47.4	52.6	0.10
Alfalfa hay	511	51.5	48.5	0.06	649	48.5	51.5	0.06
Cotton	80	37.5	62.5	5.14	163	55.2	44.8	0.21
Cowpeas	31	45.2	54.8	0.24	71	49.3	50.7	0.03
All crops	1214	51.7	48.3	0.06	1307	49.0	51.0	0.04

[a] Critical value for χ^2 at $P = 0.05 = 3.84$.

There were no substantive differences in species composition across the three landscapes (Fig. 2). *Lygus hesperus* predominated in each location. *Lygus elisus* densities were slightly higher in the Tulare County landscape in 2002 and 2003. Of the three landscapes evaluated, the highest proportion of both *L. hesperus* and *L. elisus* recovered were from Fresno County (Fig. 3). The higher proportion of both species is likely due to the significantly higher acreage in Fresno County of all four crops evaluated (NASS-USDA 2002).

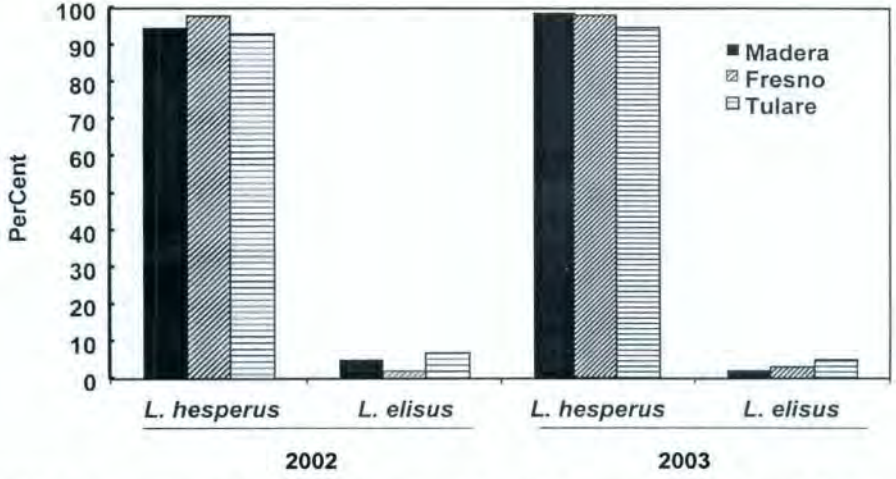

FIG. 2. Composition of male *Lygus* spp. across three landscapes in the San Joaquin Valley.

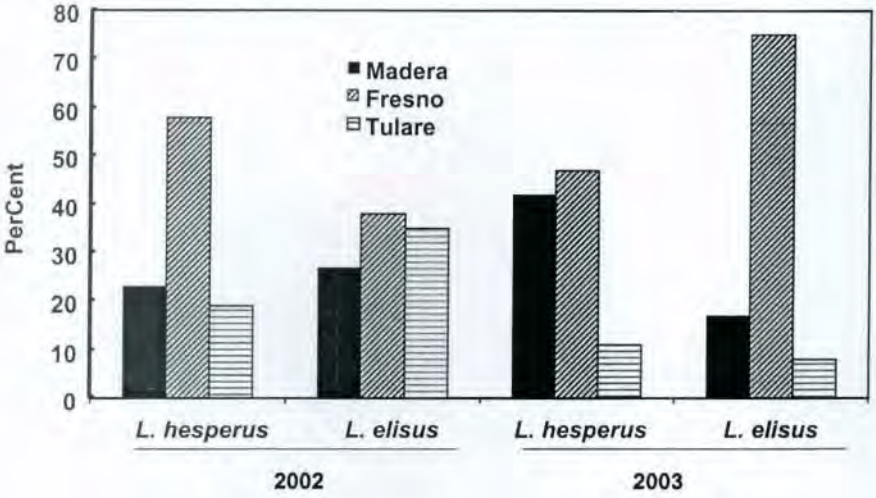

FIG 3. Proportion of male *L. hesperus* and *L. elisus* recovered from three landscapes in the San Joaquin Valley.

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POPULATIONS OF BANDEDWINGED WHITEFLIES (HOMOPTERA: ALEYRODIDAE) IN THE NORTHERN TEXAS ROLLING PLAINS

J. E. Slosser[1], M. N. Parajulee[2], G. B. Idol[1], and D. L. Hendrix[3]

ABSTRACT

During the summer of 2000, widespread infestations of whiteflies developed in dryland and irrigated cotton fields in the northern Texas Rolling Plains. The Texas Rolling Plains whitefly was identified as bandedwinged whitefly, *Trialeurodes abutiloneus* (Haldeman). This whitefly had spotted wings and cream-colored thorax and abdomen, rather than the typical dark band pattern on the wings and gray-colored thorax and abdomen. This whitefly was associated with 36 plant species, from 11 plant families, during the summer-fall period of 2000 and the spring-summer period of 2001. Laboratory studies indicated that this whitefly could transfer from at least eight species of weed hosts and reproduce successfully on cotton. Giant and western ragweeds (*Ambrosia trifida* L. and *A. psilostachya* DC, resectively) appeared to be the most important alternate hosts based on intensity of infestations and abundance of these two weeds. The primary sugars in bandedwinged whitefly honeydew were glucose and fructose, and contaminated cotton lint was slightly to moderately sticky. A significant sticky lint problem did not occur in a test where numbers of immature whteflies ≤ 33/leaf. Based on sampling data collected periodically from 1988 to 2002 in two locations in the northern Rolling Plains, high bandedwinged whitefly populations were associated with summers that had multiple days with temperatures >37.8°C.

INTRODUCTION

The bandedwinged whitefly (BWWF), *Trialeurodes abutiloneus* (Haldeman), has been present in low numbers in cotton in the northern Texas Rolling Plains since about 1988. If the BWWF was present prior to 1988, numbers were too low to attract attention. However, during the summer of 2000, heavy infestations occurred in both dryland and irrigated cotton fields, and heavy deposits of honeydew occurred on cotton leaves and on the soil surface under plants in a few fields. Infestations have been low since 2000. Boling and Schuster (1969) reported that BWWF populations were high in cotton in the Lower Rio Grande Valley, Texas, in late summer, 1965. Occasional high infestations of BWWF have been present in Louisiana since the mid-1940's, but the first damaging infestation in cotton did not occur until 1964. By the early-1970's, the BWWF had become a damaging pest in Louisiana (Clower and Watve 1973), southwest Arkansas, northeast Texas, in the Mississippi Valley, and Georgia (Jones et al. 1975).

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The BWWF has been reported as an occasional pest of cotton in localized areas in the San Joaquin Valley in California (Leigh et al. 1996). Butler and Muramota (1967) indicated that BWWF was considered a minor pest of cotton in Arizona, but Byrne et al. (1986) reported that population levels in some fields were high in the early 1980's.

Outbreaks of BWWF have been attributed to destruction of natural enemies with insecticides, development of resistance to insecticides, and changes in the agro-ecosystem (Clower and Watve 1973). Slosser et al. (1992) reported that BWWF numbers were significantly higher in cotton planted in late April and late May compared to numbers in cotton planted in late June, and BWWF numbers were negatively correlated with percentage leaf moisture. Planting pattern affects populations of BWWF, and numbers were significantly higher in cotton planted in every row (40" spacing) compared to numbers in cotton planted in skip-row patterns (plant two rows, skip two rows) (Parajulee et al. 2002).

The objectives of this report are to discuss (1) identification of the BWWF population in the Texas Rolling Plains, (2) alternate host plants in the Rolling Plains, (3) composition of the honeydew and implications for development of sticky cotton lint, and (4) influence of weather patterns on population density.

MATERIALS AND METHODS

Whitefly Sampling. Bandedwinged whitefly populations have been monitored periodically since 1988 in cotton at the Texas Agricultural Experiment Station located at Chillicothe in the northern Texas Rolling Plains. Cotton aphids, *Aphis gossypii* Glover, were sampled at Chillicothe each week during August-September from 1988-2004, and relative abundance of BWWF larvae was noted while inspecting the leaves for aphids. No samples were taken in years when BWWF were absent or were present in very low numbers. Samples were taken only when BWWF numbers appeared to be increasing. Samples for BWWF were taken at the Texas Agricultural Experiment Station at Munday from 1999-2002. Bandedwinged whitefly larvae were counted visually on five or ten leaves picked from the top-half of the plant in three or four replications of untreated cotton plots utilized in cotton aphid research. Whitefly larvae were counted once-a-week in each plot. Depending on year and location, plot sizes ranged from 8 rows wide by 15 m long to 44 rows wide by 183 m long.

Whitefly Identification. Larvae, pupae, and adults were collected on cotton leaves from a commercial cotton field in Wilbarger Co., Tex., during August, 2000. The whitefly samples were placed in a vial of 70% ethanol and sent to Dr. R. J. Gill (California Department of Food and Agriculture, Sacramento) for identification.

Honeydew Sugar Identification. A burlap bag was filled with cotton leaves contaminated with BWWF honeydew from the same commercial field as the whitefly specimens in Wilbarger Co., Tex. The leaves were placed in a cool chest and returned to the laboratory at Vernon. The leaves were then packaged in a box with ice and overnight mailed to the ARS, USDA, Western Cotton Insects Research Laboratory in Phoenix, AZ for determination of sugars present in the honeydew. The honeydew was extracted with hot, deionized water which was filtered and the water removed by lyophilization. The resulting solids were suspended in 80% ethanol, and the dissolved sugars were purified by means of activated charcoal (Hendrix 1993). Following removal of the ethanol, the purified sugars were suspended in deionized water and analyzed for carbohydrates by HPLC (Hendrix and Wei 1994).

Alternate Host Plants. Wild host plants and cultivated crops were surveyed for immature and adult BWWF in September - October 2000, and in March - June 2001. Immature and adult BWWF infestations on host plants were subjectively rated as low, moderate, and heavy. The survey was conducted in Knox and Haskell counties, located in the Rolling Plains Central Zone in the boll weevil, *Anthonomus grandis grandis* Boheman, eradication program and in Hardeman and Wilbarger counties, located within the Northern Rolling Plains Zone.

Leaves from potential host plants containing immature stages of BWWF were collected and brought to the laboratory, and the immatures were reared to adult emergence to verify that the life cycle was completed on that particular host. The rearing procedure consisted of floating the host-plant leaf, or a section of the leaf containing BWWF larvae, on distilled water in ventilated plastic petri dishes (60 x 15 mm). The petri dishes were kept at room temperature (about 24°C) and inspected daily for adult emergence. Successful emergence of adults in the petri dishes indicated the plant was a suitable host for BWWF, and the plants were identified using information provided by Diggs et al. (1999), Hatch et al. (1990), and USDA, ARS (2005). Adults reared from a weed host were transferred to a 104 x 26 mesh screen cage (BugDorm-3, MegaView Science Education Services Co., Ltd., Taichung, Taiwan) containing potted cotton plants (5-7 leaf stage) to determine if these whiteflies could be successfully reared on cotton. Cotton leaves were inspected 2-3 times per week for presence of larvae, pupae and adults. Emergence of BWWF adults in the cage and completion of one generation on the cotton leaves was evidence of successful host transfer from weeds to cotton.

Sticky Lint Analysis. A heavily infested field was selected in Knox Co., Tex. to determine degree of lint stickiness caused by BWWF honeydew. Bandedwinged whitefly larvae were counted visually on ten, 5th mainstem node leaves, and cotton aphids were counted visually on five leaves picked from the top-half of the plant and from five leaves picked from the bottom-half of the plants. Insect counts were taken at six locations in the field on 15 and 22 September.

Cotton was harvested immediately after the insect counts on 22 September to determine degree of sticky lint. Open cotton bolls were handpicked from 1 m of row at six locations in the field. The lint samples were deburred by hand and ginned on a laboratory, table-top gin. The lint was submitted to Cotton Incorporated, Cary, North Carolina (H2SD analysis, automated sticky detector) and to the ARS, USDA Cotton Quality Laboratory at Clemson, South Carolina (SCT analysis, manual sticky detector) to determine degree of lint stickiness.

Data Analysis. Pearson correlations were calculated to determine relationships between BWWF numbers and temperature and rainfall (Analytical Software 1998).

RESULTS AND DISCUSSION

Whitefly Identification. Upon close inspection under a binocular microscope, it became apparent that the adult whiteflies infesting the cotton fields were morphologically different from the BWWF pictured in the literature, which have wings with two dark wavy bands and gray-colored thorax and abdomen (Leigh et al. 1996). The BWWF from the Texas Rolling Plains has spotted wings and cream-colored thorax and abdomen (Fig. 1). However, the Rolling Plains BWWF meets the taxonomic criteria for *Trialeurodes abutiloneus* (Haldeman).

Alternate Host Plants. Immature stages of the BWWF were collected and successfully reared to adults from 27 plant species representing 10 plant families during the summer-fall period in 2000 (Table 1). Another nine species and one additional plant family were found in the spring-summer samples taken in 2001(Table 2). The BWWF was associated with 36 plant species from 11 plant families in this survey. The most common plant families included Asteraceae (12 species), Solanaceae (8 species), and Fabaceae (5 species). Two species in each of the plant families Convolvulaceae, Onagraceae, and Malvaceae were hosts. Only one species of host plant was represented in the families Cucurbitaceae, Lamiaceae, Polygonaceae, Verbenaceae, and Zygophyllaceae. More than half of the plant species found as acceptable hosts (Tables 1, 2) were not reported in the host plant list of Russell (1963).

A number of other plants apparently did not serve as hosts for BWWF. Although inspected frequently, Palmer amaranth, *Amaranthus palmeri* S. Wats., did not support BWWF.

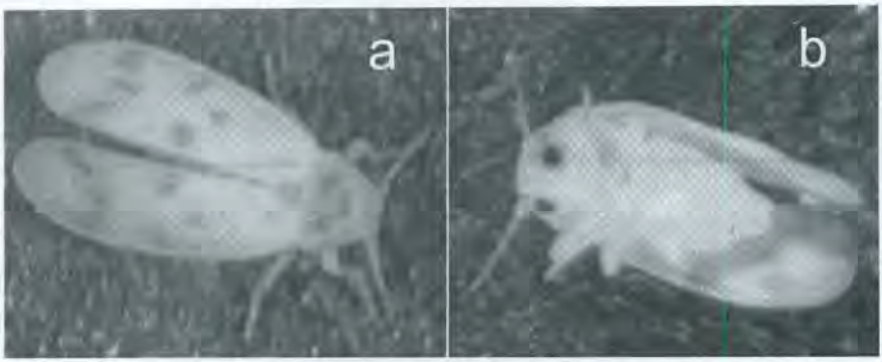

FIG. 1. Dorsal (a) and ventral (b) aspects of bandedwinged whitefly from Wilbarger Co., TX, 2000.

In Knox Co., we found only a very few adults in watermelon, *Citrullus lanatus* (Thunb.) Matsum. & Nakai var. *lanatus*, and cantaloupe, *Cucumis melo* L. subsp. *melo* var. *cantalupensis* Naudin, fields that were adjacent to heavily infested western ragweed, *Ambrosia psilostachya* DC. Because immature stages could not be located after intensive searching, watermelon and cantaloupe were not be considered to be suitable hosts. Adult BWWF were common in vacuum samples taken from evening primroses, *Oenothera laciniata* Hill and *O. grandis* (Britt.) Smyth, for cotton fleahopper, *Pseudatomoscellis seriatus* (Reuter), during the spring of 2001. However, no immature stages were recorded. No adult or immature BWWF were found in vetch, *Vicia* sp., bluebonnet, *Lupinus texensis* Hook., henbit, *Veronica* sp., wild gourds, *Citrullus colocynthis* (L.) Schrad., tansy mustard, *Descurainia pinnata* (Watt.) Britt., lambsquarters, *Chenopodium album* L., tumbleweed, *Salsola tragus* L., or canola, *Brassica napus* L. var. *napus*.

Laboratory studies indicated that this BWWF could transfer from at least eight species of weed hosts and reproduce successfully on cotton (plants identified with footnote a in Table 1). Plant families represented in this host transfer test included Asteraceae, Fabaceae, Onagraceae, and Solanaceae. Giant and western ragweeds (*Ambrosia trifida* L. and *A. psilostachya* DC., respectively) appeared to be the most important alternate hosts based on intensity of infestations and abundance of these two weeds. Liu and Stansly (2000) successfully transferred BWWF from sweet potato, *Ipomoea batatus* (L.) to cotton rose, *Hibiscus mutabilis* L., and roselle, *Hibiscus sabdariffa* L. These workers concluded that BWWF prefers plants in the Malvaceae, which explains the successful transfer of BWWF from four plant families to cotton in our studies.

Honeydew Sugars. Inositol (a polyol) and the sugars glucose and fructose were dominant in BWWF honeydew, while the sugars trehalose, trehalulose, sucrose, and melezitose were present but only in low concentrations (Fig. 2). Inositol, trehalose, glucose, and fructose are considered to be relatively non-sticky, but trehalulose, sucrose and melezitose are known to cause sticky lint. Glucose and melezitose may cause sticky lint problems at concentrations >0.60% (w/w), but sucrose and trehalulose cause sticky lint problems when at concentrations as low as 0.08-0.10% (Miller et al. 1994, Hequet 2003). Hendrix et al. (2002) presented graphs showing relative concentrations of various sugars in the BWWF honeydew, and glucose and fructose were the primary sugars in samples taken in Maricopa, AZ and Lubbock, TX. The sugar concentrations in those samples were comparable to the Vernon, TX samples (Fig. 2).

TABLE 1. Host Plants of Bandedwinged Whitefly During Late Summer and Early Fall in the Northern Texas Rolling Plains. 2000.

Scientific name	Common name	County[b]	Abundance	
			Immature	Adult
Family Asteraceae				
Ambrosia trifida L.	Giant ragweed[a]	K	heavy	heavy
A. psilostachya DC.	Western ragweed[a]	K, Hs	heavy	mod.
Grindelia lanceolata Nutt.	Spinytooth gumweed	K	few	none
Helianthus annuus L.	Common sunflower	Hd	low	low
H. maximiliani Schrad.	Maximilian sunflower	K, W, Hd	moderate	low
H. petiolaris Nutt.	Prairie sunflower	K, W, Hs	moderate	low
Heterotheca subaxillaris (Lam.) Britt. & Rusby	Camphorweed[a]	K	heavy	low
Verbesina encelioides (Cav.) Benth. & Hook.	Cowpen daisy[a]	K, Hs	heavy	mod.
Xanthium spinosum L.	Spiny cocklebur	K, Hd	heavy	mod.
Family Convolvulaceae				
Ipomoea purpurea L.	Tall morning-glory	K	moderate	none
Convolvulus arvensis L.	Field bindweed	W	moderate	mod.
Family Cucurbitaceae				
Cucurbita pepo (L.)	Zucchini squash	W	low	few
Family Fabaceae				
Arachis hypogaea L.	Peanut	K	few	none
Cyamopsis tetragonoloba (L.) Tauber	Guar[a]	K	heavy	mod.
Glycine max (L.) Merrill	Soybean	K, W	low	none
Medicago sativa L.	Alfalfa	K	low	none
Family Malvaceae				
Gossypium hirsutum L.	Cotton	Hd, K, W	heavy	heavy
Family Onagraceae				
Gaura sinuata Nutt.	Wavy-leaf gaura[a]	K	low	heavy
G. villosa Torrey	Hairy gaura	K	low	low
Family Polygonaceae				
Polygonum amphibium L.	Water smartweed	Hs	low	few
Family Solanaceae				
Physalis cinerascens (Duval) Hitchc.	Beach groundcherry[a]	Hd	heavy	heavy
Quincula lobata (Torrey) Raf.	Purple groundcherry	Hd	heavy	mod.
Solanum dimidiatum Raf.	Western horsenettle[a]	Hs	moderate	low
S. elaeagnifolium Cav.	Silverleaf nightshade	K	heavy	mod.
S. melongena L.	Eggplant	W	moderate	few
Family Verbenaceae				
Verbena bracteata Lag. & Rodr.	Prostrate vervain	Hs	low	few
Family Zygophyllaceae				
Tribulus terrestris L.	Goathead	K	low	few

[a] Adult whiteflies reared from this wild host completed one generation on cotton.
[b] Hd = Hardeman, Hs = Haskell, K = Knox, W = Wilbarger counties.

TABLE 2. Host Plants of Bandedwinged Whitefly in Spring and Early Summer in the Northern Texas Rolling Plains. 2001.

Scientific name	Common name	County[a]	Abundance	
			Immature	Adult
Family Asteraceae				
Ambrosia trifida L.	Giant ragweed	K, Hs	heavy	heavy
A. psilostachya DC.	Western ragweed	K, Hs	heavy	heavy
Aphanostephus ramosissimus DC.	Plains lazy daisy	K	low	few
Conyza canadensis (L.) Cronquist	Horse-tail Conyza	K	mod.	mod.
Dracopis amplexicaulis (Vahl) Cass.	Clasping coneflower	K, Hs, W	few	few
Grindelia lanceolata Nutt.	Spinytooth gumweed	K	few	none
Helianthus annuus L.	Common sunflower	Hd	mod.	mod.
H. maximiliani Schrad.	Maximilian sunflower	K, W	low	low
Heterotheca subaxillaris (Lam.) Britton & Rusby	Camphorweed	K	low	low
Verbesina encelioides (Cav.) Benth. & Hook.	Cowpen daisy	K	low	few
Family Fabaceae				
Trifolium hirtum All.	Rose clover	K	low	none
Family Lamiaceae				
Monarda pectinata Nutt.	Plains beebalm	K	heavy	heavy
Family Malvaceae				
Abelmoschus esculentus (L.)	Okra	Hd	low	heavy
Family Onagraceae				
Gaura sinuata Nutt.	Wavy-leaf gaura	K	low	low
G. villosa Torrey	Hairy gaura	K	low	low
Family Solanaceae				
Lycopersicon esculentum Miller	Tomato	W	few	few
Solanum elaeagnifolium Cav.	Silverleaf nightshade	K	mod.	mod.
S. tuberosum L.	Potato	K	heavy	heavy
S. rostratum Dunal	Buffalobur	K	low	low

[a] Hd = Hardeman, Hs = Haskell, K = Knox, W = Wilbarger counties.

Both cotton aphids and BWWF were present on cotton leaves in the field where cotton samples were taken for sticky lint analysis. On 15 September, average numbers of immature BWWF and cotton aphids were 67.8 ± 30.6 and 6.8 ± 3.2, respectively. Average numbers of BWWF and cotton aphids were 33.5 ± 13.7 and 7.2 ± 3.4, respectively, on 22 September. The automated sticky detector (H2SD, Cotton Inc. instrument) indicated that the cotton lint was moderately sticky with average readings of 15.5 ± 2.1. The manual sticky detector (SCT, USDA instrument) indicated that cotton lint was slightly sticky, and average sticky readings were 4.6 ± 0.1. Melezitose and sucrose concentrations on tested lint were very low (Fig. 2) indicating that cotton aphids contributed little of the sugars present on the lint.

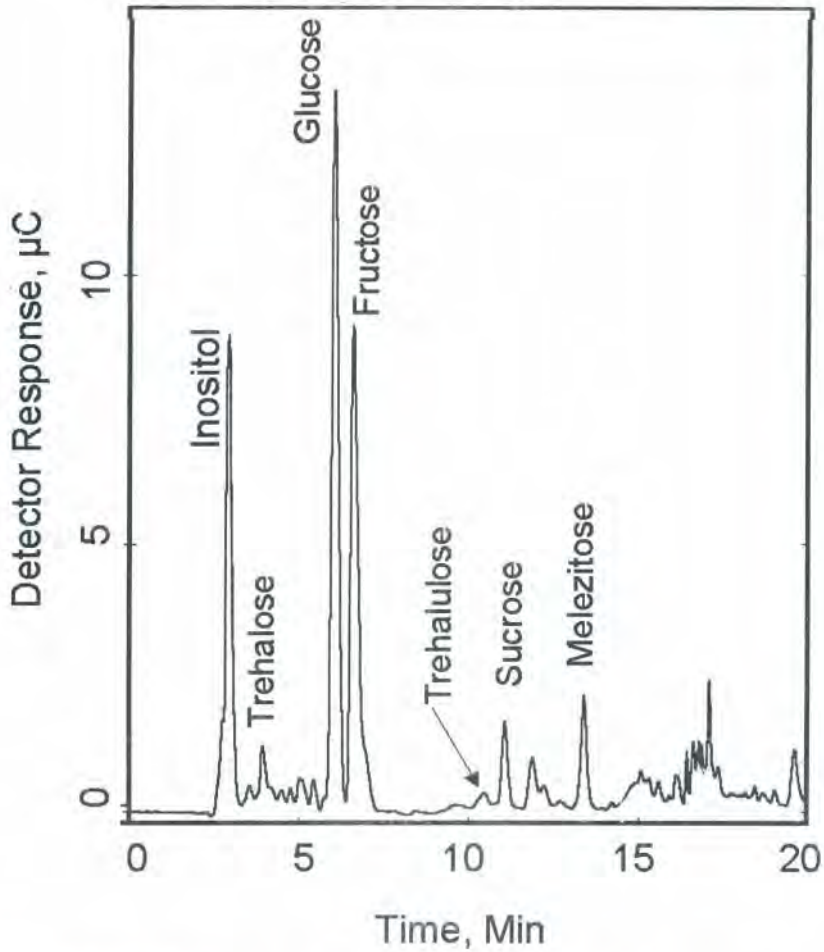

FIG. 2. HPLC of honeydew from bandedwinged whiteflies feeding on cotton leaves. Wilbarger Co., TX, 2000.

Both sticky detectors indicated that BWWF honeydew was only moderately to slightly sticky compared to equivalent concentrations of cotton aphid and silverleaf whitefly, *Bemisia argentifolii* Bellows and Pering, honeydew. Based on infestations on the day of lint harvest, 22 September, our data suggest that BWWF does not portend a significant sticky lint problem during September when numbers of immatures are ≤ 33/leaf. Populations greater than 33/leaf may not cause sticky problems, but we were not able to define the threshold.

Summer Populations. Average numbers of BWWF were high during 1988 at Chillicothe, and a second but somewhat smaller peak in average seasonal abundance occurred in 2000 (Table 3). While BWWF were probably present during the years 1990-1999, it is unlikely that high numbers occurred, otherwise samples would have been taken during the course

TABLE 3. Numbers ($\bar{x} \pm$ SE) of Immature Bandedwinged Whiteflies per Leaf at the Texas Agricultural Experiment Station, Chillicothe, Hardeman Co., Texas.

Year [a]	Planting Date	Avg. No. per Leaf	Max. No. per Leaf	Date of Max. No.	Sample Dates:	
					N	Range
1988	23 May	9.5 ± 2.2	12.7 ± 6.6	19 Aug.	3	12 - 26 Aug.
1989	24 May	0.6 ± 0.1	2.0 ± 0.4	29 Aug.	8	28 July - 19 Sept.
2000	17 May	2.3 ± 0.4	2.7 ± 0.8	7 Sept.	3	21 Aug. - 7 Sept.
2001	24 May	1.3 ± 0.6	4.0 ± 0.8	6 Aug.	6	31 July - 4 Sept.
2002	31 May	0.6 ± 0.2	2.1 ± 1.0	27 Aug.	8	7 Aug. - 25 Sept.

[a] Cotton grown dryland in 1988, 1989, 2000, and irrigated in 2001, 2002

TABLE 4. Numbers ($\bar{x} \pm$ SE) of Immature Bandedwinged Whiteflies per Leaf at the Texas Agricultural Experiment Station, Munday, Knox Co., Texas.

Year [a]	Planting Date	Avg. No. per Leaf	Max. No. per Leaf	Date of Max. No.	Sample Dates:	
					N	Range
1999	19 May	10.4 ± 1.2	12.6 ± 1.2	20 Sept.	4	30 Aug. - 20 Sept.
2000	24 May	6.9 ± 2.0	10.6 ± 6.2	13 Sept.	4	30 Aug. - 20 Sept.
2001	16 May	0.4 ± 0.1	0.9 ± 0.1	1 Aug.	12	3 July - 19 Sept.
2002	18 May	0.1 ± 0.0	0.1 ± 0.1	13 Aug.	2	7 - 13 Aug .

[a] Cotton grown dryland in 1999, and irrigated in 2000-2002.

of cotton aphid sampling in those years. Average numbers of BWWF were higher in 1999 and 2000, compared to numbers in 2001 and 2002, at Munday (Table 4). The status of BWWF populations at Munday prior to 1999 is not known. As discussed previously, sampling for BWWF commenced when it became apparent that numbers were increasing in the cotton aphid plots. The sampling intervals indicate that the timing of population peaks was inconsistent. In years when samples were taken for at least four weeks (Tables 3, 4), peak population numbers occurred from 1 August to 20 September.

Correlations were calculated between average and maximum numbers of BWWF (Tables 3, 4) and temperature and rainfall (Tables 5, 6) for the Chillicothe and Munday data combined. Average numbers of BWWF per leaf were significantly and positively correlated with number of days in August with temperatures >37.8°C and with total number of days in August and September combined with temperatures >37.8°C ($r = 0.751$, $P = 0.020$, and $r = 0.711$, $P = 0.032$, respectively, n = 9). Maximum numbers of BWWF per leaf were significantly and positively correlated with number of days in August with temperatures >37.8°C and with total number of days in August and September combined with temperatures >37.8°C ($r = 0.690$, $P = 0.040$, and $r = 0.659$, $P = 0.053$, respectively, n = 9). Correlations with average daily temperatures and

average maximum daily temperature during August or September and with total rainfall during August or September were not significant (all $r < 0.532$, all $P > 0.140$, n = 9). The lack of correlations with rainfall may have been a result of some tests being grown dryland and some being irrigated. Rainfall would not have affected leaf moisture in irrigated cotton to the same extent that it would in dryland cotton, and Slosser et al. (1992) reported that BWWF numbers were negatively correlated with percentage leaf moisture.

TABLE 5. Temperature and Rainfall at the Texas Agricultural Experiment Station, Chillicothe, Hardeman Co., Texas.

Year	Month	Average Temp. (°C)	Average Max. Temp.	No. Days Temp. > 37.8°C	Total Rain (mm)
1988	Aug.	29.5	37.1	15	14.5
	Sept.	23.7	31.0	0	162.3
1989	Aug.	27.6	33.6	6	64.3
	Sept.	21.8	28.6	1	147.6
2000	Aug.	31.2	39.1	26	0.0
	Sept.	25.5	34.1	7	2.8
2001	Aug.	28.6	35.5	7	35.6
	Sept.	23.3	30.4	1	39.9
2002	Aug.	28.5	35.3	2	14.0
	Sept.	24.2	31.9	1	25.7
1983-2002	Aug.	28.6	35.6	9	60.5
	Sept.	23.9	30.8	2	81.8

These correlation analyses indicate that high BWWF numbers were related to above average temperature patterns during August and September. High BWWF numbers in 1988 and and 2000 at Chillicothe (Table 3) and in 1999 and 2000 at Munday (Table 4) were associated with an above average number of days that temperatures exceeded 37.8°C. Daily temperatures (average daily temperatures or average maximum temperatures for a month) were not influential compared to months with a series of days when temperatures >37.8°C. Butler (1967) investigated development time from egg to adult at constant temperatures of 23.9, 29.4, and 35.0°C (75, 85, and 95°F, respectively), and he found that generation time was shortest (20.7 days) at 35.0°C. He also investigated four ranges of fluctuating temperatures, and he reported that generation time was shortest (20.3 days) under fluctuating temperatures of 29.5 - 40.6°C (85-105°F). These results indicate that the generation time for BWWF is shortest when temperatures are near 37.8°C (100°F), and this is the reason for selecting 37.8°C in the correlation analyses. Byrne and von Bretzel (1987) reported that BWWF populations peak during the hottest part of the summer in Arizona. While the timing of our sampling data was not consistent across years, our conclusion that high BWWF populations were associated with summers with patterns of high temperatures is supported by reports of these other researchers.

TABLE 6. Temperature and Rainfall at the Texas Agricultural Experiment Station, Munday, Knox Co., Texas.

Year	Month	Average Temp. (°C)	Average Max. Temp.	No. Days Temp. > 37.8 °F	Total Rain (mm)
1999	Aug.	32.6	40.9	30	63.0
	Sept.	26.0	33.8	8	27.2
2000	Aug.	31.0	38.8	21	0.0
	Sept.	26.2	35.0	9	0.0
2001	Aug.	28.4	35.3	5	43.7
	Sept.	23.3	30.4	0	78.0
2002	Aug.	28.6	35.3	3	38.9
	Sept.	24.6	32.0	0	30.0
1983-	Aug.	28.9	35.9	10	57.9
2002	Sept.	24.5	31.4	3	70.1

Influence of Boll Weevil Eradication. The Texas Boll Weevil Eradication Program was in operation during 2000 in the Northern Rolling Plains Zone (first full year at the Chillicothe location) and in the Rolling Plains Central Zone (third full year at the Munday location). There was an average of 1.52 applications per acre (per 0.405 ha) of ULV malathion in the Rolling Plains Central Zone in 2000 and 9.11 applications per acre in the Northern Rolling Plains Zone (Allen et al. 2004). However, BWWF infestations were common in both zones and included heavily infested and lightly infested cotton fields. Data for 2000 at Chillicothe and Munday (Tables 3, 4) do not represent the range in infestation severity encountered in the northern and central Rolling Plains. Bandedwinged whiteflies collected for identification were obtained from a very heavily infested field in Wilbarger Co. (northern Rolling Plains), and the lint samples for sticky analysis were collected from a heavily infested field in Knox Co. It appears that high temperature patterns, rather than the use of ULV malathion for boll weevil eradication, were responsible for BWWF outbreaks in these two zones.

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Dr. R. J. Gill, California Department of Food and Agriculture, Plant Pest Diagnostics Branch, Sacramento, confirmed the identification of the bandedwinged whitefly as *Trialeurodes abutiloneus* (Haldeman), and he has all BWWF samples collected for identification. Steve Dowhower, Texas Agricultural Experiment Station, Vernon, and J. F. Cadenhead, Texas Cooperative Extension, Vernon, assisted with plant identifications.

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LIGHT RESPONSE BY FRANKLINIELLA OCCIDENTALIS TO WHITE FLUORESCENT LIGHT FILTERED THROUGH COLOR FILMS AND ULTRAVIOLET- AND BLUE LIGHT-EMMITING DIODES

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ABSTRACT

Western flower thrips (WFT), *Frankliniella occidentalis* (Pergande), are economic pests worldwide. A study was conducted to obtain efficacy data to help develop traps for monitoring WFT population variations in the field and greenhouses. Spectral responses were determined using blue sticky card (BC) traps equipped with light-emitting diodes (LEDs) in the field and a parti-colored light array (PLA) system in the laboratory under darkroom conditions. BC traps equipped with ultraviolet (UV) LEDs were more attractive to this thrips as compared with blue LEDs. Distances from the point of thrips release to the PLA affected the number of thrips caught independently of trap color.

INTRODUCTION

Western flower thrips (WFT), *Frankliniella occidentalis* (Pergande), are among the most important pests attacking a wide variety of plant species including cotton grown in the Western United States (Leigh et al. 1996). Sampling and population monitoring methods are urgently needed in WFT management programs. Ultraviolet (UV) light has been reported to attract many insect species (Hienton 1974). Blue and white colors have been reported to be more attractive to WFT compared with other colors (Beavers et al. 1971, Chang 1990, Cho et al. 1995, Leigh et al. 1996, Mateus and Maxia 1995, Chu et al. 2000, Roditakis et al. 2001, Liu and Chu 2004). Our goal was to develop a trap for field monitoring and potential control of WFT in greenhouse crops. We report here on the results of research to determine the attractiveness of various light sources for the capture of WFT.

MATERIALS AND METHODS

A parti-colored Light Array (PLA) system was arranged in three rows of 10 juxtaposed spaced light source boxes that were stacked vertically against a wall in a (7.9 x 5.2 x 2.7m) (L x W x H) darkroom (Fig. 1). Light source treatments were randomized within two columns of three boxes each. There were five replicates. Each light source was installed in a 25 x 25 x 11cm (L x W x D) plastic box. The inside of each light source box

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was lined with aluminum foil. The front of the box was covered with black cloth with a white fluorescent 9.5 x 9.5cm window area of clear plastic. Each light source box contained one compact light bulb (15 watt, 120V, 60Hz, 750 lumens, Phillips Lighting Co., Somerset, NJ), except for boxes with one UV light bulb (fluorescent black light, BPESL15T, 15W, 120V, 50-60Hz, 200mA, Pico Rivera, CA). Front windows of each light source box were covered with one of five randomly assigned color transparency per replication. The color transparencies obtained from Rosco Roscolux (Stamford, CT) were: #26 Light Red, #10 Medium Yellow, #88 Light Green, #85 Deep Blue, and #00 Clear (used for both white and UV light bulbs). The front of each transparency (9.5 x 9.5cm) was coated with Tanglefoot® Insect Trap Coating (Grand Rapids, MI). Captured WFT were counted on the 8 x 8cm center area of each transparency. Transparencies were held in position on the cover of each light box using magnetic strips on the outside edges of the transparent windows. Spectra of the color transparencies were measured with an ASD-FR spectrometer (Full Range model, Analytical Spectral Devices, Boulder, CO). A halogen light source was directed at a 99% Spectralon panel (Labsphere, Inc., North Sutton, NH) which reflected the light energy to the spectrometer which was positioned perpendicularly to the panel. Transparency spectra were measured by positioning the transparencies about 10cm in front of the light source and recording the spectra. Peak wavelengths were determined by analyzing the spectra from each transparency. The peak wavelengths of the transparencies were: 545nm for #10 Medium Yellow, 650nm for #26 Light Red, 460nm for #85 Deep Blue, and 512nm for #88 Light Green. The peak wavelength of the UV fluorescent light was 369nm. The PLA was powered continuously during the experimental period. Darkroom conditions were 28-30° C and 20-30% RH during the experiment.

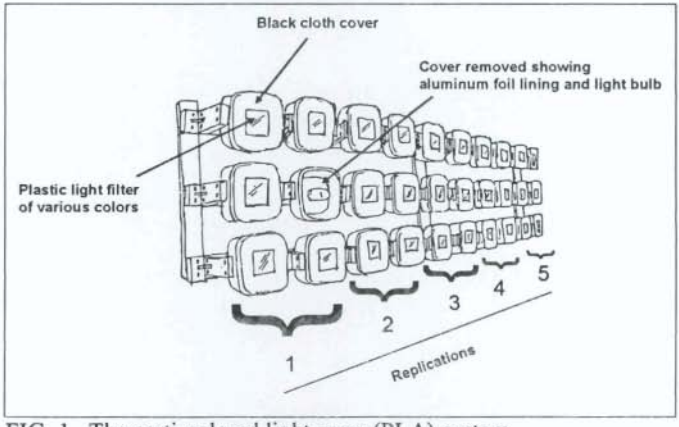

FIG. 1. The parti-colored light array (PLA) system.

LED light lamps used in the studies were 12° 398nm UV LED (L200CUV405-12D, Ledtronics, Inc., Torrance, CA) and 45° 465nm blue LED lamps (Nichia NSPB320BS, Nichia America Corp., Mountville, PA). LED light spectra were measured using the ASD-FR spectrometer by pointing the sensor directly at the LEDs. Peak wavelengths were determined by plotting the spectra from each light source. The LED lamps were fitted on hair clips and energized with 220 ohm resistors *via* a 6V direct current (DC)

adaptor (Radio Shack® Co., Fort Worth, TX, USA) as described elsewhere (Chu et al. 2003). The LED lamps used for Experiments 2 and 3 were connected to a standard 110V alternating current electricity source. For Experiment 4, the LED lamps were connected to a 12V solar/battery operated energizer (Model MAG 12-SP, Parker MaCrory MFG, Co., Kansas City, MO). The energizer was turned on manually at night.

Experiment 1 (WFT attraction to light colors) was conducted in a randomized complete block design. Treatments were six light colors as described earlier. WFT were released at the rate of 1000, 2000 or 3000 for each test 83 or 165cm from the light sources. WFT were collected in sweep nets in an alfalfa, *Medicago sativa* (L.), field at Maricopa, AZ, and released in the darkroom each day for six days. WFT caught over 24h on each Tanglefoot® coated transparencies were counted after each release.

Experiment 2 (WFT catches on LED-BC traps in greenhouse grown alfalfa) was conducted in eight (124 x 60 x 132cm) (L x W x H) wood framed cages (replicates). Cages were covered with 72 mesh plastic screen. Alfalfa *cv.* CUF 101 was planted in pots (10-15 plants per pot) and six pots were placed in each cage. Treatments were BC traps equipped with blue or UV LED lamps. BC traps without LED were the controls. BC traps (blue Takitrap®, Oecos Ltd., Kimpton, Hertfordshire, England) were 10.0 x 10.5cm in size. The traps were spaced 45cm apart in the cages. Trap bottoms were 3cm above the plant terminals. The plants were 10-15cm high during the experiment. WFT were collected as described earlier and 50 were released in each cage on each day for four weeks from 11 February to 3 March 2004. The LED lamps were illuminated from 1800 to 0700 hours. Traps were retrieved weekly and trap catches were counted. New traps were placed in cages weekly.

Experiment 3 (WFT catches on LED-BC traps in field grown fava bean, *Vicia fava* (L.), was conducted in a randomized complete block design with seven replicates in a fava bean field at Phoenix, AZ. The trap treatments were BC equipped with UV LED or blue LED traps. BC traps not equipped with LED were used as controls. The traps were set 800cm apart along a row. Fava bean rows were spaced 100cm apart, and the plants were spaced about 3cm apart. The plants were about 40-45cm high during the experiment. The traps were mounted vertically on wire stakes. The trap bottoms were about 5cm above the plant tops. Each replicate was set in every other row for seven rows. The traps were retrieved and replaced with new ones daily.

Experiment 4 (WFT catches on LED-BC traps in field grown cotton *Gossypium hirsutum* (L.), was conducted in a randomized complete block design with four replicates in cotton research plots in Holtville, CA. Cotton cv. DPL 5415 was planted and irrigated on 30 March 2004. Standard agronomic practices were followed. Treatments were the same as described for Experiment 3. Traps were retrieved weekly and WFT and bean thrips, *Caliothrips phaseoli* (Hood), catches were counted. New traps were placed in the plots at the same locations during the experimental period from 21 May to 18 June 2004.

The numbers of WFT and bean thrips caught were analyzed using ANOVA. The means were separated using Tukey's test at $P = 0.05$ (Anonymous 1994).

RESULTS AND DISCUSSION

Overwhelmingly more WFT were attracted to 369nm wavelength UV (black) fluorescent light in Experiment 1 (WFT attraction to light colors) compared with white, red (650nm), medium yellow (545nm), light green (512nm), and deep blue (460nm) colors under the darkroom experiment (Table 1).

In Experiments 2 and 3 (WFT catches on LED-BC traps in greenhouse grown alfalfa and in field grown fava bean), mean numbers of WFT caught on 398nm UV LED-BC traps were greater compared with 465nm blue LED-BC traps (Tables 2 and 3). For both experiments, Blue LED-BC traps caught more WFT compared with BC controls.

TABLE 1. Mean Thrips (± SE) Numbers of *Frankliniella occidentalis* (Pergrande) Caught per Colored Light Source When Releases Were Made at Distances of 83 and 165 Centimeter Distances from the Six Different Light Sources in a Darkroom.

	Mean numbers/trap/24 h after release		
	No. thrips released		
Light color	1000	2000	3000
	83cm from light source		
UV	82.6±25.5a[a]	148.0±27.2a	221.4±52.2a
Blue	1.8±0.6b	4.8±1.3b	5.2±3.5b
Green	1.8±0.6b	2.0±0.9b	3.4±1.7b
Yellow	1.2±0.6b	1.4±0.2b	1.8±0.7b
Red	1.2±0.6b	2.0±0.7b	2.0±0.9b
Clear	4.0±1.7b	3.6±1.2b	6.6±1.4b
F, prob.	9.9, <0.001	28.3, <0.001	17.2, <0.001
	165 cm from light source		
UV	9.2±2.9a	32.8±11.5a	40.8±2.2a
Blue	0.8±0.6b	0.8±0.6b	0.4±0.4b
Green	0.2±0.2b	0.8±0.2b	1.0±0.8b
Yellow	0.2±0.2b	0.2±0.2b	0.8±0.8b
Red	0.2±0.2b	1.6±0.7b	1.2±0.4b
Clear	0.8±0.6b	1.8±1.6b	6.4±2.9b
F, prob.	8.0, <0.001	7.5, <0.001	102.3, <0.001

[a] Means in a column not followed by the same letter are significantly different by Tukey's HSD, $P = 0.05$, df = 5, 30.

TABLE 2. Mean (± SE) Numbers of *Frankliniella occidentalis* (Pergrande) Caught on Blue Sticky Card Traps With or Without LEDs from Alfalfa in a Greenhouse, 2004.

	No./trap/week				
LED type	11 Feb	18 Feb	25 Feb	3 March	Mean
UV	4.1±0.9a[a]	4.6±0.4a	5.9±0.2a	7.6±0.4a	5.6±0.3a
Blue	3.1±0.6ab	3.9±0.6a	4.3±0.4b	6.8±0.5a	4.5±0.4b
Control[b]	1.1±0.2b	1.1±0.2b	1.5±0.2b	2.8±0.3b	1.6±0.1c
F, prob.	7.3, <0.007	76.1, <0.001	177.7, <0.001	63.5, <0.001	88.9, <0.001

[a] Means in a column not followed by the same letter are significantly different by Tukey's HSD, $P = 0.05$, df = 2, 14.
[b] No LED.

TABLE 3. Mean (± SE) Numbers of *Frankliniella occidentalis* (Pergrande) Caught on Blue Sticky Card Traps With or Without LEDs in a Fava Bean Field, Phoenix, AZ, 2004.

	No./trap/day				
LED type	12 March	16 March	17 March	18 March	Mean
UV	22.0±2.2a[a]	33.7±4.8a	29.6±3.5a	22.6±3.0a	27.5±1.5a
Blue	20.8±3.5a	31.0±2.4a	25.4±3.3a	17.4±2.8a	23.9±1.5b
Control[b]	14.5±4.7b	20.9±1.2b	14.7±4.6b	6.1±1.7b	14.5±1.2c
F, prob.	7.4, <0.006	4.6, <0.024	6.7, <0.007	10.6, <0.001	52.9, <0.001

[a] Means in a column not followed by the same letter are significantly different by Tukey's HSD, $P = 0.05$, df = 2, 12.
[b] No LED.

Mean numbers of WFT and bean thrips caught on 465nm blue LED-BC traps in Experiment 4 (WFT catches on LED-BC traps in field grown cotton) were greater compared with 398nm UV LED-BC traps (Table 4).

TABLE 4. Mean (± SE) Numbers of *Frankliniella occidentalis* (Pergrande) and *Caliothrips phaseoli* (Hood) Caught on Blue Sticky Card Traps in a Cotton Field in Holtville, CA, 2004.

Light color	No. /trap/week				
	21 May	28 May	4 June	11 June	Mean
			F. occidentalis		
UV LED	539.5±47.6b[a]	410.8±49.2ab	433.2±46.4b	324.3±12.8b	427.0±20.1b
Blue LED	858.7±45.5a	505.7±46.0a	539.2±39.4a	604.2±36.8a	626.9±31.4a
Control[b]	524.0±27.2b	328.2±53.7b	360.3±56.8b	277.8±13.4b	372.6±23.8b
F, prob.	23.0, <0.001	13.4, <0.002	12.3, <0.002	90.8, <0.001	60.5, <0.001
			C. phaseoli		
UV LED	16.0±2.5a	23.7±3.9b	17.0±2.8b	14.5±1.8ab	17.8±2.1b
Blue LED	17.3±1.4a	41.0±5.3a	34.0±4.8a	21.5±3.2a	28.5±3.0a
Control[b]	14.2±1.8a	25.8±5.3ab	19.3±3.2b	10.8±1.1b	17.5±2.2b
F, prob.	0.7, <0.544	5.1, <0.030	9.5, <0.005	8.5, <0.010	8.3, <0.008

[a] Means in a column not followed by the same letter are significantly different by Tukey's HSD, $P = 0.05$, df = 2, 10.
[b] No LED.

Reasons for the differences in trap catches between 398nm UV LED- and 465nm blue LED-BC traps in field grown fava bean in Phoenix, AZ, and cotton in Holtville, CA, are not known, but may be due to higher WFT population in cotton. More WFT were caught in traps placed 83cm from the PLA compared with 165cm from the light source (Table 1). Catches of WFT at 369nm UV light sources ranged from 21 to 111 times more compared with numbers caught at any other light source at 83cm distance and 6-464 times at 165cm distance. These results suggest that WFT response to light occurs over short distances.

Thrips orientation to colors may be compounded under field conditions compared with their behavior in darkroom. Stavisky et al. (2002) found UV reflective mulch reduced *Frankliniella* spp. densities by 45% and tomato spotted wilt incidence by 50%, suggesting that UV reflectance-induced color changes in plant canopy adversely affected *Frankliniella* spp. landing on the hosts. Other research indicates that 290-320nm UVB is related to the trophic-level interactions between algae density and the density of predator larval chironomid tubes (Diptera: Chironomidae) (Bothwell et al. 1994). Bean thrips herbivory is reduced by UVB radiation (Mazza et al. 1999) suggesting changes in leaf chemistry (Warren 2002) under field conditions that interact with the plant pests.

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FITNESS OF GREENBUG[1] ON WILD AND CULTIVATED GRASSES

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ABSTRACT

Pre-reproductive period, total fecundity, and longevity of biotype I greenbug, *Schizaphis graminum* (Rondani), were evaluated on four wild grasses, resistant 'LG35' sorghum, susceptible 'RTx430' sorghum, resistant 'GRS1201' wheat, and susceptible 'Custer' wheat. Pre-reproductive period per biotype I greenbug was longest on barnyardgrass (7.5 days). Pre-reproductive periods of greenbugs were not significantly different on susceptible versus resistant sorghum or wheat. Among the four wild grasses, average total fecundity per greenbug was least on barnyardgrass (15.9 nymphs) and greatest on Johnsongrass (57.1 nymphs). Total numbers of nymphs produced per greenbug differed significantly on resistant versus susceptible sorghum (49.1 versus 61.7) and wheat (49.1 versus 64.1). Longevity per greenbug was shortest on barnyardgrass (14.8 days) and longest on grasses of the genus *Sorghum* (27.5 to 29.4 days). The longevity of greenbugs did not differ significantly between susceptible versus resistant sorghum or wheat. Barnyardgrass was the least suitable host for biotype I greenbugs. Johnsongrass and jointed goatgrass were better hosts compared to barnyardgrass or western wheatgrass. These results show the importance of weedy grasses in the temporal survival of greenbugs and call for a thorough survey of the susceptibility of other wild grasses so damage by greenbugs can be prevented in commercial cereal crops.

INTRODUCTION

Greenbug, *Schizaphis graminum* (Rondani), occurs on more than 70 species of grasses in 44 genera (Michels 1986). The aphid is a major pest of sorghum, *Sorghum bicolor* (L.) Moench, wheat, *Triticum aestivum* L., and other small grains. Annual losses by greenbug to commercial cereal crops often exceed $100 million each year in the Great Plains (Webster et al. 2000).

In the southern United States, greenbugs survive on wild grasses when cultivated hosts such as sorghum and wheat are not available. Greenbugs live on cultivated grass crops, volunteer plants, and wild grasses during the summer (Dahms et al. 1954). Daniels

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(1961) reported oversummering greenbugs on 20 species of wild grasses. In addition to being hosts, non-cultivated grasses are believed to harbor as yet unknown biotypes of greenbug (Porter et al. 1997, Kindler and Hays 1999, Anstead et al. 2003). In northern, cooler climates, biotypic variation is believed to result from sexual reproduction of greenbugs on cool-season grasses (Porter et al. 1997). Sexual reproduction on non-cultivated grasses such as Kentucky bluegrass, *Poa pratensis* L., in temperate regions north of the 35th parallel in the United States allows genetic recombination leading to diversity among populations of greenbugs (Webster and Phillips 1912, Puterka and Peters 1990). Warm-season, non-cultivated grasses harbor as yet unidentified biotypes of greenbugs that may move to crop plants in southern, warmer regions of the United States (Porter et al. 1997, Anstead et al. 2003). Weedy grasses also are reservoirs of plant viruses transmitted by greenbugs (Daniels and Toler 1969, Blackman and Eastop 1984).

The occurrence of biotypes has challenged the strategy of using resistant plants to control greenbugs (Kindler and Hays 1999). Eleven biotypes of greenbugs (A through K) have been identified (Wood 1961, Harvey and Hackerott 1969, Teetes et al. 1975, Porter et al. 1982, Kindler and Spomer 1985, Puterka et al. 1988, Harvey et al. 1991, Beregovoy and Peters 1994, Harvey et al. 1997). Greenbug biotype I is believed to be dominant in Texas (Pendleton, unpublished data).

Despite the potential importance of wild grasses as alternate hosts and reservoirs of diseases, studies of interactions between wild grasses and greenbugs have been limited. Although preferences, reproduction, and survival of unknown biotypes of greenbugs have been evaluated on different species of wild grasses (Dahms et al. 1954, Dickson and Laird 1969, Kieckhefer and Stoner 1978, Stoner and Kieckhefer 1979, Kieckhefer 1983), only Kindler and Spomer (1986) and Kindler and Hays (1999) assessed the suitability of wild hosts for survival and reproduction of known biotypes of greenbugs, and greenbug biotype I was not known when Kindler and Spomer (1986) conducted their study. Therefore, pre-reproductive period, total fecundity, and longevity of biotype I greenbug were assessed to determine fitness on four species of wild grasses and resistant and susceptible lines of sorghum and wheat.

MATERIALS AND METHODS

Pre-reproductive period, total fecundity, and longevity per biotype I greenbug were assessed in May-June, August-October, and November-December 2002 in a greenhouse at West Texas A&M University, Canyon, TX. Seeds of Johnsongrass, *Sorghum halepense* (L.) Pers., and 'Arriba' western wheatgrass, *Agropyron smithii* Rydb., were obtained from Curtis and Curtis, Inc., Clovis, NM. Valley Seed Service of Fresno, CA, provided seeds of jointed goatgrass, *Aegilops cylindrica* (Host) Ces., and barnyardgrass, *Echinochloa crusgalli* (L.) Beauv. These grasses were selected because they are related to sorghum or wheat and/or are common weeds. Seeds of biotype I-resistant 'LG35' and biotype I-susceptible 'RTx 430' sorghum were obtained from Gary C. Peterson, Texas Agricultural Experiment Station, Lubbock, TX. David R. Porter, USDA-ARS, Stillwater, OK, provided seeds of biotype I-resistant 'GRS1201' and susceptible 'Custer' wheat used during August-October and November-December.

Seeds of the different kinds of grasses were planted separately by genotype in 20.3-cm diameter plastic pots filled with Miracle-Gro® Enriched Potting Mix (Miracle-Gro Lawn Products, Inc., Marysville, OH). Each pot was covered with a cylindrical, clear plastic cage with organdy cloth glued over the top to allow aeration. The pots were arranged in a randomized complete block design. During May-June 2002, Johnsongrass, jointed goatgrass, western wheat grass, resistant 'LG35' sorghum, and susceptible

'RTx430' sorghum were used. A total of 30 pots was arranged in six blocks, with each kind of grass in each block. During August-October and November-December, all eight kinds of grasses were arranged in seven blocks. Seedlings were thinned to three per pot. Water was added to the plastic saucer at the base of the pot of plants when the soil was dry. The temperature in the greenhouse was 21-32°C.

A pure culture of biotype I greenbugs was maintained on susceptible 'Tx399 x RTx430' sorghum in the greenhouse. A camel-hair brush was used to transfer a single adult greenbug to a 2.5-cm^3 clip cage with two, organdy cloth-covered holes for aeration. A clip cage was clipped onto a leaf of each of three plants per pot 35 days after emergence. Eighteen to 21 clip cages were used for each kind of grass each time.

The original greenbug in each clip cage was discarded after it began producing nymphs. A single nymph was retained in the clip cage. The date of birth was recorded. The greenbug was allowed to mature and produce nymphs that were counted and removed daily by using a camel-hair brush. Total numbers of nymphs produced and days each greenbug survived on each kind of grass were recorded.

Data for pre-reproductive period, total fecundity, and longevity of biotype I greenbug were analyzed by the MIXED procedure (SAS Institute 1999), with time as a random effect and kind of grass as a fixed effect. Blocks were considered replications and clip cages as subsamples for analysis. Denominator degrees of freedom were computed by the Satterthwaite approximation method. Least square means for the tested parameters were estimated by LSMEANS (SAS Institute 1999, $P = 0.05$).

RESULTS AND DISCUSSION

Average pre-reproductive period per biotype I greenbug differed significantly on the different kinds of grasses ($F = 16.13$; df = 7, 111; $P < 0.0001$). Average pre-reproductive period was significantly longest on barnyardgrass (7.5 ± 0.2 days), followed by western wheatgrass (6.5 ± 0.2 days) (Table 1). No significant differences in pre-reproductive periods of greenbugs were found for susceptible versus resistant sorghum or wheat.

TABLE 1. Development and Reproduction of Biotype I Greenbugs on Different Kinds of Grasses.

Grass	n[a]	Pre-reproductive period (days ± SE)[b]	Total fecundity (average number of nymphs/greenbug ± SE)[b]	Longevity (days ± SE)[b]
Barnyardgrass	14	7.5 ± 0.2a	15.9 ± 4.6f	14.8 ± 2.2f
Western wheatgrass	20	6.5 ± 0.2b	22.5 ± 4.2f	17.6 ± 2.0ef
Jointed goatgrass	20	5.3 ± 0.2c	54.5 ± 4.1bcd	19.3 ± 2.0cde
Johnsongrass	19	5.7 ± 0.2c	57.1 ± 4.2abc	29.4 ± 2.0a
'LG35' sorghum	20	5.5 ± 0.2c	49.1 ± 4.1cde	27.5 ± 2.0ab
'RTx430' sorghum	20	5.3 ± 0.2c	61.7 ± 4.2ab	28.8 ± 2.0ab
'GRS1201' wheat	14	5.2 ± 0.2c	49.1 ± 4.6cde	21.9 ± 2.2cd
'Custer' wheat	14	5.4 ± 0.2c	64.1 ± 4.6a	22.8 ± 2.2c

[a]n = number of replications of three clip cages.

[b]Means followed by the same letter in a column are not significantly different (t-test; $P < 0.05$).

Average total fecundity per biotype I greenbug differed significantly on the different kinds of grasses ($F = 32.21$; df = 7, 131; $P < 0.0001$). Among wild grasses, the average total number of nymphs produced per greenbug was significantly greater on Johnsongrass (57.1 ± 4.2) and jointed goatgrass (54.5 ± 4.1) compared to western wheatgrass (22.5 ± 4.2) or barnyardgrass (15.9 ± 4.6) (Table 1). Average total fecundity per greenbug differed significantly on susceptible versus resistant sorghum or wheat.

Average longevity per biotype I greenbug differed significantly on the different kinds of grasses ($F = 14.18$; df = 7, 133; $P < 0.0001$). Average longevity per biotype I greenbug was significantly shortest on barnyardgrass (14.8 ± 2.2 days) (Table 1). Average longevity was significantly longest on grasses of the genus *Sorghum* (27.5 to 29.4 days). Average longevity per greenbug did not differ significantly on susceptible versus resistant sorghum or wheat.

Biotype I greenbugs survived and reproduced on different wild grasses. Of the wild grasses, Johnsongrass and jointed goatgrass were better hosts than barnyardgrass or western wheatgrass, probably because the former two grasses are related to sorghum and wheat, respectively. These results agree generally with Dahms et al. (1954) who found that Johnsongrass and jointed goatgrass were favored hosts for survival of greenbugs of an unknown biotype, perhaps biotype A. In contrast, Dickson and Laird (1969) found that greenbugs, perhaps biotype C, did not survive on barnyardgrass, and did not survive well on Johnsongrass. Biotype I greenbug seems better able to adapt to hosts inhospitable to other biotypes. Kindler and Hays (1999) reported that populations of eight biotypes, including biotype I, lived on western wheatgrass. In our experiment, biotype I greenbugs confined in clip cages survived on western wheatgrass, but average total fecundity per greenbug was less on western wheatgrass than on Johnsongrass or jointed goatgrass. Among the wild grasses, biotype I greenbugs matured latest, produced fewest nymphs, and survived for the shortest time on barnyardgrass.

Damage was visible on susceptible 'RTx430' sorghum and 'Custer' wheat. The area of the leaf inside the clip cage was reddened on susceptible 'RTx430' sorghum and yellowed on susceptible 'Custer' wheat. No symptoms of damage by greenbugs were observed on resistant 'LG35' sorghum or 'GRS1201' wheat. The mechanism of resistance in biotype I greenbug-resistant 'LG35' sorghum and 'GRS1201' wheat seems to be tolerance rather than antibiosis.

Many publications report interactions between greenbugs and cereal crops, but interactions between greenbugs and non-cultivated grasses seldom have been studied. Our results emphasize the importance of wild grasses for sustenance of greenbugs when sorghum and wheat are not available. That biotype I, unlike some previous biotypes of greenbug, survived and reproduced on barnyardgrass underscores the need to understand interactions between greenbugs and wild grasses that aid in the temporal harbor of as yet unidentified biotypes so that future infestations of and damage to cultivated cereal crops might be avoided.

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INTERVALS BETWEEN BOLL WEEVIL (COLEOPTERA: CURCULIONIDAE) OVIPOSITION AND SQUARE ABSCISSION, AND DEVELOPMENT TO ADULTHOOD IN LOWER RIO GRANDE VALLEY, TEXAS, FIELD CONDITIONS

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ABSTRACT

This study reports that 6.2 and 5.3 d in 2002 and 2003, respectively, elapsed between boll weevil, *Anthonomus grandis grandis* Boheman, oviposition and square abscission under field conditions in the Lower Rio Grande Valley of Texas. Oviposition to adult weevil emergence from the square took an average of 18.5 d in 2002 and 16.2 d in 2003. Although significant minimum and maximum daily ambient temperature differences were detected between the separate sampling periods in May and June of 2002, oviposition to square abscission and adult emergence periods were not significantly affected.

INTRODUCTION

Cotton squares abscise after boll weevils, *Anthonomus grandis grandis* Boheman (Coleoptera: Curculionidae), oviposit and second or third instars develop (Coakley et al. 1969). Only two studies, however, have reported on the time from oviposition to square abscission, an important part of the boll weevil's life cycle which affects the time of boll weevil development from egg to adult (Hunter and Pierce 1912). One of these studies concluded that an average of 9.5 d elapsed (Hunter and Pierce 1912). The time from oviposition to emergence of the adult was reported to be 17.3 d (Hunter and Pierce 1912). However, no standard errors and other statistics were provided on either time interval, and the data were collected from a temperate cotton growing area and may not be relevant to boll weevil development in subtropical areas such as the Lower Rio Grande Valley of Texas (Guerra et al. 1982, Summy et al. 1988). In the other study, 3.5-4.9 d elapsed between oviposition and square abscission (Davich et al. 1965), and the time interval was calculated using squares that had boll weevil eggs artificially implanted, which may have unnaturally influenced square abscission cotton growing areas. Another report indicated that square abscission ranged from 5.8 to 18.6 d at 34 to 18°C, respectively, but the study was conducted in the laboratory using excised cotton branches placed in water (Bacheler et al. 1975). This study was conducted to determine the time from oviposition to square abscission, and to adult emergence under field conditions of the Lower Rio Grande Valley.

MATERIALS AND METHODS

Sixty cotton (var. 'NK-2387') plants were flagged in two commercial fields, one planted on 19 March 2002 and the other on 5 March 2003, in Hidalgo County, TX. Squares on the flagged plants were checked daily, 17-24 May and 12-17 June 2002 in the first field, and 2 May-6 June 2003 in the second field. A 1-cm^2 paper tag was fastened by thread to one bract of each oviposition-punctured square, and the date of the puncture and the size of the square were written on the tag. When each square dropped to the ground, the date was recorded and the square was placed alone in a 20-cm^3 cage. Cages with

squares (2002, $n = 27$; 2003, $n = 30$) were placed in furrows that were largely shaded by the crop canopy. All of the squares were 5-7-mm in diameter, except for three in 2002 that were 2-4 mm in diameter. The cages were checked daily for adult boll weevils. Ten squares in which eggs were laid in May 2002 produced adult weevils (three others died as larvae in 5-7-mm-diameter squares), and 14 squares in which eggs were laid in June 2002 produced adults. Data on square abscission and adult emergence times were compared between the squares on which oviposition occurred in May 2002 (abscission, $n = 13$; adult emergence, $n = 10$) versus June 2002 (abscission and adult emergence, $n = 14$), and pooled 2002 data (variances in the May and June sampling periods were homogeneous) was compared to the 2003 data.

Ambient daily minimum and maximum temperatures were recorded at a USDA-ARS weather station located ≈3 km away. Mean (±SE) minimum and maximum air temperatures recorded between first oviposition and last square abscission during the two periods 17 May-3 June and 12-20 June, and between first oviposition and last adult emergence during the two periods 17 May-6 June and 12 June-9 July 2002, and between the pooled 2002 data and data collected 2 May-22 June 2003 were compared. All comparisons were made using the two sample *t*-test (Analytical Software 1998).

RESULTS AND DISCUSSION

No significant differences were detected between the May and June 2002 sampling periods in terms of oviposition to abscission and to adult emergence intervals, so statistics describing pooled data are presented. Square abscission occurred 6.2±0.4 (median=6; range=2-12) d after oviposition in 2002, and 5.9±0.3 (median=6; range=2–9) d in 2003. Adult boll weevils emerged 18.5±0.9 days after oviposition (median=16; range=13-26) in 2002, and 16.2±0.4 (median=17, range=11-21) days in 2003. The mean minimum temperatures that occurred from oviposition until square abscission between 17 May and 3 June, and between 12 and 20 June, were 21.0±0.3 °C and 23.4±0.3 °C (t=2.9, df=1, 25, P=0.0074), respectively, and the mean maximum temperatures between 17 May and 3 June, and between 12 and 20 June, were 31.9±0.4 °C and 36.0±0.4 °C (t=4.89, df=1, 25, P<0.0001), respectively. In 2003, the mean minimum temperature between 2 May and 11 June, from oviposition through abscission, was 23.7±0.3 °C, and the mean maximum temperature was 34.0±0.5 °C. The mean minimum temperatures that occurred from oviposition until adult emergence between 17 May and 6 June, and between 12 June and 9 July, were 21.4±0.2 °C and 23.8±0.3 °C (t=4.5, df=1, 47, P<0.0001), respectively, and the mean maximum temperature between 17 May and 3 June, and between 12 and 20 June was 32.2±0.3 °C and 34.4±0.3 °C (t=3.2, df=1, 25, P=0.0025), respectively. The significant differences between mean minimum and maximum temperatures in the first (May) and second (June) sampling periods did not significantly affect the duration of intervals between oviposition and square abscission and development to the adult stage. The mean minimum and maximum temperatures between 2 May and 22 June, from oviposition through adult emergence, were 23.7±0.3 °C and 34.1±0.3 °C, respectively. No significant differences in square abscission and adult emergence intervals, and pooled mean minimum and maximum temperatures, were detected between years.

The 3.3-3.6-d difference between the longer mean abscission interval in central and northern Texas (Hunter and Pierce 1912) and the Lower Rio Grande Valley was probably the result of environmental differences, including temperature, that are likely greater than those observed between the May and June sampling periods in the Lower Rio Grande Valley, 2002. The 1.0-3.6 shorter abscission intervals observed by Davich et al. (1965) than in Lower Rio Grande Valley field conditions might have been due to the insertion of the boll weevil eggs by drilling holes in the squares with a glass pipette. Adult boll

weevils emerged from the fallen squares 1.2 d later in 2002, and 1.1 d earlier in 2003, than in central and northern Texas (Hunter and Pierce 1912). Variations in adult emergence times were likely affected to some extent by conditions on the soil surface, where most of the boll weevil development, and mortality, occurs (Summy et al. 1986).

This information establishes the period in which boll weevils are protected inside cotton squares in the Lower Rio Grande Valley, even from the insecticides that are most commonly applied (Heilman et al. 1979, Gage et al. 1984, Showler and Scott 2003). Knowledge of the intervals between visible events associated with the weevil's development might help to establish a more rational insecticide intervention trigger (Showler and Cantú 2003) than at 10% oviposition on randomly selected squares, a commonly used threshold in the Lower Rio Grande Valley. The intervals between sprays also might be revised if calculations involve the duration of insecticide residue toxicity and the interval during which boll weevils are protected from the effects of insecticides (Showler and Scott 2003).

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ELECTRICAL STIMULATION OF *SOLENOPSIS INVICTA* TO ENHANCE PHORID FLY, *PSEUDACTEON TRICUSPIS*, DETECTION

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ABSTRACT

An electrical stimulation device consisting of a modified livestock prod was tested for its ability to enhance detection of the phorid fly *Pseudacteon tricuspis* Borgmeier, a parasitoid of the red imported fire ant, *Solenopsis invicta* Buren. At a site heavily infested with phorid flies in Burleson County, Texas, pairs of active fire ant colonies were disturbed simultaneously using the device with light mechanical disturbance on one mound and mechanical disturbance only on the other. Electrical stimulation proved much more effective than mechanical disturbance resulting in less than half the response time (108 seconds vs 271 seconds), more flies per mound (5.7 vs 0.55), and, most importantly, a detection rate of 100% versus only 27% (3 of 11 mounds) for mechanical disturbance.

INTRODUCTION

The decapitating phorid fly, *Pseudacteon tricuspis* Borgmeier, (Diptera: Phoridae) was been released at a number of sites around the United States for control of the red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae). In Texas alone, over 20 releases have been made with an establishment rate of around fifty percent (Gilbert and Patrock 2002). Field detection of phorids is a tedious and often unsuccessful endeavor. The usual procedure is to mechanically disturb a fire ant mound, crush a number of ants, then simply observe the mound for the appearance of phorids (Porter 1998). Another method used by researchers at the University of Texas is to place small piles of fire ant midden material in plastic containers or white ceramic tiles and watch for the appearance of the flies (Gilbert and Patrock 2002). It is common to spend at least 15 minutes observing a single disturbed mound and numerous mounds must be disturbed to have a reasonable degree of confidence of the presence or absence of flies.

In rearing facilities at the USDA-ARS, CMAVE center in Gainesville, Florida, the fly rearing boxes contain two electric plates spaced approximately 1.5 mm apart. When the plates are charged, an ant crossing the gap is "electrically stimulated" to release an alarm pheromone. The result is attack stimulation by the flies and increased oviposition (S.D. Porter, personal communication). The basis for this mechanism is that electrical stimulation causes the release of numerous semiochemicals including alarm and orientation pheromones by the ants. (Vander Meer et al. 2002). Building on this concept, we felt that electrical stimulation of ants in the field could possibly increase the success rate of phorid fly detection. A commercially available electric livestock prod was modified and tested for effectiveness.

MATERIALS AND METHODS

The stimulation device was constructed using an electric livestock prod, TheBlueOne™ LMPlus®, manufactured by Hot Shot® (5441 W. 125th St., Savage, MN 55378). This particular model had a 76 cm wand length. Modifications were made by soldering two #1 size metal paper clips to the prod's electrode prongs. The paper clips overlapped in a parallel manner with a spacing of approximately 2 mm apart and not touching the opposite prong (Fig. 1). Ideally, when the prod is energized, there will be no electrical arc until an ant walks between the paper clips. In practice, however, there is usually an intermittent arc as the prod charges and discharges.

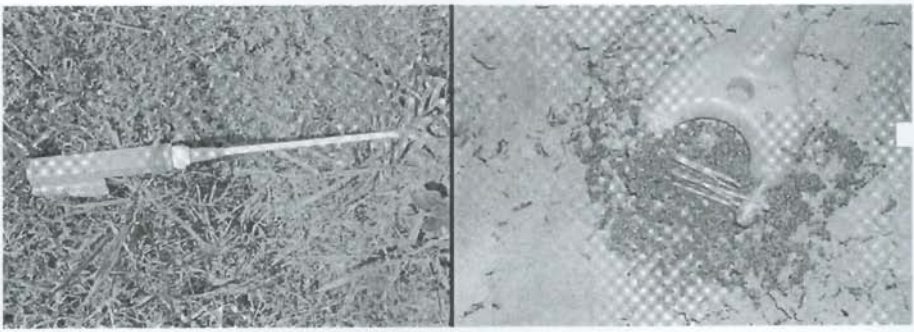

FIG.1. Modified electric livestock prod for phorid fly detection.

This test was conducted at the Five Eagle Ranch, located approximately five miles north-north-east of Caldwell, Texas. Two releases of *P. tricuspis* were made on the ranch in May 2002 and April 2003. Overwintered flies were first detected in April 2003 using this device. Subsequent evaluations showed there to be substantial numbers of flies in the area.

The test was conducted on the afternoon of 15 September 2003. Two active fire ant mounds were selected in the phorid-infested area to compare the effects of mechanical disturbance versus mechanical plus electrical disturbance. Efforts were made to select mounds of similar size and activity level in similar habitats. A total of 11 pairs of mounds were tested.

At approximately the same time, each of us would disturb one of a pair of mounds. One would be disturbed using mechanical means (usually a boot or stick) followed by the crushing of a number of ants between the fingers. The other mound was less vigorously disturbed by mechanical means, then the electric prod tip was placed on the mound and energized intermittently until the first phorid fly appeared. That mound was then observed for five minutes and the total number of flies counted. If no flies appeared on a mound, time recording was stopped after five minutes. Stopwatches were used for timing. Data were taken on the time of first fly appearance and maximum number of flies for each mound. The procedure was then repeated on another pair of mounds and continued until a total of 11 pairs were observed. Mound pairs were located in different habitats: closed canopy woods (2 pairs), edge of woods or near large trees (3 pairs), open pasture with nearby herbaceous vegetation ranging in height from approximately 15 cm to over 2 meters (6 pairs). Distance between mounds in a pair ranged from approximately seven meters to over 30 meters.

Data were analyzed using SAS PROC MEANS procedures with paired observations. Data were also analyzed using PROC TTEST with Cochran and Cox Approximation.

RESULTS AND DISCUSSION

The mean time of first fly observation for electrically stimulated mounds was 108 (± 66) seconds versus 271 (± 88) seconds for mechanically disturbed mounds. Probability for the paired analysis was $P = 0.0027$; unpaired t-test, $P = 0.0001$, (df = 20). Phorid flies were observed over all 11 electrically stimulated mounds, but only three mechanically stimulated mounds within five minutes of disturbance. Time to first observation of the electrical stimulation ranged from 47 to 240 seconds. Time to first observation of the mechanical disturbance ranged from 61 to 300 seconds, but only on the three mounds.

The mean number of flies per electrically stimulated mound was 5.7 (± 2.05) and 0.55 (± 1.04) flies for mechanically disturbed mounds. Probability for the paired analysis was $P = 0.0001$; unpaired t-test, $P = 0.0001$, (df = 20). Results are summarized in Table 1.

TABLE 1. Comparison of Electrical vs Mechanical Stimulation of Red Imported Fire Ant Mounds for the Detection of *Pseudacteon tricupsis*. Mound pairs = 11. (15 Sept., 2003, Burleson County, Texas).

Parameter	Electrical	Mechanical
Mean time to first observation (sec.)	108 ± 6	271 ± 88
Mean number of flies per mound (5 min.)	5.7 ± 2.05	0.55 ± 1.04
Mounds with flies (max. time 340 seconds)	11 (100%)	3 (27%)
Max/min. number flies on any mound	9 / 2	3 / 0

Results indicate that the modified electric livestock prod was highly effective at increasing the number of phorid flies observed over fire ant mounds, and it greatly reduced the amount of time required to observe the first fly. Perhaps most importantly, flies were observed on 100% of the mounds where electrical stimulation was used, versus 27% where only mechanical disturbance was used. The livestock prod has also been used during *P. tricuspis* release procedures with observations indicating increased fly retention over mounds and increased oviposition attacks.

The prod itself can be used in different ways. One is to allow only a few ants on the tip and electrocute them almost individually. Another way is to lay the prod tip on the mound and electrocute ants constantly until an arc can no longer be seen or heard. Our more commonly used method, the one used in this experiment, is to lay the tip on the mound and intemittently electrocute ants. Ants quickly form a large cluster around the paperclips. Such behavior is typical of damage caused by red imported fire ants invading electrical equipment (Slowik et al. 1996, MacKay et al. 1992). Flies are often seen attacking ants in close proximity to this cluster and even on the plastic part of the tip itself. Activation of the prod also seems to re-stimulate fly attack after their initial rush of attacks subsides.

Though there are undoubtedly ways in which the existing device can be improved, the model tested was inexpensive (approximately $60), quickly modified, rugged, and is easily carried in vehicles and by personnel. It appears to greatly enhance the chances of success when attempting to detect phorid flies in the field and the device has already been adopted by researchers in other states.

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NEW AND NOTABLE RECORDS OF ODONATA FROM TEXAS

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ABSTRACT

A dramatic increase in interest in the North American Odonata (dragonfly and damselfly) fauna in the last few years has lead to many new discoveries, particularly in southern areas where subtropical species seem to be expanding their range northward. I report the occurrence of eight Odonata species previously unknown from Texas: *Argia oenea* Hagen *in* Selys [Coenagrionidae], *Enallagma antennatum* (Say) [Coenagrionidae], *Leptobasis melinogaster* Gonzáles-Soriano [Coenagrionidae], *Aeshna persephone* Donnelly [Aeshnidae], *Anax concolor* Bauer [Aeshnidae], *Phyllocycla breviphylla* Belle [Gomphidae], *Erythemis attala* (Selys) [Libellulidae], and *Erythemis mithroides* (Brauer) [Libellulidae]. These discoveries include four species previously unknown from the United States and the first occurrence of the genus *Leptobasis* in the country. Additionally, I discuss recent records of several other species rarely reported from Texas.

INTRODUCTION

The Odonata fauna of Texas has been documented sporadically by several authors (Tinkham 1934; Ferguson 1942; Gloyd 1958; Johnson 1972; Donnelly 1978; Abbott 1996, 2001; Abbott and Stewart 1998; Abbott et al. 1997, 2003) over the last century. The advancing northern ranges of a number of species have been attributed to global temperature increases (Paulson 1998, Krotzer 1999, Behrstock 2000). Recently, however, there has been a surge in the number of avocational odonatologists which has lead to many field observations and the discovery of many species previously unknown from Texas and the United States. This surge in interest started in 2000 with Sid Dunkle's field guide, *Dragonflies through Binoculars*. This and the recent *Dragonflies and Damselflies of Texas and the South-central United* States (Abbott 2005) have made the group accessible to a much larger audience that is now regularly contributing to the field of odonatology. As a result, gaps in our knowledge of distribution, habitat utilization, life history and flight periodicity of Texas Odonata are also being filled at a rapid rate.

MATERIALS AND METHODS

Voucher specimens of all species, except *Anax concolor* and *Erythemis mithroides*, are deposited in the Brackenridge Field Laboratory Insect Collection at the University of Texas at Austin. Photographic vouchers of both species above are on file in the same collection.

RESULTS AND DISCUSSION

This surge in interest over the last few years culminated in an unprecedented number of new discoveries for Texas in 2004 and 2005 (Table 1). Eight species were confirmed in Texas for the first time, bringing the total documented species for this state to 223; at least 25 species more than any other state. Additionally, the known ranges of several other species were dramatically extended.

TABLE 1. New and Unusual Records of Odonata for Texas and the United States in 2004 and 2005.

Species	New to the U.S.	New to Texas	Major Range Extension	Breeding Population[a]	Unusual Record(s)
Acanthagrion quadratum				x	
Argia leonorae					x
Argia oenea		x			
Enallagma antennatum		x			
Leptobasis melinogaster	x	x		x	
Aeshna persephone		x			
Anax conclor	x	x			
Coryphaeschna adnexa					x
Rhionaeschna psilus			x		
Phyllocycla breviphylla	x	x		x	
Erythemis attala		x		x	
Erythemis mithroides	x	x			
Tholymis citrina					x

[a]First report of a breeding population in Texas.

Acanthagrion quadratum Selys [Coenagrionidae]: Mexican Wedgetail

This species was previously only known from Cameron and Val Verde Counties (Abbott 2001). These records represented historical and infrequent collections and a breeding population was unknown in Texas. It had only been documented as occurring in Texas during the first two weeks of May (Abbott 2001). A well-established breeding population is now known to occur on Borregos Creek of the King Ranch (Kleberg Co.). Its seasonal distribution has also been extended to 27 October. This is a common tropical species that extends south to Nicaragua. It inhabits slow reaches of intermittent streams. Within the United States its distribution is limited to Texas. Though these new records do not represent a northern increase in their distribution, it does represent the first confirmed breeding population of this species in the United States.

Argia leonorae Garrision [Coenagrionidae]: Leonora's Dancer

This species has been known since 1928, but was only recently described (Garrison 1994). At the time of its description, it was only known from four widely distributed counties in Texas and Nuevo Leon, Mexico. Abbott (2001) increased the distribution to nine counties in Texas. Garrison (1994) stated that "further collecting will show it to occur in southeastern New Mexico and in other areas of southern and central Texas." On 27 May 2003 I took a single male of this species at Rattlesnake springs in Carlsbad Caverns National Park (Eddy Co.; 4.2 km W of US 180 on Co. Rd. 418; N32.1083° W104.4677°) representing the first report of this species in New Mexico. Numerous additional populations of this species have been found in Texas during the last 3 years, bringing its known distribution in the state to 14 counties. Recently discovered populations in Burnet, Kerr, Hudspeth, Terrell, and Travis counties represent new records for the state. Despite the widespread distribution of this species, it appears to have fairly narrow habitat

requirements consisting of side streams and back channel areas with slow-moving or standing waters and a thick coverage of short reeds along the margins and emergent in the water.

Argia oenea Hagen *in* Selys [Coenagrionidae]: Fiery-eyed Dancer

A population of this species was discovered for the first time in Texas at Chinati Hot Springs (N30.0381° W104.5999°) in Presidio County by R. Tizard on 1 May 2004. A well-established population was seen along the run-off from the hot springs. This is a tropical *Argia* that extends from Panama northward to six counties in Arizona and now into Texas. On 15 September 2004, G. Lasley and K. Bryan, visited the same location and photographed and collected a single male. It is one of two *Argia*'s found in North America where the males have bright red eyes and a metallic copper thorax. Both a blue and a violet form of this species are known. The individuals collected at Chinati Hot Springs were violet. The violaceous forms are thought to be associated with xeric habitats (Garrison 1994).

Enallagma antennatum (Say) [Coenagrionidae]: Rainbow Bluet

G. Lasley found this species on 1 September 2004 along Palo Duro Creek in southern Hansford County at the bridge crossing of Texas FM 520 (N36.1248° W101.4654°; elevation 951 m). This is approximately 29 km SW of Spearman and represents the first record of this species in Texas, though it is known from just over the border in Cimarron County, Oklahoma. This colorful damselfly is widely distributed from the Great Plains north and east to Quebec. This species was found along a largely dry creek, with occasional pools and grassy banks along the water's edge. It generally inhabits slow, muddy-bottomed streams, ponds and lakes.

Leptobasis melinogaster Gonzáles [Coenagrionidae]: Cream-tipped Swampdamsel

On 19 June 2004, T. Langschied and J. Sinclair discovered a population of this Mexican species on the King Ranch (Kleberg County). It represents a new species and genus for the United States. This species was only recently described from Mexico, by Enrique Gonzáles based on two males collected in Jalisco and Oaxaca states (Gonzáles, 2002). This is a particularly interesting damselfly unlike other members of the genus *Leptobasis*. Gonzáles reported, "When I examined the first male of this species from Jalisco state, I hesitated assigning it to any known genus of Coenagrionidae because the specimen has some unusual characters." Breeding populations have been discovered both at the King Ranch and at Santa Ana National Wildlife Refuge. Individuals at Santa Ana NWR are restricted to the vegetation hanging 3-7 ft above the ground, over a small running water source. At King Ranch they are restricted to canopy covered areas of Santa Gertrudis Creek, a permanent, slow-moving creek. I collected a single female at Santa Ana NWR and will be presenting a full description in another publication. Larvae remain undescribed.

Aeshna persephone Donnelly [Aeshnidae]: Persephone's Darner

On 27 September 2004, J. Lasswell and J. Brady observed and collected this species flying along Calamity Creek as it was approaching dark. A single specimen was seen, representing the first record of its presence in Texas. The specific locality was on the Woodward Ranch, 20 miles south of Alpine in Brewster County. This species seems to be expanding its range. In the original description, it was listed as confined to Arizona (Donnelly 1961). In 2000, Dunkle reported the eastern limits of its range as the extreme western edge of New Mexico. Since then it has been found in southern Utah (Abbott 2002), southeastern New Mexico (Catron County), and now 450 km further east in the

Texas Trans Pecos. Despite its perceived range extension, this species, which inhabits mountain streams, is never common. This also represents the first indication that it flies at dusk.

Anax concolor Brauer [Aeshnidae]: Blue-spotted Comet Darner

Dennis Paulson observed two individuals of this species on 5 June 2005 at Santa Ana National Wildlife Refuge. This species is very similar to *A. longipes* Hagen which has also been observed in the Lower Rio Grande Valley. *Anax concolor* was considered a synonym of *A. longipes*, but Geijskes (1968) and Peters (1988) have separately presented evidence to support their distinctness. No specimen was collected, but a single male was photographed, confirming this species' presence in Texas and the United States. See Paulson (2005) for additional details.

Coryphaeschna adnexa (Hagen) [Aeshnidae]: Blue-faced Darner

Abbott (2001) reported this species from Cameron and Hidalgo Counties stating that it was unclear whether a breeding population existed in the state or if these records represented strays. A well-established, thriving population of this species has been discovered further north in Kleberg County on the King Ranch. It is known from southern Florida and the Greater Antilles, Mexico and south to Argentina. This species may be extending its range northward. It occurs all year in southern Florida (Dunkle 2000) and is known from June through 27 October in Texas.

Rhionaeschna psilus (Calvert) [Aeshnidae]: Turquoise-tipped Darner

Abbott (2001) reported this species from Cameron and Comal Counties. Since then, this uncommon Texas species has been found in Starr County and extended its known range further north into Travis County. It inhabits ponds, ditches and intermittent streams (Dunkle 2000) and has been found flying in Texas from 11 March to 16 November.

Phyllocycla breviphylla Belle [Gomphidae]: Ringed Forceptail

In 2002 a single teneral female gomphid was photographed at Santa Ana National Wildlife Refuge in Hidalgo County (Czaplack 2003). The species was thought to be *Phyllocycla breviphylla*, but a positive identification was not possible from the photograph. Subsequently, two females were collected on 29 May 2004 (Bocanegra and Czaplack 2004) at Anacua Wildlife Management Area in Cameron County. These captures represented the first confirmation of this species in the United States. Additional individuals were later observed (M. Reid and D. Dauphine, personal communication) at the Mission West RV Park in Mission (Hidalgo Co.) on 26 August 2004. I observed several individuals just outside Bentsen-Rio Grande Valley State Park on 21 June 2005. These observations suggest the establishment of a population of this species in the Rio Grande Valley. All of the individuals seen or collected are somewhat intermediate between *P. breviphylla* and the morphologically similar *P. elongata* in the appearance of the thoracic stripes. The presence of these intermediates raises the question of whether *P. breviphylla* is actually a synonym of *P. elongata.* The south Texas populations are found along small streams within the riparian forest.

Erythemis attala (Selys in Sagra) [Libellulidae]: Black Pondhawk

On 22 September 2004, T. Langschied observed *Erythemis attala* on Santa Gertrudis Creek at the King Ranch in Kleberg County. This species was observed up to 27 October 2004. Within the United States, this species was only known as a stray to Alabama. Its normal range includes the Antilles and Mexico south to Brazil. Indications are that there is an apparent breeding population at this south Texas locality. This species is unusually wary, presenting a great challenge to the collector. It usually inhabits ponds,

swamps, and slow streams. The newly discovered habitat in Texas is a canopy covered slow-moving stream.

Erythemis mithroides (Brauer) [Libellulidae]: Claret Pondhawk

A single male of this species was photographed at Santa Ana National Wildlife Refuge (Hidalgo Co.) on 1 May 2004 by M. Reid. It was seen along a narrow trail deep within thick wood-scrub, with a low, closed canopy, approximately 100 m from water (M. Reid, personal communication). This represents the first record for this species in the United States, and thus far it has not been seen again. *Erythemis mithroides* is tropical, ranging southward to Brazil and Paraguay. There is evidence that the Mexican populations of this taxon may actually represent an undescribed species (D. Paulson, personal communication).

Tholymis citrina Hagen [Libellulidae]: Evening Skimmer

Abbott (2001) reported this species from Hidalgo County based on the historical 1950 collection of a specimen now housed in the Florida State Collection of Arthropods. On 22 October 2003 T. Langschied found a single tattered male on the Borregos Creek within the Santa Gertruis Division of the King Ranch (Kleberg County). This species has only been reported in the United States on three previous occasions and certainly represents a rare find. This species is crepuscular, possibly aiding to its inconspicuousness, and was found flying late in the day. It is widely distributed in the tropics extending south into Chile. It has the tendency to hang vertically on twigs in densely wooded areas when not patrolling at dusk.

CONCLUSIONS

All of these finds indicate the tremendous opportunity for discovery in the Odonata, especially in southern states like Texas. In a reasonably well-known group like Odonata, it is exceptional that in such a short period of time, eight species were discovered for the first time in Texas and that four represent new country records. The geographic location and size of Texas are in part responsible for its diverse odonate fauna. Within the political boundaries of the state, there is a mixing of eastern and western faunas and subtropical and temperate faunas. This has resulted in the most diverse fauna of any state, with 223 species currently known from Texas. I believe some of these species are certainly expanding their ranges, but I attribute most of these discoveries to the growing number of people now studying dragonflies and damselflies.

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RELEASE AND ESTABLISHMENT OF *MACROCENTRUS PROLIFICUS* (HYMENOPTERA: BRACONIDAE), A PARASITOID OF SUGARCANE STALKBORERS (LEPIDOPTERA: CRAMBIDAE), IN NORTHWESTERN MEXICO

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ABSTRACT

Field studies were conducted in the sugarcane area of Los Mochis, Sinaloa, Mexico, from December 1994 to December 2001 to determine the establishment and impact of the introduced braconid *Macrocentrus prolificus* Wharton on the crambid stalkborers *Diatraea considerata* Heinrich, *D. grandiosella* Dyar, and *Eoreuma loftini* (Dyar). A total of ca. 200,000 *M. prolificus* adults were liberated in each of five, 10-ha fields during May-December 1995. Fields were sampled monthly for stalkborer larvae and *M. prolificus* parasitism from 1995 to 2001. Total stalkborer larvae averaged 2.9 per stalk during the period of study, with *D. considerata*, *D. grandiosella*, and *E. loftini* comprising 78.1, 21.2, and 0.7% of the total, respectively. Recovery of *M. prolificus* from field-collected stalkborer larvae began soon after initial releases, with average parasitism <3% during 1995-1997, increasing to 6% in 1998, and stabilizing at 10-12% during 1999-2001. Average (all-years) parasitism was highest for *D. considerata* and lowest for *E. loftini*.

INTRODUCTION

Sugarcane is grown on nearly 14,000 ha in Los Mochis area of northern Sinaloa, Mexico, a region where the sugar industry is nearly a century old. By far, the most important sugarcane insect pests are the crambid stalkborers *Diatraea considerata* Heinrich, *D. grandiosella* Dyar, and *Eoreuma loftini* (Dyar). Damage to sugarcane, scored as average bored internodes at harvest, during the last decade has ranged from 20 to 30%, representing annual losses up to six million U.S. dollars (Vejar 2003). Most damage is attributed to *D. considerata*, the most abundant and destructive of the three stalkborer species in this area (Rodriguez-del-Bosque and Smith 1997).

The stalkborers *E. loftini* and *D. grandiosella* are apparently indigenous to the Pacific Mexican coast (van Zwaluwenburg 1926). *D. considerata* was introduced into the Los Mochis sugarcane circa 1950 in infested seed cane originating from sugarcane produced in the Culiacan area, 220 km south of Los Mochis (Box 1951, Abarca et al. 1958). Similarly, *D. considerata* occurrence in Culiacan is probably a result of earlier seed cane movements from areas farther south in Nayarit, Colima, Jalisco, or Michoacan states

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(Smith and Rodríguez-del-Bosque 1994). This recent and progressive, northern movement of *D. considerata* has apparently occurred without a concomitant range extension of its natural enemies; thus, opportunities exist for classical biological control (Smith and Rodríguez-del-Bosque 1994, Rodríguez-del-Bosque and Smith 1997).

The braconid *Macrocentrus prolificus* Wharton is a polyembryonic, gregarious, larval endoparasitoid of *Diatraea* spp. and *E. loftini* (Wharton 1984, Smith et al. 1993). After being attracted by chemical cues provided by host frass or plant damage, *M. prolificus* attacks early-instar hosts within the leaf sheath, using a "probe-and-sting" strategy (Smith et al. 1993). Progeny emerge from later-instar hosts tunneling in the plant stalk (Smith et al. 1993). *M. prolificus*, originally described from individuals emerging from *D. considerata* near Culiacan (Wharton 1984), is present in several locations of western Mexico, but absent farther north of Culiacan (Melton et al. 1986, Rodríguez-del-Bosque and Smith 1989). Systematic field collections of stalkborers in the Los Mochis sugarcane area during the 1980s to the mid-1990s failed to yield either *Diatraea* spp. or *E. loftini* larvae parasitized by *M. prolificus* when hosts were reared to adult in the laboratory (A. Caro, unpublished data).

The objective of this study was to periodically release *M. prolificus* into the Los Mochis sugarcane area in selected fields, and routinely field collect stalkborer larvae to determine its establishment and estimate its impact. This investigation was part of an area-wide IPM program against stalkborers in this region implemented in 1990, when chemical insecticide applications were discontinued for the entire Los Mochis sugarcane area (Pérez and Hernández 1994). Without insecticidal pressure, extant native enemies were expected to reestablish and exert their natural mortality, and introduced biocontrol agents were believed to a high probability of colonization and establishment.

MATERIALS AND METHODS

On 1 December 1994, 196 larvae of *D. considerata* and 155 larvae of *D. grandiosella* were collected from sugarcane near Ingenio Costa Rica in Sinaloa, 253 km south of Los Mochis, an area where *M. prolificus* had been previously reported (Melton et al. 1986). Larvae were placed singly in 6-cm diameter plastic petri dishes containing 1 cm^3 of an artificial diet (Badilla et al. 1994), transported to Los Mochis, and reared under laboratory conditions (27 ± 1°C; 70% RH). Larvae were inspected for parasitism every 24 h, and provided with 1 cm^3 of fresh diet every 2-3 d. A foundation culture of *M. prolificus* was initiated with a single cocoon mass (98 females, 4 males), emerging from a *D. grandiosella* larva, 28 d after field collection. Subsequently, *M. prolificus* was mass reared on *D. saccharalis* (F.) in the laboratory at Ingenio Los Mochis from May to August 1995 according to procedures described by Smith et al. (1993).

Cocoons of *M. prolificus* were harvested from parasitized larvae every 24 h, and placed in groups of 17 in 150-ml plastic cups covered with organza secured with a rubber band, and held in the laboratory for emergence. About 2-3 d before emergence, two vertical lines of a 1:1 mixture of honey and distilled water as a food source were applied inside the cups with a fine brush. Nearly 1,000 adults emerged in each cup, 25-30 d after cocoon were harvested.

M. prolificus was released monthly during May-August 1995 in five, 10-ha sugarcane fields (6-8 month old), separated by a minimum of 7-km, near Los Mochis. Each month, approximately 50,000 *M. prolificus* adults were liberated in each field. Adult parasitoids were released early in the morning with most adults emerging the previous night. Thus, in each field, a total of ca. 200,000 *M. prolificus* adults were released during the 4-month period. All fields were planted with the cultivar NCO-310, and were surrounded by other commercial sugarcane fields. No insecticide was applied throughout

the study, and other cultural practices were made in all fields accordingly to local recommendations (Sánchez 1990).

Each field was sampled monthly for stalkborer larvae before (January-April 1995) and after parasitoid liberations (May 1995-December 2001). In each field and on each sampling date, 50 stalks were randomly selected within the center 5 ha of the field, excised, transported to the laboratory, and dissected for stalkborer larvae. Larvae were identified based on pigmentation pattern differences among the three species (Flores and Abarca 1961). Larvae were reared on artificial diet and parasitization determined by parasitoid cocoon formation. Adults of *M. prolificus* emerging from cocoons obtained from field parasitized larvae were recycled into the laboratory culture for subsequent field release.

For each sampling date, percentage parasitism was estimated for each stalkborer species by dividing the number of parasitized larvae by the total number of collected larvae, and multiplying by 100. Means of larvae per stalk were compared among stalkborer species by LSD ($P<0.05$). Percentages of parasitism by *M. prolificus* were compared among stalkborer species by chi-square tests, $P<0.05$ (SAS Institute 1999).

RESULTS AND DISCUSSION

Stalkborer larval infestation averaged a total of 2.93 per stalk throughout the study, ranging from 2.26 in 1995 (May-December) to 4.02 in 2000 (Table 1). Stalkborer densities increased progressively from 1995 to 2000, then declined in 2001. Overall (1995-2001), *D. considerata*, *D. grandiosella*, and *E. loftini* comprised 78.1, 21.2, and 0.7% of total larvae collected, respectively. These data confirmed that *D. considerata* remains the predominant sugarcane stalkborer in Los Mochis (Pérez and Hernández 1994, Rodríguez-del-Bosque and Smith 1997). The low numbers of *E. loftini* collected in the field may represent an underestimation of actual densities as a result of its tunneling behavior, which is not as conspicuous as that of *Diatraea* species (Rodríguez-del-Bosque et al. 1988, Smith et al. 1993).

TABLE 1. Stalkborer Density and Parasitism by *M. prolificus* on *D. considerata*, *D. grandiosella*, and *E. loftini* on Sugarcane. Averages from Five Fields near Los Mochis, Sinaloa, Mexico. 1995-2001.

	Larvae (Ave±SE)/stalk [a,b]				% Parasitism [a,c]			
Year	*D. con.*	*D. gra.*	*E. lof.*	Total	*D. con.*	*D. gra.*	*E. lof.*	Total
1995 [d]	1.92±0.20 a	0.50±0.05 b	0.03±.002 c	2.45	0.0 a	0.0 a	0.0 a	0.0
1995 [e]	1.67±0.17 a	0.56±0.06 b	0.03±.003 c	2.26	3.2 a	2.1 a	0.0 a	2.9
1996	1.71±0.10 a	0.60±0.03 b	0.02±.001 c	2.33	2.0 a	1.3 a	0.0 a	1.8
1997	1.98±0.10 a	0.52±0.03 b	0.03±.002 c	2.53	2.1 a	1.9 a	0.0 a	2.0
1998	2.94±0.14 a	0.71±0.03 b	0.02±.001 c	3.67	6.4 a	4.8 b	3.0 b	6.1
1999	2.98±0.17 a	0.86±0.05 b	0.01±.001 c	3.85	12.9 a	7.4 b	0.0 c	11.6
2000	3.28±0.19 a	0.72±0.04 b	0.02±.001 c	4.02	10.4 a	11.7 a	1.0 b	10.8
2001	1.80±0.09 a	0.47±0.02 b	0.02±.001 c	2.29	12.9 a	9.5 b	14.7 a	12.2
Ave.	2.29 a	0.62 b	0.02 c	2.93	8.1 a	6.1 b	3.2 c	7.6

[a] *D. con.* = *D. considerata*; *D. gra.*= *D. grandiosella*; *E. lof.*= *E. loftini.*

[b] Averages of stalkborer larvae within rows followed by the same letter are not significantly different (LSD, $P<0.05$).

[c] Percentage parasitism within rows followed by the same letter are not significantly different (chi-square, $P<0.05$).

[d] January-April, before liberations initiated.

[e] May-December. After liberations initiated in May-August.

Seasonal abundance was significantly different between the two most common stalkborers (Fig. 1). *D. considerata* was abundant all year, with peak densities in February-March. In contrast, *D. grandiosella* was most abundant in November-December, when densities were similar to those of *D. considerata*, then decreased progressively during January-March to become scarce during April-July, and increased again in August.

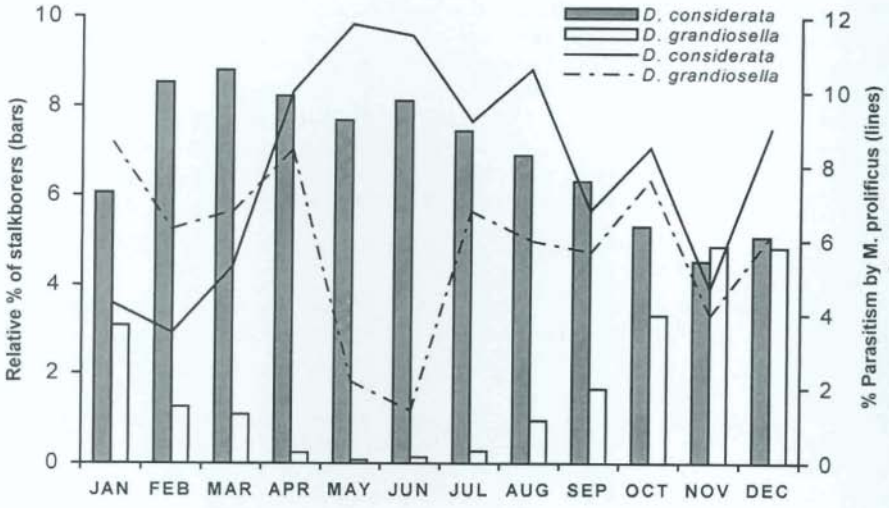

FIG. 1. Seasonal abundance of two sugarcane stalkborers (bars) and their respective parasitism by *M. prolificus* (lines); pool data (1995-2001); Los Mochis, Sinaloa, Mexico.

Recovery of *M. prolificus* from field collected stalkborer larvae began soon after first releases, with total parasitism <3% during 1995-1997, increasing to 6% by 1998, and stabilizing at 10-12% during 1999-2001. This suggests a permanent establishment of the parasitoid in this region (Table 1). A maximum of 52% parasitism of *D. considerata* by *M. prolificus* was observed in one field during August 1998. Average (all years) parasitism was highest on *D. considerata*, and lowest on *E. loftini*. However, parasitism on *E. loftini* during 2001 reached a level not significantly different from that of *D. considerata* (Table 1).

Regardless of stalkborer species, *M. prolificus* was able to locate and successfully parasitize host larvae all year. However, peak parasitism on *D. considerata* occurred in May-June, when conversely, parasitism on *D. grandiosella* was the lowest (Fig. 1). *M. prolificus* appears to have acted in a density-dependent manner (Huffaker et al. 1974) parasitizing each of the two most common stalkborers, according to their respective seasonal abundance. The ability of *M. prolificus* to find and successfully parasitize stalkborers all year increased its propensity to successfully colonize and establish in the Los Mochis region. *M. prolificus* was also released and established in southern Texas sugarcane during the 1980's. However, although the parasitoid is recovered consistently from field collections, parasitism levels are usually low (<1%) on *E. loftini* and *D. saccharalis* (F.) (Meagher et al. 1998). Conversely, *M. prolificus* was not able to establish on *D.*

grandiosella in northern Texas maize, possibly because of harsh winters (Overholt and Smith 1990).

Although we did not measure the ability of *M. prolificus* to disperse, recent findings show the parasitoid is now widely distributed in the sugarcane area of Los Mochis. The parasitoid has been released at multiple sites other than those studied here (Vejar 2003). In conclusion, the present study showed *M. prolificus* has become an additional mortality factor of sugarcane stalkborers in this region, and may contribute to reductions in their population levels.

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PARASITOIDES Y PARASITISMO NATURAL DEL PICUDO DEL CHILE[1] *ANTHONOMUS EUGENII* EN EL NORTE DE SINALOA, MÉXICO

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RESUMEN

Se identificaron en el norte de Sinaloa los parasitoides relacionados directamente con el picudo del chile *Anthonomus eugenii* Cano y se definió el parasitismo natural que ejercen sobre él a través del desarrollo del cultivo. El trabajo se llevó a cabo en parcelas comerciales y experimentales de chile jalapeño durante dos temporadas, realizando semanalmente recolección de frutos infestados con la plaga y mediante golpes de red entomológica. En los lotes experimentales no se aplicaron insecticidas, con el fin de fomentar el incremento de poblaciones parasitoides. Los parasitoides se colectaron mediante técnicas de confinamiento y disección de frutos, y red entomológica. La mayoría de los ejemplares se obtuvieron de lotes experimentales, a partir del mes de marzo y principalmente durante abril, hasta que las temperaturas se incrementaron en primavera. En las parcelas comerciales, únicamente se encontraron dos ejemplares, posiblemente debido al uso de insecticidas. Se identificaron en total cinco especies de parasitoides relacionados directamente con el picudo del chile: *Catolaccus hunteri* Crawford, *Eupelmus* sp. *Urosigalphus* sp. *Bracon* sp., y *Eurytoma* sp. *C. hunteri* fue el parasitoide más abundante, aunque el máximo porcentaje de parasitismo que ocasionó fue 2%. A partir de los resultados obtenidos se pueden realizar estudios orientados a desarrollar un programa para el control biológico de *A. eugenii*.

ABSTRACT

The parasitoids of the pepper weevil *Anthonomous eugenii* Cano were identified in north Sinaloa from samples taken during two planting seasons, 2001/2002 and 2002/2003, in commercial and experimental plots of Jalapeño pepper. Crop production practices were typical of the region, with the exception that insecticides were not used in the experimental farms to promote the presence of the natural enemies. Weekly samples were taken of infested fruit, along with sweep net samples; parasitoids were obtained by confinement and dissecting fruit, along with identification of specimens captured with insect nets. Most of the specimens were obtained from the experimental plots beginning March and peaking in April, with confined fruits. Practically no parasitoids were obtained from the commercial farms, probably due to excessive application of insecticide sprays, with 9 applications in the 2001/2002 season, and 13.5 in the 2002/2003 season. Five parasitoid species directly associated with *A. eugenii* were identified: *Catolaccus hunteri* Crawford, *Eupelmus* sp. *Urosigalphus* sp. *Bracon* sp., and *Eurytoma* sp. The first of these, *C. hunteri,* was the most common and abundant

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species, though, the highest parasitism rate for this species was 2%. The information obtained in this study is useful for future development of biological control tactics against *A. eugenii* in north Sinaloa.

INTRODUCCION

El picudo del chile es la plaga más importante de este cultivo porque ataca a todos los tipos de chile y puede dañar el 100% de las fructificaciones, por su amplia distribución, y porque origina la mayor cantidad de gastos dentro del rubro fitosanitario (Burque y Wodruff, 1980; Bolaños y Aranda, 1991; Bujanos, 1993). El método de control más utilizado, y prácticamente el único dirigido intencionalmente para combatir a éste insecto plaga, es el uso de insecticidas, que puede exceder de 15 aplicaciones por temporada. Lo anterior, incrementa los costos del cultivo, afecta la salud del hombre, provoca contaminación ambiental, resistencia a los insecticidas, eliminación de enemigos naturales, y otros problemas ya bien conocidos (CATIE, 1993).

El empleo de enemigos naturales, en conjunto con otras prácticas de manejo, representa una alternativa para el control del picudo del chile (Mariscal et al., 1998) que permitiría reducir el número de aplicaciones de insecticidas, y con ello disminuir también los efectos indeseables ocasionados por su empleo. Antes, es indispensable realizar la búsqueda de sus enemigos naturales con fines de identificación. Lo anterior es importante para i) establecer la identidad de los parasitoides presentes, ii) definir el estatus de dichos agentes de control en espacio, tiempo y cantidad, y iii) definir la relación de candidatos potenciales a introducir y/o reproducir en un programa de liberación. El objetivo general de este trabajo fue contribuir al manejo integrado de *A. eugenii* a través de la identificación de los parasitoides que lo atacan en la región norte de Sinaloa y determinar el porcentaje de parasitismo que proveen en forma natural a través del desarrollo del cultivo.

MATERIAL Y METODOS

Durante los ciclos agrícolas otoño-invierno 2001/2002 y 2002/2003, se establecieron dos parcelas experimentales, una en el terreno agrícola del Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional (CIIDIR), del Instituto Politécnico Nacional, en Guasave, Sinaloa, y otra en el Campo Experimental del Valle del Fuerte (CEVAF), del Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP), en Juan José Ríos, Sin. En ambas no se aplicaron insecticidas, para favorecer la presencia de parasitoides, mientras que el resto del manejo agronómico fue similar al de las parcelas comerciales. Las parcelas experimentales se establecieron en lugares aislados de otras áreas de cultivo, para evitar arrastre de insecticida, y ocuparon una superficie de 2000 m^2 cada una.

En la temporada 2001/2002, se seleccionaron cuatro lotes comerciales para realizar el estudio, buscando que fueran representativos del área de producción de chile en la región. En estos lotes se realizaron las prácticas generales que realizan los productores de chile en el norte de Sinaloa, incluyendo las aplicaciones de insecticidas. Las fechas de transplante fueron entre el 2 de septiembre y el 15 de octubre del 2001, en la primera temporada, y entre el 25 de agosto y el 11 de octubre de 2002, en la segunda temporada. En la primera temporada las cuatro parcelas comerciales se ubicaron en el municipio de Guasave, Sin., y en la segunda se seleccionaron tres lotes próximos a Guasave y un cuarto en el municipio de Ahome, Sin., al norte del CEVAF. La superficie cultivada varió de seis a 10 ha.

Las aspersiones de insecticidas en los lotes comerciales se realizaron de manera calendarizada. En el ciclo agrícola 2001/2002, se ejecutaron un promedio de nueve aspersiones de insecticidas, mientras que en el 2002/2003 el promedio fue de 13.5

aspersiones. En la mayoría de los casos los insecticidas aplicados se mezclaron con otros agroquímicos como fungicidas, nutrientes foliares, y antibióticos, entre otros.

Muestreo de frutos con síntomas de daño por picudo del chile. En el período comprendido entre los meses de diciembre del 2001 a abril del 2002, y de diciembre del 2002 a mayo del 2003, se recolectaron frutos infestados por el picudo del chile en muestreos semanales. La recolección se realizó en cinco sitios de cada lote, un sitio en cada esquina de la parcela y uno más en el centro; en cada punto se recolectaron al azar cuarenta frutos tiernos, ya sea de la planta ó caídos en el suelo, todos con síntomas de infestación por el picudo; en total se recolectaban 200 frutos por lote por semana. Después, los frutos se trasladaban al laboratorio de entomología del CEVAF, en bolsas de papel estraza. Se realizaron un total de 16 muestreos para cada lote durante el ciclo agrícola 2001/2002, y 22 muestreos para cada lote en el ciclo 2002/2003.

Confinamiento de frutos infestados con A. eugenii. Una vez en el laboratorio, se limpiaron y secaron los frutos con un pedazo de tela, con el fin de evitar pudriciones y eliminar externamente insectos u otros organismos que pudieran estar presentes. Después, se colocaron en 20 vasos desechables de plástico transparente de 260 ml en grupos de cinco frutos por vaso y se cubrieron con tela organza asegurada por una liga en la parte superior del vaso (Hunsberger y Peña, 1997; Mariscal et al., 1998). Los vasos con las muestras se mantuvieron en observación a temperatura y humedad ambiente, esperando un tiempo razonable, alrededor de 15 días, para que los insectos emergidos murieran y se facilitara su conteo.

Disección de frutos infestados con A. eugenii. Este muestreo se realizó en el segundo año de estudio, con el propósito de complementar los resultados del muestreo en frutos confinados. De cada muestra semanal por lote, la mitad de los frutos recolectados por lote (100 frutos) fueron disectados, cortándolos longitudinalmente con una navaja delgada y filosa, luego con la ayuda de un microscopio estereoscopio, aguja de disección y pinzas entomológicas se determinó la presencia o ausencia de estados inmaduros, así como de adultos de parasitoide dentro de los frutos. Una vez que estos eran localizados, se obtuvieron fotografías de cada uno de ellos, para después cerrar los frutos y depositarlos en vasos transparentes hasta la emergencia de los parasitoides adultos.

Muestreo de parasitoides con red entomológica. Durante el ciclo 2002/2003, además del muestreo en frutos infestados con picudo, se incluyó el muestreo con red entomológica. Este consistió en 100 golpes de red por semana en cada uno de los lotes, en los cinco sitios por lote. La muestra obtenida se introdujo en frascos con alcohol al 70%, y se transportó al laboratorio para su revisión bajo el microscopio. En el laboratorio se separaron las diferentes especies, las cuales fueron introducidas en frascos viales con alcohol al 70%. Este muestreo se hizo para cotejar la identidad y el número de los parasitoides que se podrían encontrar parasitando estados inmaduros del picudo del chile, con los adultos obtenidos mediante esta técnica de muestreo.

Preparación e identificación de los parasitoides obtenidos. En la temporada 2001/2002, los especímenes de los parasitoides se obtuvieron vaciando en una caja de Petri el contenido de cada vaso (frutos de chile, picudos y parasitoides), y con la ayuda de un microscopio estereoscopio se separaron, y se registraron los resultados. Los parasitoides obtenidos fueron depositados en cápsulas de gelatina, debidamente etiquetadas. Posteriormente, se mantuvieron bajo refrigeración hasta ser enviadas para su identificación a un taxónomo especialista. Para la preparación de los parasitoides del ciclo agrícola 2002/2003, se siguió el mismo procedimiento, con diferencia de que aquí los ejemplares se colocaron en frascos viales de 50 ml con alcohol al 70%.

Los ejemplares obtenidos fueron identificados por taxónomos especialistas en himenóptera parasítica (Alejandro González Hernández, Facultad de Ciencias Biológicas, Universidad Autónoma de Nuevo León, San Nicolás de los Garza, N. L., México; José Antonio García Sánchez, Instituto de Fitosanidad, Colegio de Postgraduados, Montecillo, Estado de México, México; Marco Antonio Reyes Rosas, Campo Experimental Río Bravo (INIFAP), Río Bravo,

Tamaulipas, México; utilizando las claves taxonómicas de Wharton *et al.* (1997) y Gibson *et al.* (1997). En el laboratorio del CEVAF se conservan fotografías y muestras de las especies de parasitoides obtenidos.

Determinación del porcentaje de parasitismo. Los datos empleados para la determinación del porcentaje de parasitismo se obtuvieron de la relación de adultos de picudos del chile y de parasitoides emergidos del total de frutos confinados por vaso. Aparte, se emplearon los datos obtenidos de la disección de frutos. En ambos casos, éste se calculó dividiendo el número de parasitoides emergidos entre la cantidad de parasitoides emergidos más el número de picudos emergidos, y multiplicado por 100, según metodología usada por Hunsberger y Peña (1997).

RESULTADOS Y DISCUSIÓN

Parasitoides obtenidos de frutos confinados. En los lotes experimentales del CIIDIR y del CEVAF se obtuvieron 70 ejemplares de parasitoides asociados a *A. eugenii* en el primer ciclo, y 104 en el segundo. Estos pertenecen a dos superfamilias, en cuatro familias de Hymenoptera. Únicamente se identificó a especie a *Catolaccus hunteri* Crawford (Pteromalidae), el cual fue el más frecuente y abundante con 65 especímenes en el primer ciclo (93%) y 90 en el segundo (87%). Los demás géneros identificados fueron: *Eupelmus* (Eupelmidae), *Urosigalphus* (Braconidae), *Bracon* (Braconidae) y *Eurytoma* (Eurytomidae), en números muy reducidos (Cuadro 1). Especímenes de estos géneros no se identificaron a especie. Los parasitoides se detectaron durante los meses de marzo a abril en el primer ciclo, y de febrero a mayo en el segundo ciclo.

CUADRO 1. Parasitoides de picudo del chile obtenidos de frutos confinados en dos ciclos agrícolas en el norte de Sinaloa. TABLE 1. Pepper weevil parasitoids obtained from confined fruits in two agricultural seasons in north Sinaloa.

Superfamilia	Familia	Género	Especie	No.	Frutos*
		Temporada 2001/2002			
Chalcidoidea	Pteromalidae	*Catolaccus*	*Catolaccus hunterii*	65	1600
	Eupelmidae	*Eupelmus*	*Eupelmus*	1	1600
	Eurytomidae	*Eurytoma*	*Eurytoma*	2	1600
Ichneumonoidea	Braconidae	*Urosigalphus*	*Urosigalphus*	1	1600
	Braconidae	*Bracon*	*Bracon*	1	1600
		Temporada 2002/2003			
Chalcidoidea	Pteromalidae	*Catolaccus*	*C. Hunteri*	90	2200
	Eupelmidae	*Eupelmus*	*Eupelmus*	4	2200
	Eurytomidae	*Eutytoma*	*Eurytoma*	10	2200

*Total de frutos confinados de los que se obtuvieron los especímenes parasitoides reportados.

Sólo en un lote comercial (León Fonseca) se obtuvieron ejemplares parasitoides: uno de *C. hunterii* y el otro de *Eupelmus* sp., en el mes de abril de 2003. Cabe señalar, que para la fecha en que se obtuvieron los parasitoides, la parcela ya había sido abandonada por el bajo valor de la producción; la última aplicación de plaguicidas se realizó tres meses y medio antes de la detección de los enemigos naturales.

Parasitoides obtenidos mediante la disección de frutos. Todos los parasitoides detectados mediante esta técnica provinieron de frutos recolectados en lotes experimentales en los meses de abril a mayo del 2003 y correspondieron a *C. hunteri* (Cuadro 2) no se obtuvo especímen alguno mediante ésta técnica en los lotes comerciales.

Parasitoides obtenidos mediante el empleo de red entomológica. Se obtuvieron 28 ejemplares de parasitoides del picudo en los lotes experimentales, entre febrero y mayo de

2003, y todos correspondieron a *C. hunterii*. En los lotes comerciales no se obtuvo ejemplar alguno (Cuadro 3).

La familia Pteromalidae fue la mejor representada de las cuatro familias detectadas. Con relación a esto Borror *et al.* (1989) señalan que los miembros de dicha familia son parasitoides que atacan una amplia variedad de hospederos, y muchas especies son muy importantes en el control biológico de plagas insectiles de varios ordenes, incluyendo Coleoptera, al cual pertenece *A. eugenii*. Wilson (1986), reportó a *C. hunteri* parasitando larvas de picudo del chile en Florida, en el interior de las flores de chile tipo bell pepper. Posteriormente Hunsberger y Peña (1997), lo reportaron parasitando a *Anthonomus macromalus* Gyllenhal, en acerola *Malpighia glabra* L., en el sur de Florida. El mismo *C. hunteri* ataca al picudo del algodón, con una efectividad similar a *C. grandis*, el parasitoide más exitoso en el control biológico de esa plaga (Cortez et al., 2001). Por otra parte, Mariscal *et al.* (1998), reportaron a *C. hunteri* en el estado de Nayarit causando un 2.9% de parasitismo sobre *A. eugenii*. Wilson (1986), lo reportó en el estado de Florida causando 5% de parasitismo sobre la misma plaga. No obstante el parasitismo tan reducido que ocasionó en todos los casos, éste parasitoide podría ser un candidato efectivo y un importante agente de control biológico del picudo del chile (Wilson, 1986; Mariscal et al., 1998).

CUADRO 2. Parasitoides obtenidos en frutos disectados en laboratorio. Temporada 2002/2003. TABLE 2. Parasitoids obtained from dissected fruits in laboratory. Season 2002/2003.

Lote	Fecha	Frutos disectados	No. de parasitoides
CIIDIR	10-Abr-03	100	6
	23-Abr-03	100	8
	30-Abr-03	100	6
	05-May-03	100	11
	13-May-03	100	4
CEVAF	10-Abr-03	100	4

CUADRO 3. Especímenes de *Catolaccus hunteri* obtenidos durante la temporada 2002/2003, mediante el empleo de red entomológica en chile, en el norte de Sinaloa. TABLE 3. Specimens of *Catolaccus hunteri* obtained during the season 2002/2003, in sweep net samples in pepper, in north Sinaloa.

Lote	No. de muestreos	No. de parasitoides
CIIDIR	13	17
CEVAF	15	11

Eurytoma sp., fue la segunda especie en importancia, considerando su abundancia, aunque el número de individuos encontrados fue reducido; sólo se obtuvieron 12 especímenes en los dos ciclos agrícolas. Cross y Chesnut (1971) reportan algunos miembros de esta familia como parasitoides de curculiónidos, entre ellos citan a *Eurytoma herrerae* (Ashmead) parasitando al picudo del algodón, así como *E. pini* Bugbee que parásita entre otros al picudo del algodón *A. grandis* y *A. vestitus* Boheman (CABI, 2000). Pérez (1985), encontró en el estado de Morelos a *Eurytoma* sp., parasitando larvas del primer ínstar del picudo del ejote *Apion aurichalceum* Wagner. Además, lo reporta como un hiperparasitoide de *Zatropis* sp. (Hymenoptera: Pteromalidae). CABI (2000) reporta a *Eurytoma goidanichi* Bugbee, *E. pini* y *E. braconidis* Ferriére, como hiperparasitoides de otros himenópteros.

Eupelmus sp., fue el tercero en frecuencia de aparición, con sólo cinco ejemplares. Mariscal *et al.* (1998) encontraron en Nayarit nueve adultos de *Eupelmus* sp., relacionados

directamente con *A. eugenii.* Rodríguez y Reyes (2003), en un trabajo desarrollado en Río Bravo, Tamaulipas, encontraron solamente cuatro parasitoides de este género. Cross y Chesnut (1971) reportan a *Eupelmus cushmani* Crawford y a *E. cyaniceps* Ashmead, como parasítoides del picudo del algodón. Además, señalan a *E. cyaniceps* como posible hiperparasitoide de *Bracon mellitor* Say. Por su parte CABI (2000), reporta a *E. cushmani* como parasitoide de *A. grandis* y de *A. eugenii* igual que *C. hunteri.*

Se obtuvieron dos ejemplares de la familia Braconidae, de los géneros *Urosigalphus* y *Bracon.* Mariscal *et al.* (1998) encontraron en Nayarit, únicamente tres hembras y dos machos de *Urosigalphus* sp. atacando al picudo del chile. A este género se le ha encontrado parasitando a otros curculiónidos. Charlet y Seiler (1994), reportaron a *U. femoratus* atacando al picudo del girasol *Smicronyx fulvus* Le Conte. En el estado de Texas, se crió la especie *Urosigalphus schwarzi* Gibson, para controlar biológicamente al picudo del algodón, pero sin resultados favorables (Cate et al., 1990).

En cuanto a *Bracon,* Rodríguez y Reyes (2003) reportan a éste género parasitando a *A. eugenii* en el estado de Tamaulipas, y Mariscal *et al.* (1998) recolectaron sólo nueve ejemplares en Nayarit a partir de *A. eugenii.* La especie *Bracon mellitor* Say es un parasitoide importante del picudo del chile y del picudo del algodón (Cross y Chesnut, 1971; CABI, 2000). *Bracon vesticida* Vierek se reporta como parasitoide de *A. eugenii* en E.U.A., y de *A. vestitus* en Perú (CABI, 2000). Por su parte, Riley y King (1994) mencionan que se intentó realizar control biológico clásico del picudo del chile con *B. vesticida* en el sur de California, pero no se documentó éxito.

Porcentaje de parasitismo del picudo del chile en frutos confinados. El porcentaje de parasitismo en los lotes comerciales durante ambos ciclos agrícolas fue prácticamente nulo, pues únicamente se detectaron dos especímenes de parasitoides en el lote comercial de León Fonseca, en el segundo ciclo agrícola. La mayoría de los parasitoides se obtuvieron en los lotes experimentales del CEVAF y del CIIDIR (Cuadro 4), pero el parasitismo fue reducido. El parasitoide que presentó el mayor porcentaje de parasitismo fue *C. hunteri* aunque apenas rebasó el 1.0 y el 2.0%, en las dos temporadas: estos números están cercanos al porcentaje de parasitismo natural del mismo pteromálido sobre el picudo de la acerola *A. macromalus,* 1.0% (Hunsberger y Peña, 1997).

CUADRO 4. Porcentaje de parasitismo de picudo del chile en frutos confinados, durante dos temporadas de siembra, en el norte de Sinaloa. TABLE 4. Percent parasitism of pepper weevil in confined fruits, during two seasons, in north Sinaloa.

Lote	No de Picudos	Parasitoides	% de parasitismo
	Temporada 2001/2002		
CIIDIR	3186	42 *C. hunteri*	1.30
		1 *Urosigalphus*	0.03
		1 *Bracon*	0.03
		1 *Eupelmus*	0.03
CEVAF	2750	24 *C. Hunteri*	0.86
	Temporada 2002/2003		
CEVAF	1834	46 *C. hunteri*	2.40
		2 *Eupelmus*	0.10
		6 *Eurytoma*	0.32
CIIDIR	1980	43 *C. hunteri*	2.10
		1 *Eupelmus*	0.05
		4 *Eurytoma*	0.20
León Fonseca	1965	1 *C. Hunteri*	0.05
		1 *Eupelmus*	0.05

El parasitismo total en frutos confinados en la temporada 2001/2002 fue de 2.3%, mientras que en el 2002/2003, fue de 5.3%. Todas las especies de parasitoides arrojaron en la segunda temporada mayor parasitismo respecto a la primera, pero fueron menos especies las detectadas (Cuadro 4).

Porcentaje de parasitismo en frutos de chile disectados. En ninguno de los lotes comerciales se detectó parasitismo con ésta técnica; sólo se detectó en los lotes experimentales. Las primeras larvas parasitoides se detectaron el 10 de abril de 2003, todas de *C. hunteri* (Cuadro 5). El porcentaje de parasitismo fue bajo, parecido al obtenido en los frutos confinados, y muy diferente al reportado por Mariscal *et al.* (1998), quienes en un trabajo similar desarrollado en el estado de Nayarit, encontraron nueve especies de parasitoides asociados con *A. eugenii*, en donde *Triaspis* sp., el cual no se encontró en éste estudio, ocasionó un 29.2% de parasitismo natural sobre estados inmaduros del picudo. Esto posiblemente se deba a que en el lugar donde se desarrolló el estudio de Mariscal *et al.*, existen condiciones ambientales diferentes a las del norte de Sinaloa, y que favorecen mayor presencia y actividad de los parasitoides del picudo del chile.

CUADRO 5. Porcentaje de parasitismo obtenido en frutos disectados de chile, durante la temporada 2002-2003, en el norte de Sinaloa. TABLE 5. Percentage parasitism obtained in dissected pepper fruits, during the season 2002/2003, in north Sinaloa.

Lote	Picudos	Parasitoides	No.	% de parasitismo
CEVAF	1834	*Catolaccus*	28	1.5
CIIDIR	1980	*Catolaccus*	35	1.7

En el primer ciclo agrícola, se obtuvieron menos parasitoides que en el segundo. Esto pudo deberse a que en el segundo se incrementó el número de muestreos durante cuatro semanas.

Aunque los muestreos para detectar parasitoides se realizaron a partir de diciembre en las dos temporadas, la presencia de los enemigos naturales ocurrió hasta la primera semana de marzo en la temporada 2002/2003, y en la última semana de febrero en la temporada 2003/2004. En ambos casos, la obtención de parasitoides fue mayor en abril (Cuadro 6). Esto pudo deberse al efecto de la temperatura, ya que es notorio que los enemigos naturales del picudo del chile encontrados en este estudio, se presentaron en primavera, cuando la cantidad de horas con temperaturas altas por día aumentaron (datos no presentados). Se desconocen reportes sobre el impacto de bajas temperaturas en el desarrollo de los parasitoides encontrados, sin embargo, Greenberg *et al.* (1996) señalan que *Catolaccus grandis* (Burks), parasitoide de picudo del algodón, incrementó el rango de mortalidad 2.4 veces y disminuyó el número de hembras emergidas de larvas de picudo parasitadas en 78.2% cuando se redujo de 25 °C a 15 °C la temperatura. Además, mencionan que a temperaturas por debajo de 20 °C se redujo la sobrevivencia de 1.8 a 9.4 veces comparada con un testigo mantenido a 25 °C.

Es posible que la presencia nula de parasitoides en las parcelas comerciales de la región norte de Sinaloa esté relacionada con el efecto que ocasionan los plaguicidas sobre ellos, pues se sabe que los insectos entomófagos son más susceptibles a estos que los insectos fitófagos (Croft, 1990; citado por Badii et al., 2000). Esta aseveración también se apoya en lo señalado por Flint y Dreistadt (1998), quienes mencionan que los insecticidas oxamyl, malatión, azinfos metílico, metomilo, permetrina, carbaryl, dicofól, metamidofós, clorpirifos, y otros, tienen un impacto inmediato, y una larga residualidad sobre un gran número de grupos de enemigos naturales. El uso excesivo de los plaguicidas oxamyl, endosulfán y amitráz, tienen un efecto negativo en *C. grandis*, ya que cantidades pequeñas de estos insecticidas son suficientes para eliminarlo, afectando gravemente sus poblaciones (Escobar et al., 1999). Elzen *et al.* (1999), reportan que los plaguicidas fipronil, spinosad, tebufenozide, cyflutrin,

oxamyl, endosulfán, profenofós, azinfos metílico y malatión, fueron altamente tóxicos para *C. grandis*. Sumy *et al.* (1992), reportan que una aplicación del insecticida oxamyl, en dosis de 226 g de ingrediente activo/ha, causó un 100% de mortalidad de dicho parasitoide una hora después de su aspersión y tuvo un efecto residual de 16 a 20 días. De los plaguicidas señalados anteriormente, el amitráz es el insecticida que se usa menos en el norte de Sinaloa, mientras que el resto integran el paquete de insecticidas usados comúnmente contra plagas insectiles del cultivo de chile. Además, se debe considerar que en los lotes comerciales donde se realizó el estudio se efectuaron aplicaciones con estos productos, en la mayoría de los casos en mezcla o acompañados con algún otro agroquímico. Esto puede explicar, en parte, el número reducido de especies parasitoides encontradas, y porqué aún cuando las temperaturas favorecieron la presencia de los enemigos naturales, en los lotes comerciales con insecticidas no se obtuvieron parasitoides.

CUADRO 6. Número y porcentaje de parasitoides obtenidos por mes en frutos confinados, en las temporadas 2001/2002 y 2002/2003, en el norte de Sinaloa. TABLE 6. Number and percent of parasitoids obtained by month in confined fruits, season 2001/2002 and 2002/2003, in north Sinaloa.

Mes	No. de Parasitoides	% de Parasitoides
	Temporada 2001/2002*	
Diciembre	0.0	0.0
Enero	0.0	0.0
Febrero	0.0	0.0
Marzo	6.0	8.6
Abril	64.0	91.4
	Temporada 2001/2002**	
Diciembre	0.0	0.0
Enero	0.0	0.0
Febrero	2.0	1.9
Marzo	20.0	19.2
Abril	58.0	55.8
Mayo	24.0	23.1

* Muestreo en frutos confinados. ** Muestreo en frutos confinados, frutos disectados y golpes de red.

En resumen, es interesante señalar que a pesar de la diversidad y abundancia reducidas de parasitoides de *A. eugenii* en el norte de Sinaloa, los resultados obtenidos son importantes porque se definió cuales son los parasitoides asociados directamente con esta plaga en la región y su distribución temporal. Esta información servirá para el desarrollo de futuros programas de control biológico de *A. eugenii*. Una de las recomendaciones importantes que pueden desprenderse de los resultados obtenidos, es que *C. hunteri* debe ser considerado seriamente en esfuerzos futuros de control biológico del picudo del chile en el norte de Sinaloa. La introducción de otras especies como *Triaspis* sp., el parasitoide más abundante detectado en Nayarit, deberá ser examinada críticamente, ya que si no se encuentra de manera natural en ésta región es debido posiblemente a que no se adapta, y porque podría tener un efecto ecológico negativo para las especies nativas que se presentan en la región.

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PARASITISM OF *MELANIS PIXE* (LEPIDOPTERA: RIODINIDAE) ON GUAMUCHIL IN NORTHERN MEXICO

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The legume tree "guamuchil", *Pithecellobium dulce* (Roxb.) Benth, is native along the coastal areas from California through Mexico to South America, and has colonized the tropics worldwide (Allen and Allen 1981). Several species of Homoptera, Hemiptera, Lepidoptera and Coleoptera feed on guamuchil, although damage is commonly insignificant (Browne 1968, N.A.S. 1980). However, occasional outbreaks of *Melanis pixe* (Boisduval) (Lepidoptera: Riodinidae) in northeastern Mexico may result in severe defoliation of guamuchil (Rodríguez-del-Bosque 2005), which is probably its preferred host plant (Scott 1986, Neck 1996, DeVries 1997). The objective of this study was to determine the survival of *M. pixe* larvae and pupae on guamuchil in northern Tamaulipas, Mexico.

The study was conducted near Río Bravo, Tamaulipas, during May 2001 and December 2003, respectively when *M. pixe* outbreaks occurred. A total of 411 pupae in 2001 and 494 pupae in 2003 were collected from five guamuchil trees (4-6 m tall). A total of 140 late-instar larvae were collected only in 2003. The trees had been defoliated (visually estimated at 30-40%) during the previous 2-3 weeks, and larvae had ceased feeding and descended from the leafy twigs to the trunk and main branches to search for pupation sites. Larvae and pupae were placed singly in 30-ml plastic cups with plastic lids, transported to the laboratory (25°C, 70% RH), and observed every 24 h for 30 d for survival. Fate of collected individuals were classified as: (a) normal development (pupation or adult emergence), (b) parasitism, (c) infection by entomopathogens, and (d) desiccation.

Fate of collected larvae in 2003 was as follows: 72.9% pupated, 26.4% died due to desiccation, and 0.7% were infected by *Beauveria bassiana* (Balsamo) Vuillemin. No parasitism was observed in larvae. Survival of pupae was lower than larvae (Table 1).

TABLE 1. Survival of *Melanis pixe* pupae on guamuchil. Río Bravo, Tamaulipas. 2001 and 2003.

	May 2001		December 2003	
Mortality factor	n	%	n	%
Adult emergence	57	13.9	177	35.8
Desiccation	70	17.0	148	30.0
Beauveria bassiana	0	0	48	9.7
Brachymeria annulata	284	69.1	121	24.5
Total	411	100	494	100

The most important mortality factor of pupae was the chalcidid *Brachymeria annulata* (F.) (identified by G. Delvare, CIRAD, Montpellier, France) with parasitism ranging from 69.1% (2001) to 24.5% (2003). Desiccation of pupae ranged from 17% (2001) to 30% (2003). Infection of pupae by *B. basssiana* ranged from 0% (2001) to 9.7% (2003). The hyperparasite *Eupelmus* sp. (Hymenoptera: Eupelmidae) emerged from 1.8 and 8.0% of *B. annulata* in 2001 and 2003, respectively. Differences among mortality factors of pupae in 2001 and 2003 may be attributed to the different time of the year in which *M. pixe* outbreaks occurred. Yet, factors favoring such occasional outbreaks remain unknown. This study represents the first report of *M. pixe* survival near its northern distribution (Rodríguez-del-Bosque 2005).

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CRÍA DE *DIATRAEA SACCHARALIS* (F.) EN DIETA NO ESPECIFICA

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El barrenador del tallo de la caña de azúcar *Diatraea saccharalis* (F.) tiene una amplia distribución en el continente americano (Davidson, 1992). En México, es el insecto plaga más importante sobre la caña de azúcar (Johnson, 1984; Legaspi et al., 1999); sus larvas causan daño al cavar galerías dentro de los tallos, reduciendo el crecimiento y causando la muerte de plantas (Davidson, 1992). Las épocas de mayor incidencia son entre mayo y septiembre y dependiendo de la región donde se localice, se presentan 4 o 5 generaciones por año (Legaspi et al., 1997; Grupo Azucarero México, 1998).

Existen diversas dietas para la cría de este insecto a nivel de laboratorio (Pan y Long, 1961; Hensley y Hammond, 1968; Mihm, 1984). Sin embargo, algunos de los ingredientes utilizados son difíciles de encontrar en México y las dietas comerciales tienen un costo elevado. Por este motivo, el objetivo de este trabajo fue evaluar una dieta para *Trichoplusia ni* (Hübner) (Shorey, 1963) modificada para la cría de *D. saccharalis*.

Una colonia de *D. saccharalis* fue iniciada con 80 pupas proporcionadas por USDA-ARS, Weslaco, Texas. Estas pupas se mantuvieron con un fotoperíodo de 14:10 h (luz:oscuridad), 65% de HR y 26°C. Al emerger los adultos se transfirieron manualmente a cubetas de plástico recubiertas en el interior con polietileno transparente y tapadas con gasa. Los adultos se alimentaron utilizando algodón impregnado con una solución de sacarosa al 15% contenida en un recipiente completamente cubierto con polietileno transparente y perforado en la parte superior. Los huevecillos fueron recolectados manualmente cada 24 h y lavados con agua corriente durante 3 min, se dejaron secar y se incubaron en recipientes de plástico de 100 ml con 50 ml de dieta fresca.

La dieta de Shorey (1963) se modificó incrementado la cantidad de azúcar de 13g a 25g y el agar-agar de 14g a 20g. El procedimiento de preparación no fue modificado. La dieta preparada se vació en recipientes de plástico transparente de 100 ml y al solidificar se adicionaron de 10 a 15 larvas y se dejaron en incubación. Diez días después se cambiaron a dieta nueva donde permanecieron hasta la pupación. Las pupas se colectaron manualmente, se desinfectaron con hipoclorito de sodio al 2.5% por 2 min y se colocaron nuevamente en las cubetas

El ciclo completo de *D. saccharalis* en laboratorio utilizando la dieta modificada fue de 33 días (rango = 28 a 39 días) en promedio en cada una de cinco generaciones. Los tiempos de desarrollo fueron: huevecillo, 5 días (rango = 4 a 6 días); larva, 21 días (rango = 19 a 24 días); pupa, 5 días (rango = 4 a 6 días) y adulto, 2 días (rango = 1 a 3 días). El peso promedio de las pupas macho fue 0.072 ± 0.013 g (n = 16) en la primera generación y 0.076

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± 0.017 g (n = 228) en la quinta generación (P = 0.01, gl = 242, t = 5.38); los pesos promedio correspondientes de las pupas hembra fueron 0.118 ± 0.012 g (n = 10) y 0.124 ± 0.029 g (n = 261) (P = 0.01, gl = 269, t = 26.88). El pequeño incremento en peso de las pupas macho y hembra sugiere que la dieta fue adecuada para los insectos.

Los resultados sugieren que la dieta modificada y las condiciones de humedad, temperatura y fotoperíodo empleados, resultaron adecuados para el desarrollo de *D. saccharalis*. Los pesos de las pupas obtenidos con la dieta modificada fueron mayores que aquellos reportados por Pan y Long (1961) (pupas macho, 0.0607 g y pupas hembra, 0.1044 g) utilizando una dieta específica para *D. saccharalis*. Además se observa que a nivel poblacional la dieta les permite un desarrollo adecuado. La práctica del recubrimiento con polietileno resultó de gran utilidad para la recolección de huevecillos ya que las hembras tienen una marcada tendencia a ovipositar en el lugar de alimentación (Mihm, 1984). La transferencia de las larvas a dieta fresca a la mitad del ciclo de desarrollo evitó la contaminación por hongos y bacterias y el aletargamiento de las larvas, que sucede cuando la dieta se deshidrata. El tiempo de generación de la cría resultó menor al reportado (43 días) por Legaspi et al. (1997). El acortamiento del ciclo biológico permitió la obtención de una generación por mes y representó una ventaja para la obtención de larvas comparado con las poblaciones naturales que se presentan 4 ó 5 veces al año (Legaspi et al., 1997). Actualmente, a 4 años del establecimiento de la cría es posible contar con aproximadamente 10,000 larvas cada mes.

En conclusión, el establecimiento a nivel de laboratorio de la cría de *D. saccharalis* fue exitoso. La modificación realizada a la dieta de *T. ni* produce larvas sanas y robustas para la realización de bioensayos a nivel de laboratorio, y reduce el costo de mantenimiento de la colonia.

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TABANUS ABACTOR[1] PHILIP RESPONSES TO CHAINING AND BURNING REDBERRY JUNIPER STANDS

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ABSTRACT

Ground-level chaining and elevated chaining followed by fire were evaluated as methods to eliminate stands of redberry junipers (*Juniperus pinchotii* Sudw.) which provide a habitat for *Tabanus abactor* Philip. Adult captures on bucket and board traps and larval numbers in the soil were significantly influenced by year. When fly abundance was high in 1997, 1999, and 2002, numbers of fly captures were lower in both chaining treatments compared to captures in the untreated check. The five-year, post-treatment average indicated that ground-level chaining coupled with burning significantly ($P \leq 0.05$) reduced the numbers of adult females captured by 67% compared with numbers captured in the untreated check. The site was dominated by mature juniper trees pre-treatment, and canopy cover was 32%. Chaining rates were not significantly different for the two chaining methods. In 2003, the juniper canopy cover percentage in the untreated check (50.8%) was significantly greater than that of the chain and burn treatments which were not different from each other (5.4% avg.). Juniper mortality decreased significantly over time, and there was no indication that elevated chaining plus burning increased mortality over ground-level chaining and burning. The addition of the fire treatment four years after chaining did not increase juniper mortality.

INTRODUCTION

Tabanus abactor Philip, known colloquially as the "cedar fly", has been considered one of the most damaging livestock pests in the Rolling Plains of Texas (Davis and Sanders 1981) due to their painful bites. Research has shown a potential monetary loss from weight gain reduction in heifers exposed to tabanids for 84 days of over $10.00 per head (Perich et al. 1986). The distribution of *T. abactor* includes 46 of 62 counties of the Rolling Plains (Davis and Sanders 1981) as well as other parts of Texas, Oklahoma, Kansas, and Arkansas (Schomberg and Howell 1955). Insecticide control strategies are generally not practical under field conditions (Leprince et al. 1992).

Montandon et al. (1993) found *T. abactor* larvae in the soil under leaf litter of large redberry junipers (*Juniperus pinchotii* Sudw.), but not under mesquite (*Prosopis glandulosa* Torr. var. *glandulosa*). An effective integrated pest management strategy is to modify or eliminate winter habitats of pest insects. For example, Wiedemann et al. (1979) used a low-energy grubber to selectively remove tree rows in a shelterbelt, and Slosser et al. (1984) demonstrated that this technique significantly reduced winter survival and altered spring emergence patterns of the boll weevil (*Anthonomus grandis* Boheman). Slosser et al. (1985) used tebuthiuron herbicide to kill sand shinnery oak (*Quercus havardii* Rybd.), thus

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owners were forced to re-introduce grazing because the prolonged drought had reduced available forage, and they needed the land returned to production.

Treatments were (1) untreated check, (2) ground-level chaining followed by fire four years later, and (3) elevated chaining followed by fire four years later. Chaining was conducted in early March of 1997, one week following at least 7.8 mm of rainfall at both locations. A spherical ball, 1.2-m diameter, was fabricated from 13-mm steel plate. A 102-mm diameter axle through the center of the ball allowed the ball to rotate as the unit was pulled by chains attached to each end of the axle (Wiedemann 2004). Two crawler tractors, 123 kW and 149 kW, pulled 54 m of 52-mm diameter anchor chain (12.4 kg $link^{-1}$ or 58.4 kg m^{-1}) for standard, ground-level chaining, after which the ball was attached midway in the chain for elevated chaining at a tree striking height of 0.6 m. Chaining time was measured with a stopwatch for each plot, and the rate in hectares/hour was calculated based on time and measured plot area. Clock time was recorded for equipment servicing and transportation.

Prescribed burning was conducted in February and March 2001, four years after the chaining treatments following guidelines by White and Hanselka (1991). Plots were burned as individual units with a total of eight plots burned (four at each location). At the Halsell site, air temperatures, relative humidity (RH) and wind speed just prior to the fires averaged 18.1°C, 44.8% and 4.3 m s^{-1}, respectively. At the Johnson site, air temperatures, RH and wind speed just prior to the fires averaged 19.0°C, 35.3% and 5.2 m s^{-1}, respectively. The average herbaceous fine fuel just prior to burning was 1,117 kg ha^{-1} for the standard-chain treatment and 864 kg ha^{-1} for the elevated chain treatment. These amounts were less than the minimum of 2,000 kg ha^{-1} that is recommended to generate sufficient fire intensity to suppress junipers (Wright and Bailey 1982). As a result, intensity of all fires, as estimated by visual observation of flame heights, was considered low to moderate (flame heights 1-4 m). Large patches of bare ground disrupted flame fronts and most plots had to be re-ignited numerous times. Approximately 50-70% of the surface area of each plot was blackened by the fires.

Two types of sticky traps with a 0.2-0.4 cm thick coat of polyisobutlene (Tangle-Trap® stickum, The Tanglefoot Co., Grand Rapids, MI 49504) were used to capture adult *T. abactor* (Moore et al. 1996, Slosser et al. 2000). One trap type consisted of a cylindrical white bucket (18.9-l capacity), with top removed, suspended upside down over a T-post. A 19-cm piece of plastic pipe (4.4 cm I.D.), with two 23.5-cm long x 0.5-cm diameter wire rods welded at right angles and inserted through the pipe near one end, was placed over the top of the T-post. The inside bottom of the bucket rested on top of the wire rods which kept the bucket centered over the top of the T-post. Bucket traps were 1 m above ground level. The second trap type consisted of a 1.3-cm thick piece of plywood cut into 46.4 x 46.4 cm squares. The boards, painted brown before coating with stickum, were placed on the ground with brown side up. The white bucket traps were not painted. Moore et al. (1996) reported that white bucket traps capture females almost exclusively (99.5%), while brown board traps placed on the ground capture mostly males (79.1%).

The two types of traps were placed adjacent to each other near the center of each plot. Traps were inspected once-a-week from early June through mid-September, 1996-1999. However, flies were not sampled in 2000 because it appeared that the drought conditions that had persisted since 1998 would prevent the burning portion of the study. After burning was completed successfully, fly trapping was resumed in 2001 and 2002. After counting, the flies and other large insects were removed with forceps from the buckets and boards. The stickum was removed with a large spatula 2-3 times each summer, and a fresh coat of stickum was applied.

All flies in the untreated, ground-level chaining, and elevated chaining treatments were removed from the board traps on 21 and 28 June 2001. The 268 flies were sexed to

eliminating this overwintering habitat of the boll weevil. Habitat modification is an effective means of disrupting the life cycle and reducing populations of the horn fly (*Haematobia irritans* Linnaeus) (March and Bay 1983).

Junipers (*Juniperus* spp.) have encroached upon 8.8 million hectares of rangeland in Texas and 1.4 million hectares in Oklahoma (SCS 1983, SCS 1988, Engle 1985). Ansley et al. (1995) estimated that redberry junipers in northwest Texas have increased their distribution by 61% from 1948 to 1982. This rangeland is basic to the livestock, wildlife, and recreation industries of both states. Herbaceous plant production has been doubled and even tripled by juniper removal (Arnold 1964, Steuter and Wright 1983). Prescribed burning for control of juniper has been validated by Wright and Bailey (1982) in Texas and Rollins (1985) in Oklahoma. The mortality of Ashe junipers (*Juniperus ashei* Buchholz) and eastern redcedars (*Juniperus virginiana* L.) achieved by burning is high when trees are less than 2 m in height, but fire is seldom effective when they are over 2.4-m tall (Dalrymple 1969, Martin and Crosby 1955, Owensby et al. 1973). Mortality of redberry juniper, a sprouting species, is high (70%) when trees are less than 1-m tall and the bud zone is still above ground (Steuter and Britton 1983). When the bud zone is below ground, trees resprout following fire.

Moderate to dense stands of juniper must be mechanically treated to release herbage production necessary for successful burns (Rasmussen et al. 1986). Chaining is the felling of trees by an anchor chain pulled between large crawler tractors (Fisher et al. 1973). Wiedemann and Cross (1996) determined that an elevated chaining technique could reduce pulling requirements of individual trees by 84% in redberry junipers and 67% in Ashe junipers while maintaining efficacy similar to ground-level chaining, the standard practice. Tests showed that 0.6-m height would give the best chaining results in redberry junipers (Wiedemann and Cross 1996). The elevated chain will partly uproot redberry juniper trees exposing the bud zone. The potential is present to increase plant mortality by using fire to destroy the exposed bud zone. An elevated chain is also less likely to damage soil crusts or grasses or spread unwanted species, such as prickly pear cactus (*Opuntia* spp.), than is a chain pulled on the ground.

Our research is based on the hypothesis that mechanical removal of junipers, followed by prescribed fires, would destroy the larval habitat of *T. abactor* and break the fly's life cycle without the use of insecticides or herbicides. We also hypothesized that elevated chaining followed by fire would induce more mortality in redberry junipers than would standard chaining. The research objectives were to quantify the (1) rate of chaining (ha hr^{-1}) for elevated and standard chaining methods, (2) effect of chaining and prescribed fire on juniper reduction, and (3) reduction in *T. abactor* populations. Another paper reports the effects of these treatments on the herbaceous community (Ansley et al. 2005).

MATERIALS AND METHODS

Study sites occurred in the Rolling Plains ecological region of northwest Texas on the Johnson (33° 59' N, 99° 50' W) and Halsell (33° 50' N, 99° 48' W) ranches south of Crowell, TX. Soils at both sites were complexes of the Cottonwood (silt loam; thermic Lithic Ustorthents), Talpa (loam; thermic Lithic Calciustolls), and Knoco (clay loam; thermic shallow Aridic Ustorthents) series. Mean annual precipitation is 616 mm with most occurring between April and October (Climatological Data, NOAA, Asheville, N.C.).

The study used a randomized block design with three treatments per block and four replications, two blocks at two locations. Dense stands of redberry juniper occurred in each of the 12 plots. Plot size, ranging from 12-17 ha, were side by side at each location. Recording gauges at both locations measured each rainfall event of at least 0.25 mm, and data were extracted each month. The research area was fenced and livestock grazing was deferred from both sites from 1996 to 2001. Two months after the fires in 2001, the ranch

determine if chaining (1997) and fire (2001) altered the sex ratios of the flies that were active in the three treatments.

Larvae were sampled each year that traps were used to monitor adults. Sampling was conducted in early May of each year. A square frame, which enclosed an area measuring 0.25 m^2, was randomly placed on the litter surface under the canopy area of redberry juniper trees. Four samples were taken, each under a different tree, in every plot. The juniper leaf litter was removed and all soil to a depth of ca. 10 cm was excavated and carefully examined. Large soil clumps were broken apart and examined for larvae. After burning, juniper leaf litter was greatly reduced, and samples were taken near the base of remaining, uprooted juniper trees, especially in areas where a thin litter layer remained.

Pre-treatment samples were taken in all plots during the summer of 1996, and there were no significant differences among the designated chaining treatments in adult numbers captured on bucket and board traps or in larval numbers in the soil (Holmes 1998, Holmes et al. 1998). These pre-treatment samples indicated that *T. abactor* was evenly dispersed throughout the study areas on both ranches prior to chaining in the spring of 1997.

Juniper density, canopy cover, and average height were determined using the point-center-quarter method using 25 equally spaced points along a 300-m long diagonal line across each plot. The diagonal lines were marked at each end and a compass azimuth was used to maintain direction. Juniper apparent mortality was measured after the first growing season, eight months following chaining. At least 100 trees were rated dead or alive within a belt-transect along a diagonal line across each chained plot. The tree was rated alive if any sprouts were present on roots remaining where a tree had been or if a tree was not completely uprooted and green tissue was present.

Herbaceous vegetation was sampled in November-December of each year along two parallel 300-m-long lines in each plot. A 0.25-m^2 frame was placed at 20 equally spaced points per line, and percentage basal cover of each herbaceous species and percentage bare ground and leaf litter were visually estimated. Proximity of each frame to juniper canopy (either intact or downed from chaining) was noted and identified as interstitial space between canopies, canopy drip line or sub-canopy. Herbaceous standing crop for the entire frame was clipped to ground level and weighed. Because livestock grazing was introduced in 2001, six, 1 x 2 m cages were randomly placed in each plot along the original sample lines to continue sampling un-grazed vegetation in 2001-2003.

A randomized block analysis was used to determine main effects of different chaining techniques, chaining and burning, sites, and years on captured cedar larvae and adults. There were two replications at each of two locations (Halsell and Johnson ranches). A General Linear Model procedure (SAS System v8.1, SAS Institute, Inc., Cary, N.C. 27511) was used for analysis of variance with a protected Least Significant Difference method of means separation at P=0.05. The error term for sites, the error term for treatments, and the error term for years rather than the whole-model error term was used to test sites, treatments, and years for means separation. Contrasts for fly data were calculated for a pre-burn versus post-burn comparison, and chaining versus the check. The appropriate error term was used in each case. Contrasts for larval data were calculated for pre-treatment and post-treatment comparison, pre-treatment versus chaining, pre-treatment versus burning, and chaining versus burning. The same analysis as used for cedar flies was used for main effects of chaining on juniper canopies, densities, and height for pre- and post-treatment analysis. Selected treatment means were compared within years using Least Significant Means analysis. The male:female ratios of the flies in the three treatments were evaluated with χ^2.

RESULTS

Rainfall during the summer months of the study were compared to long-term (LT) averages in Table 1. May and September are typically peak rainfall months. May rainfall was well below the LT average in all years but 1999. September rainfall was below LT average in all years and locations except for the JJ Ranch in 1997. July rainfall was below normal in four of the six years. May, June, and July rainfall were below the LT average in 1997, 1998, and 2001. There were only two (1999, 2002) of the six years that the 5-month total was normal or above for both ranches.

TABLE 1. Rainfall Each Month of the Growing Season, Growing Season Totals, and Annual Totals at Johnson and Halsell Ranches and Long-Term (LT) Averages at Nearby Crowell, TX. (Climatological Data, NOAA, Asheville, N.C. 28807).

	Rainfall (mm)						
Year	May	Jun	Jul	Aug	Sep	Sub-total	Annual
				Johnson Ranch			
1997	56	66	21	116	99	358	689
1998	27	7	4	33	16	87	304
1999	155	147	20	26	12	360	610
2000	34	185	2	0	0	221	799
2001	51	10	6	71	36	174	423
2002	52	80	217	24	25	398	831
				Halsell Ranch			
1997	51	46	34	111	50	292	620
1998	47	25	7	69	0	148	405
1999	173	101	59	30	20	383	649
2000	39	88	10	0	0	137	609
2001	72	19	0	120	47	258	522
2002	52	85	253	53	18	461	939
LT Avg.	91	76	48	54	87	356	616

Adult captures on bucket and board sticky traps and larval numbers in the soil were not significantly affected by ranch location ($F = 13.31$; df = 1,2; $P = 0.07$ for buckets; $F = 7.48$; df = 1,2; $P = 0.11$ for boards; $F = 4.84$; df = 1,2; $P = 0.16$ for larvae). Numbers of adults captured on both sticky traps were significantly influenced by year (Table 2), but there was no indication of a decreasing trend in fly numbers over time. Numbers of larvae in soil samples were significantly lower from 1999 through 2002 compared to numbers in 1997 and 1998.

Numbers of adults captured on bucket traps were significantly influenced by treatment and by the year by treatment interaction (Table 3). During years of low fly

TABLE 2. Average Number (± SD) Adult and Larval *Tabanus abactor* Each Year Across All Treatments.

Year	Description	Adults[a] caught on:		Larvae in soil[b,c]
		Buckets[c]	Boards[c]	
1997	1st season post chaining	11.6 (7.0) a	3.4 (2.0) b	2.9 (2.0) a
1998	2nd season post chaining	0.9 (1.1) c	4.3 (5.0) b	2.3 (1.9) a
1999	3rd season post chaining	6.4 (5.7) b	2.9 (1.9) b	0.3 (0.9) b
2001	1st season post burning	2.8 (3.0) c	8.3 (7.8) a	0.3 (0.9) b
2002	2nd season post burning	7.8 (7.0) b	3.9 (4.0) b	0.3 (0.6) b

[a]Average number per month per treatment. Sampled June through September.
[b]Average number per m^2 per treatment.
[c]Means are compared among years, and values with a common letter are not significantly different (P>0.05).

TABLE 3. Average numbers (± SD) of adult and larval *Tabanus abactor* in two chaining treatments and the untreated check.

Year	Treatment[b]	Adults[a] caught on:		Larvae in soil[c,d]
		Buckets[c]	Boards[c]	
1997	Untreated	18.2 (6.8) a	4.7 (1.9) a	2.3 (1.3) a
	E-chain	10.8 (5.1) b	3.2 (2.5) a	2.8 (1.0) a
	GL-chain	5.8 (2.4) c	2.4 (1.3) a	3.8 (3.3) a
1998	Untreated	1.4 (1.8) a	7.2 (6.0) a	3.3 (2.1) a
	E-chain	0.7 (0.9) a	3.8 (6.0) a	2.3 (1.9) a
	GL-chain	0.5 (0.2) a	1.9 (1.2) a	1.3 (1.5) a
1999	Untreated	10.1 (7.2) a	3.9 (2.1) a	0.8 (1.5) a
	E-chain	5.5 (5.1) b	3.3 (2.0) a	0.0 (0.0) a
	GL-chain	3.6 (3.0) b	1.4 (1.2) a	0.3 (0.5 a
2001	Untreated	4.9 (3.5) a	10.0 (6.0) a	0.3 (0.5) a
	E-chain	2.4 (3.1) a	9.5 (12.0) a	0.0 (0.0) a
	GL-chain	1.2 (1.0) a	5.3 (4.9) a	0.8 (1.5) a
2002	Untreated	12.4 (8.8) a	5.6 (4.4) a	0.8 (1.0) a
	E-chain	6.7 (7.0) b	4.2 (5.2) a	0.0 (0.0) a
	GL-chain	4.2 (2.8) b	1.9 (1.5) a	0.0 (0.0) a
All	Untreated	9.4 (8.2) a	6.3 (4.5) a	1.5 (1.7) a
	E-chain	5.2 (5.5) ab	4.8 (6.3) a	1.0 (1.5) a
	GL-chain	3.1 (2.8) b	2.6 (2.6) a	1.2 (2.1) a

[a]Average number per month per treatment. Sampled June through September.
[b]E=Elevated chaining, GL=Ground-level chaining.
[c]Treatment means are compared within years and values with a common letter are not significantly different (P>0.05).
[d]Average number per m^2.

abundance as in 1998 and 2001, there were no differences in numbers of adults captured among chaining treatments. When fly abundance was high, as in 1997, 1999, and 2002, numbers of adults captured were lower in both ground-level and elevated chaining treatments compared with numbers captured in the untreated check. The five-year post treatment average indicated that ground-level chaining coupled with burning significantly reduced the numbers of adults captured compared with numbers captured in the untreated check. Numbers of adults captured in the elevated chaining plus fire treatment were intermediate. Numbers of adults captured on board traps and larval numbers in the soil were not significantly affected by treatment or by the year by treatment interaction (Table 3).

The ratios of male:female flies were equivalent in untreated, ground-level chaining, and elevated chaining treatments ($\chi^2 = 3.04$, df = 2, $P = 0.218$) on 21 and 28 June 2001. This indicates that the chaining and burning treatments did not alter the sex ratios of flies that were active in the cleared plots compared with the sex ratio in the untreated plots.

Pre-treatment juniper canopy cover, density, and height averaged 32%, 294 trees ha^{-1}, and 2.8 m, respectively, and were not different among designated treatments. Chaining rates for the two methods were not significantly different (P=0.32). Therefore the data were pooled and the average chaining rate was 9.2 ha hr^{-1}. Using the contractor's cost of $140 for the two tractors, the actual chaining cost was $15.15/ha (100% field efficiency). However, when downtime for tractor and equipment servicing and transportation of tractors and personnel to the site was included, costs increased to $26.65/ha (57% field efficiency). These were customary charges by the contractor, and our cost was in-line with chaining costs for the area. Canopy cover for the check increased from 30 to 51% from 1996 to 2003, an increase of 21% in seven growing years, or 3% per year (Table 4). Juniper apparent mortalities after chaining in 1997 were ca. 20% in both chain treatments. Juniper mortalities in 2003, two years after burning, were significantly lower than mortality measured in 1997 and again were not different between chain+burn treatments. By 2003, both treatments had reduced juniper canopy cover to <6% compared to 50% in the check (P<0.001), but there was no difference between the two chain and burn treatments. Tree densities were not different between treatments or between 1996 and 2003 (P>0.10) measurements. Site was not a factor in any of the parameters.

Herbaceous production initially was about 700 kg ha^{-1} in all treatments in 1996. Production did not increase in treated plots over the untreated control until 1999, three years after chaining. In 1999, the sites received average rainfall while 1998 rainfall was below normal (Table 1). Production declined in all treatments the first growing season following the 2001 fire treatments, but production increased in chained and burned plots to 2-3 times the amount in the untreated control the second and third year after fire. There was no difference in herbaceous production between ground-level and elevated chaining treatments. More details on herbaceous production and species diversity responses to these treatments are available in Ansley et al. (2005).

Fine (leaf) litter cover declined in all treatments from 1998 to 2001, probably due to drought (Fig. 1). Litter sharply increased in all treatments in 2003 in response to above normal rainfall. The chaining and burning treatments decreased litter cover compared to the untreated control, but differences were not significant until after the fire treatments were applied in 2001. There were no significant differences in fine litter between the two chain treatments throughout the study.

The yearly variation in numbers of flies captured on bucket traps was probably caused by climatic conditions rather than by erratic treatment suppression of fly numbers in the study areas. Slosser et al. (2000) reported that adult captures on sticky traps during the summer were enhanced by rainfall and suppressed by high temperatures. Fly activity was low in 1998 and 2001 which were years with below average rainfall during most of the

TABLE 4. Untreated Check, Ground-Level Chaining Plus Fire (GLC+F) and Elevated Chaining Plus Fire (EC+F) Influence on Various Juniper Parameters. Values Reported as Means (± SD).

Year	N	Treatments			P value
		Untreated	GLC+F	EC+F	
			Tree canopy cover, %		
1996	12	30.3 (10.0) a	34.3 (14.7) a	30.3 (3.6) a[a]	0.57[b]
2003	12	50.8 (16.2) a	4.3 (1.7) b	6.5 (0.6) b	<0.001[b]
			Tree density, trees ha^{-1}		
1996	12	325 (110) a	279 (90) a	279 (97) a	0.73[c]
2003	12	292 (53) a	230 (77) a	299 (124) a	
			Tree height, m		
1996	12	2.7 (0.04) a	2.8 (0.4) a	2.8 (0.3) a	0.73[d]
2003	12	2.9 (0.04) a	0.9 (0.1) b	1.0 (0.1) b	<0.001[d]
			Chaining rate, ha hr^{-1}		
1997	8	--	8.2 (1.6) a	10.2 (1.6) a	0.32[e]
			Tree mortality, %		
1997	8	--	21 (4) a	19 (3) a	0.48[f]
2003	8	--	8 (5) a	4 (2) a	0.09[f]

[a] Means in rows followed by the same letter were not different, comparisons by LS means. Site was not a factor in any of the 5 comparisons (P>0.10).
[b] Main effect of years & treatments (P<0.01).
[c] Main effect of years (P=0.45) & treatments (P=0.73).
[d] Main effect of years & treatments (P<0.006).
[e] Main effect of chaining (P=0.32)
[f] Main effect of years (P<0.001) & chaining (P=0.23).

summer (Table 1). Fly activity was high in 1997, 1999, and 2002 which were years with above average rainfall between May and September.

The differences in captures that were apparent between bucket and board traps among chaining treatments reflect differences in flight behavioral characteristics of male and female flies. Female flies, which bite livestock to obtain a blood meal (Goodwin and Drees 1996), are caught almost exclusively on the white bucket traps (Moore et al. 1996), and ground-level chaining plus fire reduced female flight activity by 67% (Table 3) compared with numbers caught in the untreated plots. Males, which probably feed on pollen and nectar and do not bite, were not influenced by chaining plus fire treatments as indicated by lack of differences in numbers captured on the brown board traps. These results indicate that flight and host searching activities of females, but not males, are strongly influenced by amount of canopy cover in an area. Moore et al. (1996) reported that adult *T. abactor* did not prefer

open areas devoid of a canopy cover provided by redberry juniper or honey mesquite trees; however, Holmes (1998) investigated fly activity prior to chaining in 1996 and concluded that brush management needed to reduce canopy cover below 20% to achieve suppression of fly numbers. Juniper canopy cover was reduced by chaining and burning to ca. 5% in this study. Cooksey and Wright (1987) reported that significantly more *T. abactor* were captured at the edge of a wooded area compared to numbers captured in a cleared area that was 150 m from the wooded area.

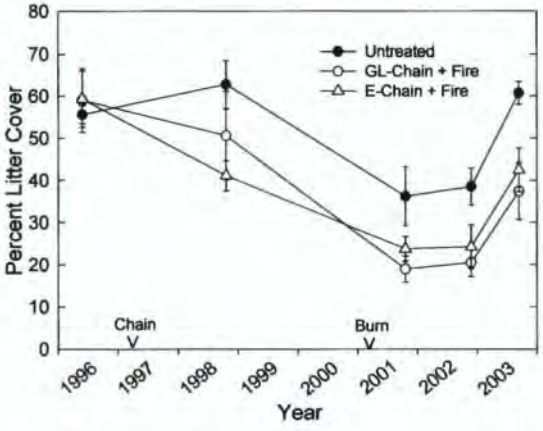

FIG. 1. Percentage litter cover beneath intact juniper canopies (all treatments in 1996 and untreated in all years) or in areas that were beneath juniper canopies prior to treatment (chain + fire treatments after 1997). Vertical bars are ± 1 standard error (n=4). GL-Chain = ground level chain; E-Chain = elevated chain.

The significant decrease in larval numbers in all treatments after 1998 (Table 2) could be the result of exclusion of cattle from the study areas from 1996 through April 2001. Cooksey and Wright (1987) indicated that *T. abactor* populations disperse from areas lacking hosts. Other factors that might reduce larval numbers are the loss of mating and resting sites. We found *T. abactor* mating within the foliage of redberry juniper trees, and these mating sites in chained and burned plots (two of the three treatments) were destroyed. Blood-engorged females may not fly very far after feeding, and we found blood-engorged females resting on trunks and large limbs of junipers. Kingston et al. (1986) indicated that resting *T. abactor* preferred habitats with dense cover as opposed to areas lacking cover. Cooksey and Wright (1987) reported that 58-74% of marked, blood-engorged females were recaptured within 0.4 km, and 76-100% were recaptured within 0.8 km of the release site.

Tabanus abactor females undoubtedly re-invaded the study areas from adjacent juniper habitats. However, destruction of juniper trees which serve as mating and resting sites and absence of cattle as a blood meal source probably lead to a reduction in oviposition and subsequent larval numbers throughout the fenced study areas. Although differences among treatments were not significant (Table 3), no larvae were found in either chained treatment in 2002, the first year that larvae were found only in untreated plots. By the end of 2001, nine months post-fire, there was less litter cover in the chained and burned plots (Fig. 1), and this may have reduced larval habitat in the soil (Montandon et al. 1993). It should be noted that percentage litter cover only indicated that the soil surface had some litter cover, but this value did not provide an estimate of litter depth.

Burning did not provide additional suppression in numbers of female flies compared with suppression obtained by chaining. A single degree of freedom comparison between untreated and chained plots prior to and after burning was not significant ($F = 0.05$; df = 1,36; $P = 0.833$ for bucket traps), indicating that felling junipers by chaining was the dominant factor involved in reducing captures of female flies on the white bucket sticky traps. Numbers of flies captured on board traps and larval numbers in the soil were not affected by burning as indicated by a single degree of freedom test comparing untreated and chained plots prior to and after burning ($F = 0.001$; df = 1,36; $P = 0.982$ for board traps; $F = 0.01$, df = 1,36; $P = 0.93$ for larvae).

Activity of female flies remained low in chained and burned treatments even after cattle were allowed back into the study areas in 2001. The results of this study demonstrate that one major benefit derived from redberry juniper management is reduced flight activity of female *T. abactor* in cleared areas. Although we were not able to document that chaining and burning reduced larval numbers, destruction of the litter layer under redberry junipers by chaining and burning removed suitable soil habitat for larvae. Elimination of redberry juniper stands should reduce fly populations, especially if treatments cover large land areas.

Although the chaining rates were similar, the smaller tractor experienced a minor overheating problem at the Halsell site during the ground-level chaining. This indicated that the pulling requirement was greater for the ground-level chaining than elevated chaining. At 2.8 m, tree height may not have been sufficient to give a major advantage to the reduced pulling requirement of the elevated chain as outlined by Wiedemann and Cross (1996). Therefore, the ground-level chain would be the better choice mechanically for trees of this size.

The reduction in percentage juniper mortality after six years was unexpected. Some uprooted tree locations were lost over the period due to environmental conditions and the burning treatment, and some root systems must have sprouted after the first growing season, thus lowering the mortality percentage. We reject our hypothesis that burning would increase the mortality of the partly uprooted redberry junipers with exposed buds or exposed roots with some meristematic bud tissue remaining following tree removal. This response was most likely attributed to the low levels of herbaceous fine fuel at the time of burning and percentage bare ground which remained at approximately 40% during the first four years of the study (Ansley et al. 2005). We also noticed that in many instances herbaceous production was lowest in the areas where juniper canopies shaded the soil surface prior to felling by chaining. Thus, our expectation that the uprooting process of chaining would expose the basal meristem of juniper to fire and thus increase mortality was inhibited by the lack of herbaceous fuel immediately near the uprooted trunk.

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RICE STINK BUG[1] DEVELOPMENT RELATIVE TO TEMPERATURE

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ABSTRACT

The rice stink bug, *Oebalus pugnax* (F.), is an important pest of rice, *Oryza sativa* L., in the United States. Little is known about the effects of temperature on its developmental rate. An experiment was conducted in temperature-controlled chambers to determine the effect of four constant temperatures on development of rice stink bug from egg to adult and to develop a degree-day model. The base of the rice panicle was immersed into a vial of water to provide food for nymphs and adults. The number of days required for egg hatch and the duration of each instar was recorded until adult eclosion. Developmental time of each instar differed significantly among temperatures. Egg hatch occurred at all temperatures except 15°C. At 15°C, second-instar development was monitored for 27 d but discontinued when food panicles became unavailable. Total development time from egg to adult was 36.8 d at 21°C, 31.6 d at 23°C and 17.9 d at 29°C. The lower developmental threshold varied from 12.1°C (third instar) to 14.7°C (second instar). The total degree-days accumulated above the average lower developmental threshold of 14°C from egg laying to adult eclosion were 281.1, 280.8 and 249.4 at 21, 23 and 29°C, respectively.

INTRODUCTION

The rice stink bug (RSB), *Oebalus pugnax* (F.) (Hemiptera: Pentatomidae), feeds on developing seeds of gramineous plants in the southern United States. The host range of RSB includes cultivated crops such as rice, *Oryza sativa* L.; barley, *Hordeum vulgare* L.; rye, *Secale cereale* L.; oats, *Avena sativa* L.; corn, *Zea mays* L.; sorghum, *Sorghum vulgare* Pers.; and wheat, *Triticum aestivum* L. (Odglen and Warren 1962). In Arkansas, from mid-April to early May, RSB emerge from overwintering sites, mostly woodland trash and bunch grass, (Odglen and Warren 1962). A rapid increase in RSB population has been noted with sweep net samples in rice during the grain-filling period and attributed to adult migration and egg hatch (Jones and Cherry 1986). Direct monitoring of RSB can be made using a sweep net, but patchiness of weed hosts often makes locating weed hosts tedious. The eggs are laid in two parallel rows and may be deposited on stems, leaves or panicles of different grasses (Nilakhe 1976). Freshly laid eggs are green and become red prior to hatching. The incubation is four to eight days, depending on temperature (Odglen and Warren 1962).

Under natural conditions, temperature largely determines the rate of development in insects (Liu et al. 1995). Several computational procedures exist for calculating daily degree-days and the practical application of predictive degree-day models (Pruess 1983, Higley et. al. 1986). The degree-day approach has been successfully applied for sucking insect pests, such as lygus bugs (Sevacherian et. al. 1977). No information is available for

Hemiptera: Pentatomidae[1]

RSB in regards to temperature effects on developmental rate or a predictive degree-day program. This experiment was conducted to estimate a lower developmental threshold and degree-days accumulations for the development of RSB.

MATERIALS AND METHODS

In 2002, the duration of RSB development from egg to adult was determined in environmental chambers equipped with white 20-watt fluorescent lamps (Powers Scientific Inc., Hatboro, Pa; Model: DS33SD) maintained at 14:10 L:D photoperiod. The chamber temperatures were 15±1, 21±2, 23±2 and 29±2°C, respectively. Temperatures were recorded every 30 min using a StowAway™ Data Logger. RSB eggs were obtained from a laboratory caged colony. Adults were collected from rice fields and grass hosts at Stuttgart, AR, and maintained at the University of Arkansas Agricultural Experiment Station, Fayetteville at 28±2°C, 14:10 L:D photoperiod. The food was provided as greenhouse-grown, excised developing barnyardgrass, *Echinochloa crusgalli* (L.) Beauv., and rice, *Oryza sativa* L. panicles in milky stage with panicle bases immersed in 0.5-liter glass bottles half filled with water and secured with paper towels. The cages consisted of 19-liter plastic pails covered with 64-mesh nylon cloth. Cages were checked at 0800, 1300, and 1800 hours CDT for freshly laid cohorts of eggs. Eggs collected only at 1300 and 1800 hours CDT were used in this experiment. Cohorts of 30 eggs were incubated at each temperature in petri dishes containing a piece of moistened cotton (three replicates). In 1987, newly laid egg masses were removed daily from caged colonies (as above) (J. L. B., unpublished data). Seven to 20 egg masses of RSB were incubated in environmental chambers (Calumet Scientific Inc., Calumet, IL) set at 21.1±2°, 23.9±2°, 26.7±2°, 29.4±2°, 32.2±2°, 35.0±2° and 37.8±°C, respectively. Total hours to hatch were recorded for 90% of the eggs in a mass to hatch at each temperature. Total days to hatch were recorded for each egg. After hatch, first instars were provided with water-moistened cotton until molting into second instars.

Food was provided to nymphs for development rate studies. Typically, many pentatomids do not feed during the first instar (McPherson and McPherson 2000). Developing rice panicles were excised from the plants grown in the greenhouse. Panicles were cut in small pieces each with six to eight kernels in the milk or soft dough stage. Eppendorf® pipette tips were cut and modified to prepare a water-containing vase for the rice panicles. The base of each piece of rice panicle was inserted into the tip filled with water and secured with cotton. This feeding apparatus was placed in a 30–ml clear plastic diet cup (Solo Cup Co., Urbana, IL). Fifteen nymphs, newly molted to second instar, were randomly selected for each replication and placed at each temperature. A second-instar nymph was introduced onto the rice kernels in each diet cup with a camel's hair brush. The rice panicles were replaced every other day at 23±2 and 29±2°C, and after every two days at 15±1 and 21±2°C. Cups were examined twice daily at 0800–0900 and 1600–1700 hours CDT for mortality or cast exuviae until adults eclosed. The duration of each nymphal instar and sex of adult were recorded.

Daily degree-days were calculated by *x*-intercept method (Arnold 1959) and accumulated over days. The following degree-day formula was used:

$$\text{Degree-days} = \frac{\text{Daily Max. Temp.} + \text{Daily Min. Temp.}}{2} - \text{Lower Dev. Threshold}$$

Data on the effect of temperature on development time for each sex were analyzed with analysis of variance (ANOVA) and student's *t*-test to separate means (P=0.05). The stage-specific and overall lower developmental thresholds were determined by simple linear

TABLE 1. Mean Duration (Days ± SE) for Development and Percent Mortality of Eggs and Nymphs of the Rice Stink Bug, *Oebalus pugnax* (F.), at Different Temperatures.

Temp. (°C)	Stages (Egg or Instar)						
	Egg	1	2	3	4	5	All
			Duration (days)				
15	-[a]	-	16.8 ± 0.54a[b]	17.0 ± 0.52a	-[c]	-	-
21	7.3 ± 0.07a	4.0 ± 0.04a	5.6 ± 0.21b	5.4±0.17b	5.9 ± 0.11a	8.7 ± 0.14a	36.8±0.32a
23	6.7 ± 0.07b	3.3 ± 0.07b	4.1 ± 0.13c	4.9 ± 0.20c	5.3 ± 0.13b	7.4 ± 0.13b	31.6±0.36b
29	4.3 ± 0.07c	2.0 ± 0.04c	2.0 ± 0.12d	2.3 ± 0.09d	3.1 ± 0.10c	4.2 ± 0.09c	17.9±0.20c
			Percent Mortality				
15	-	-	28.9	18.8	-	-	-
21	16.7	8.9	0.0	4.9	2.6	7.9	-
23	33.5	6.7	7.1	7.7	5.6	2.9	-
29	13.3	11.1	5.0	2.6	0.0	2.7	-

[a]Not hatched after being observed for 28 days.
[b]Means in same column with different letters are significantly different ($P<0.05$, Student's t test).
[c]Terminated due to unavailability of rice panicles.

regression of mean proportional growth (1/d, where, d = days for development) by temperature for the egg and each nymphal instar and across all stages (SAS Institute 1996).

RESULTS AND DISCUSSION

The duration of the period from oviposition to hatch became significantly shorter with increase in temperature (Table 1). No egg hatch occurred at 15±2°C although eggs were monitored for 28 d. There was no significant difference in development time between males and females at different temperatures ($P > 0.05$, Student's t test). The average duration from egg to adult at 29±2°C was 17.9 d compared to 31.6 and 36.8 d at 23±2 and 21±2°C, respectively (Table 1). Because no egg hatch was observed at 15±1°C, the experiment was initiated with second-instar nymphs from eggs incubated at 28±2°C. From the 1987 egg development experiment, total days to hatch were 11.2, 7.4, 5.5, 4.3, 3.8, 3.0 and 0 at 21.1±2, 23.9±2, 26.7±2, 29.4±2, 32.2±2, 35.0±2 and 37.8±2°C, respectively. Since no egg hatch occurred at 37.8°C, the upper developmental threshold for egg hatch was estimated between 35 and 37.8°C. At 15±1°C, the second instar required an average of 16.8 d. Third-instar nymphs were still feeding after 17 d, but the experiment was terminated due to unavailability of rice panicles. Percentage survival from egg to adult was 77.8, 73.3 and 80% at 21±2, 23±2 and 29±2°C, respectively.

The linear equations relating proportional growth to temperature for each stage are presented in Table 2. The lower developmental thresholds were 15.9, 13.2, 14.7, 12.1, 13.6 and 14.1°C for egg, and first to fifth instars, respectively. The average threshold across all stages was 14°C. Degree-days were calculated using this average lower developmental threshold (Table 2). The total degree-days accumulated from oviposition to adult eclosion were 281.1, 280.8 and 249.4 at 21, 23 and 29°C, respectively (Table 3). No developmental maximum was estimated for the other RSB stages. These findings need to be validated in the field.

TABLE 2. Linear Regression Equations of Mean Proportional Growth (Y) by Temperature (T) for Eggs and each instar of the Rice Stink Bug, *Oebalus pugnax* (F.).

Egg/Instar	Linear Equation	R^2	LDT[a] (°C)
Egg[b]	Y = 0.017 T – 0.27	0.99	15.9
1st	Y = 0.032 T – 0.42	0.99	13.2
2nd	Y = 0.038 T – 0.56	0.90	14.7
3rd	Y = 0.024 T – 0.29	0.86	12.1
4th	Y = 0.022 T – 0.30	0.96	13.6
5th	Y = 0.016 T – 0.23	0.99	14.1

[a]Lower developmental threshold.
[b]Egg development (J. L. B., unpublished data).

Given a biofix of first egg hatch, this degree-day information may be used to assist growers in predicting the earliest date when the first RSB nymphs will eclose to adults. At that time growers could begin scouting grass hosts in rice field margins for RSB presence and make decisions about their management. This information could be used to alert farmers with fields in heading, milk and dough (vulnerable stages) which coincide with adult eclosion. Similar control strategies based on degree-day models have been suggested for other hemipterans, such as, *Biprorulus bibax* (Breddin) (Pentatomidae), a major pest of citrus in Australia (James 1990) and *Calocoris norvegicus* (Gmelin) (Miridae), an important pest of pistachios in California (Purcell and Welter 1990).

TABLE 3. Cumulative Degree-Days (Lower Developmental Threshold = 14°C) for Eggs and Nymphs of Rice Stink Bug, *Oebalus pugnax* (F.), at Different Temperatures.

Temp. (°C)	Nymphal Stage						Total
	Egg	1	2	3	4	5	
21	55.3 (52.05 – 61.6)[a]	29.2 (29.1 – 29.3)	42.4 (39.7 – 43.7)	42.1 (39.8 – 43.2)	50.1 (49.8 – 50.8)	62.0 (56.3 – 65.2)	281. 1 (274.2 – 290.4)
23	59.7 (56.2 – 66.54)	28.1 (18.6 – 37.4)	40.7 (37.6 – 47.0)	37.5 (37.4 – 37.6)	49.1 (34.4 – 56.4)	65.7 (56.4 – 75.0)	280.8 (259.6 – 291.5)
29	50.2 (46.9 – 52.7)	25.2 (22.1 – 28.0)	36.2 (30.6 – 39.2)	27.7 (22.1 – 30.6)	43.5 (38.8 – 45.8)	66.6 (61.1 – 77.5)	249.4 (237.9 – 259.3)

[a]Range in parentheses.

The degree-day model could improve the timing of releases of the egg parasitoid of RSB, *Telenomus podisi* (Ashmead). This parasitoid was found to parasitize up to 71% RSB eggs in the field (Sudarsono 1989). Mass releases of this parasitoid could be based on cumulative degree-days predicting the RSB oviposition period. This strategy may enhance the potential of the parasitoid as a biological control agent.

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SURFACE ACTIVITY OF NATIVE ANTS CO-OCCURRING WITH THE RED IMPORTED FIRE ANT, *SOLENOPSIS INVICTA* (HYMENOPTERA: FORMICIDAE)

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ABSTRACT

We investigated surface activity patterns of the red imported fire ant, *Solenopsis invicta* Buren, and five species of native ants that commonly co-occur with it in the southern United States. Species differed temporally in abundance, both according to time of year and time of day. These differences appeared largely independent of time per se, however, while strongly associated with temporal differences in substrate temperature. We also found differences among species in their location above ground; some were more likely to occur at ground level, while others were more likely to occur above ground on plants. Above ground frequency of the five native species decreased significantly with increasing overlap with *S. invicta* in temperature-dependent surface activity and location, suggesting that activity overlap could be one important factor in determining the impact of *S. invicta* on native species. In addition, environmental matching by *S. invicta*, allowing for extensive, relatively constant activity could be one factor important in their successful invasion of the southeastern United States.

INTRODUCTION

In portions of the southeastern United States, the red imported fire ant, *Solenopsis invicta* Buren, has largely eliminated two species of native fire ants, *Solenopsis geminata* (Fabricius) and *Solenopsis xyloni* McCook, and has negative effects on other species as well (e.g., Wilson and Brown 1958, Hung and Vinson 1978, Porter et al. 1988, Porter and Savignano 1990, Gotelli and Arnett 2000, Wojcik et al. 2001). However, some native ants appear to coexist rather well with *S. invicta* (e.g., Helms and Vinson 2001, Morrison 2002, Morrison and Porter 2003). Mechanisms by which *S. invicta* excludes closely related and ecologically similar native *Solenopsis* (*geminata* group) have been fairly well studied (e.g., Tschinkel 1988, Tennant and Porter 1991, Morrison 2000); however, the mechanisms inhibiting or promoting coexistence of other native ants have not.

Competition for limited resources is likely to be important in determining the degree that native ants coexist with an invasive species (Human et al. 1998, Holway et al. 2002), and similarities and differences in activity patterns could influence the strength of competition among species (e.g., Schoener 1974, Klotz 1984, Cerda et al. 1998, Davidson 1998, Albrecht and Gotelli 2001). We studied whether activity overlap was important in the degree to which native ants coexist with *S. invicta* by studying the surface activity of *S. invicta* and five native ant species that commonly co-occur in the southeastern United States, *Monomorium minimum* Buckley, *Forelius pruinosus* (Roger), *Dorymyrmex flavus* McCook,

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Pogonomyrmex barbatus (Smith F.), and *Paratrechina* sp. (*P. vividula* (Nylander) or *P terricola* (Buckley)). Our study expands upon more limited studies addressing this topic (Claborn and Phillips 1986, Phillips et al. 1986, Claborn et al. 1988, Wuellner and Saunders 2003) by observing the natural activity of a relatively large group of species occurring within a single habitat, and by providing the first quantitative estimates of overlap in surface activity among *S. invicta* and native species.

MATERIALS AND METHODS

Our study was conducted from September 1998 through September 1999 in a 0.6-hectare meadow, 8 km south of College Station, Brazos Co., Texas. The meadow is typical of the post oak (*Quercus stellata*) savannah region of eastern Texas (Helms and Vinson 2001). *Solenopsis invicta* has occurred on the site for at least 10 years. At this site, most, if not all, *S. invicta* colonies were the monogyne, or single queen, form (Helms and Vinson 2001).

Six ant species comprised approximately 98% of all ants observed in the meadow: *S. invicta*, *D. flavus*, *F. pruinosus*, *M. minimum*, *P. barbatus*, and *Paratrechina* sp. (*P. vividula* or *P. terricola*; these *Paratrechina* could not be reliably distinguished based on worker morphology [Trager 1984]; Helms and Vinson 2001). These species often co-occur with *S. invicta* in the southeastern United States (Baroni Urbani and Kannowski 1974, Apperson and Powell 1984, Claborn and Phillips 1986, Claborn et al. 1988, Stein and Thorvilson 1989, Camilo and Phillips 1990, Hook and Porter 1990, Porter and Savignano 1990). We determined their frequency above-ground by walking along four randomly selected transects and recording the number of each ant species. The transects were 0.6 m wide and varied in length, with a total area of 100 m^2. Observations were conducted close to ground level, and each species was distinguished visually in the field. Other ant species occurring in the meadow are rarely observed (Helms and Vinson 2001) and, when they occurred, could be differentiated from the six species studied. Our observational procedure has advantages over baiting and pitfall traps for assessing spatial and temporal abundances of ants in that it does not bias the results toward species that recruit particularly well or that are easily trapped. However, our method requires that difference in size and activity among species does not make some more likely to detect than others. As an independent assessment of our procedure, we compared transect observations with observations at nest entrances, as described below.

Observations along transects took approximately one hour and were repeated at two-hour intervals throughout the day and night. Night observations were conducted with a low intensity headlamp. The number of ants per observation period was the sum of the number of each species observed during the period, and the time of day assigned to each observation period was the median time between the beginning and the end of each period. Twenty-six observation periods were at night and 61 were during daylight. Daytime observations were conducted on 13 days during September, October, November, and December of 1998, and January, February, April, July, August, and September of 1999. Nighttime observations were conducted on five nights during September and October of 1998, and August and September of 1999. The number of one-hour daytime observation periods ranged from three to six per day (mean = 4.7), and the number of nighttime observation periods ranged from two to six per night (mean = 5.2). Overall, 87 hours of transect observations were made over 156 consecutive hours.

We defined above-ground activity as that which occurred on the ground surface as well as on the stems and leaves of plants (grasses, broadleaf forbs). During an eight-day period near the end of our study, the locations of ants above ground were noted. If an ant's location was on an upright plant stem or leaf, the recorded location was above ground level, whereas an ant on the ground surface or on a prostrate plant was recorded as at ground level. These

observations were made during four days in September 1999, over 11 daytime and 6 nighttime observation periods.

Beginning in December 1998, substrate (surface) temperature was measured with a "Raynger ST" (Raytek Co., Santa Cruz, CA) infrared temperature sensor, accurate to $\pm$ 0.5°C, at a single random point along each of the four transects during each one-hour observation period. From the four measurements, a mean temperature was calculated for each observational period. Mean surface temperatures were determined for 47 hours of transect observations over 84 consecutive hours on 10 calendar days occurring in seven months.

As an evaluation of our transect data, the numbers of workers leaving and entering colony nest entrances were counted during two days and two nights in mid-August 1999. A single nest entrance of each species was observed sequentially, and the numbers of workers entering and leaving the nest entrance were counted for 10 minutes. Five minutes later, the numbers at another species' nest entrance was counted for 10 minutes, and so on, until worker numbers entering and leaving nest entrances of the six species were recorded. The procedure was then repeated, resulting in eight, 10-minute observation periods per species per day and night, with a 1.5-hour interval between each observation for each species. Temperature was measured with the Raynger ST infrared sensor at the nest entrance at the start and end of each 10-minute observation. Mean temperatures were calculated as the average temperature between the start and end of the observation period. The time of day of the observation was defined as the mean time between the start and finish of the observation. The numbers of ants active on the surface were the sums of workers entering and leaving nests over the 10-minute period.

In many cases, distributions of our data were significantly different from normal and/or exhibited variances significantly different from equal. The reason appears largely attributable to frequent inactivity, resulting in zeros for surface activity, and distributions skewed towards zero. Transformations failed to normalize the distributions or to equalize the variances, so we employed non-parametric statistical analyses in those cases. Unless noted, measures of statistical variance are presented as standard errors of the mean. In our analysis of activity as a function of time of day and surface temperature, we employed the non-linear regression curve-fitter of Sigma Plot 5.00 (SPSS Inc., Chicago, Illinois), with the best overall fit for the six species achieved by the peak (normal, or gaussian) function:

$$y = ae^{\left(-0.5\left(\frac{x-x_0}{b}\right)^2\right)}$$

where y = surface activity in number of individuals observed, a = the proportion of all individuals predicted to be on the surface at peak surface activity, e is the base of the natural log, x is surface temperature, x_o is surface temperature where peak activity occurs, and b = the standard deviation of the surface temperature distribution. From these equations, the temperature at which peak surface activity occurred was estimated, as was the temperature range within which significant surface activity occurs.

Areas under the peak regression curves represent the sum of predicted surface activity according to surface temperature for each species. Overlap in surface activity among species was therefore estimated by the degree to which curve areas overlapped. However, curves generated from raw data could not be used because regressions resulted in size differences of the areas under species' curves. We equalized the areas with a transformation, where we first assigned a total number of individuals for each species (N) of 10,000. This number is arbitrary; any could be used, so long as it is the same for each species. For each species, this number was multiplied by the proportion of the total number predicted from our original regressions at each 1°C difference in temperature, beginning at the lowest temperature where one individual was predicted to be on the surface and ending at the high temperature where

one individual was predicted to be on the surface (i.e., where the proportion of the total number predicted in the original analysis was $1/N$, i.e. 1/10,000). We then fitted a new peak regression to each species' transformed data which resulted in regressions with equal areas under the curves for each species, with estimates of peak surface activity temperature and range identical to those in the original analyses for each species. The transformation equalized the areas under the species' curves but left the original relationships between temperature and activity intact. These new curves were then used to estimate overlap in surface activity among species (Fig. 1). The curves generated are based on transect data.

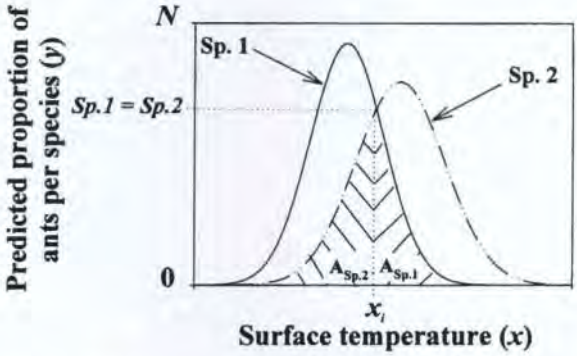

FIG. 1. Procedure used for estimating overlap in surface activity between species. After generating regression equations predicting equal areas under the peak regression curves for each of the six species (see Methods), we determined x_i, the temperature at which two species' curves intersect. This occurs when y is equal for two species (at Sp.1 = Sp.2 in the figure). We set the equations of two species (1, 2) equal to one another: $a_1 e^{\left(-0.5\frac{(x_i - x_{01})^2}{b_1^2}\right)} = a_2 e^{\left(-0.5\frac{(x_i - x_{02})^2}{b_2^2}\right)}$ and solved for x (x_i when Sp.1 = Sp.2). The percent overlap under the curves was then estimated by generating probability distribution functions from the species curves using Minitab 14 (Minitab, Inc.). These distributions show the cumulative percent of the area under each species curve according to surface temperature. As shown in the figure, overlap was then estimated as $A_{Sp.2} + A_{Sp.1}$, the percent of the area under the curve of species 1 that remains at temperature x_i ($A_{Sp.1}$) added to the percent of the area under the curve of species 2 up to temperature x_i ($A_{Sp.2}$).

We also incorporated differences in species locations (on-ground vs. above-ground) into estimates of overlap according to temperature, to arrive at an overall estimate of surface activity overlap between each species (O_{TL}). We did so with the equation

$$O_{TL1,2} = \left(1 - abs\left(\frac{G_1}{G_1 + P_1} - \frac{G_2}{G_2 + P_2}\right)\right) \cdot O_{T1,2}$$

where $O_{TL1,2}$ is the percentage overlap between species *1* and *2*, G_1 (or G_2) is the mean proportion of the total number of individuals of species *1* (or *2*) observed per observation period at ground level (vs. above ground on plants), P_1 (or P_2) is the mean proportion of the number of individuals of species *1* (or *2*) observed per observation period above ground on plants, and $O_{T1,2}$ is the percentage overlap in temperature dependent surface activity between species *1* and *2*.

RESULTS

The invasive species *S. invicta* was the most frequent ant occurring on transects, approximately three-times more than any other (Fig. 2). All six species were active throughout much of the year, although all were more common in summer through early fall than late fall through early spring (Fig. 3). The proportion of individuals per species active on the surface differed according to julian date (Friedman Repeated Measures ANOVA, $\chi^2 = 52.72$, $d.f. = 5$, $P < 0.001$). *Solenopsis invicta* differed significantly from each of the native species (Tukey test for all pairwise multiple comparisons, $P < 0.05$ for each comparison); whereas no native species differed significantly from one another. This difference arises from the relative constancy of *S. invicta* surface activity relative to that of the native species (Fig. 2).

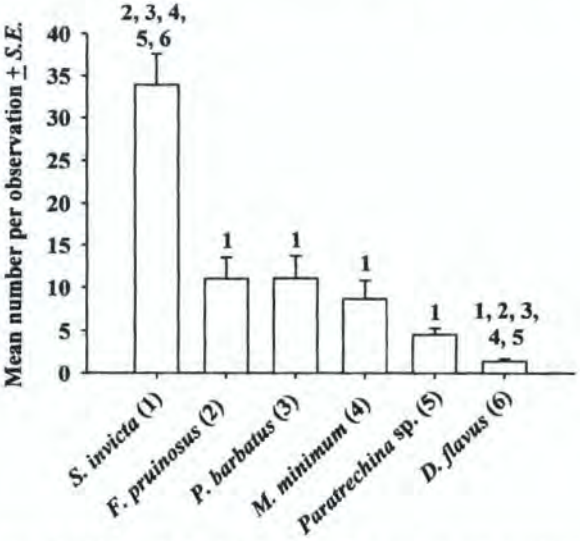

FIG. 2. Abundance of *Solenopsis invicta* and the five native ant species. Data are from transect observations ($N = 87$ observation periods). Abundance differs significantly among species (Kruskal-Wallis One Way ANOVA: $H = 111.87$, $P < 0.001$). Numbers above bars/columns indicate which species differ significantly from one another ($P < 0.05$; All Pairwise Multiple Comparisons, Student-Newman-Keuls Method).

Daily activity of species often resulted in one of three basic patterns (Fig. 4). In the summer and fall, both diurnal and nocturnal surface activity occurred, and diurnal activity often exhibited a bimodal pattern, where activity peaked in the morning and afternoon; bimodal patterns of activity occurred when mid-day temperatures were hot (Fig. 4A).

During winter, surface activity was largely or completely diurnal, and exhibited a unimodal pattern, peaking sometime during the middle portion of the day (Fig. 4 C). The surface activity of the species generally peaked at different times of the day, and these differences were also associated with differences in surface temperature (Fig. 4 A, C).

Nocturnal surface activity was restricted to warmer periods of the year, and not all species are active at night. When nighttime activity occurred, it remained stable or decreased gradually throughout the night. Night temperatures were relatively low compared those during the day, and also more stable, decreasing gradually from sunset until sunrise the following day (Fig. 4 B).

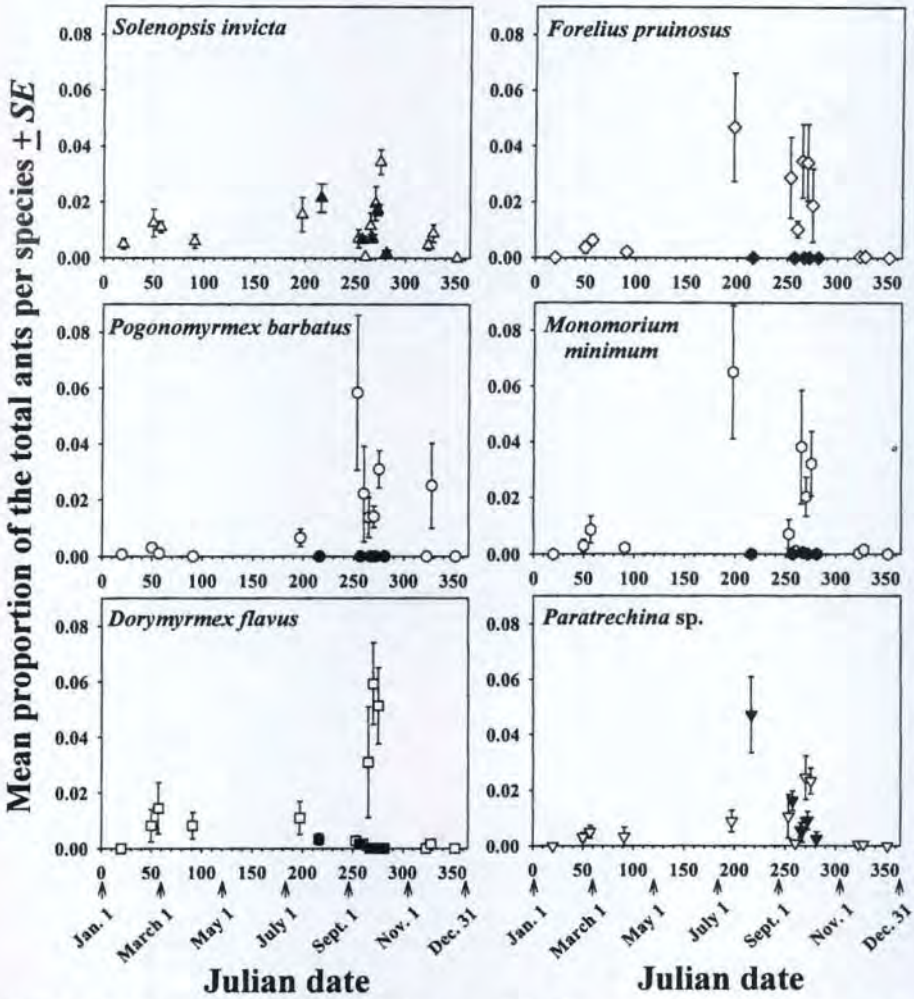

FIG. 3. Surface activity of *Solenopsis invicta* and the five native ant species on transects over the course of one year. Open symbols designate observations made during daylight hours, while filled symbols designate observations made during the night.

Pogonomyrmex barbatus and *F. pruinosus* were never observed active on the surface at night (Fig. 5 A, B). Although *P. barbatus* often occurred above-ground around nest entrances on warm nights, they were never observed venturing away from the nest perimeter. The remaining species, *Paratrechina* sp., *S. invicta*, *D. flavus*, and *M. minimum*, were observed on the surface at night as well as during the day. *Paratrechina* sp. was significantly more active at night than during the day in nest observations, while *M. minimum* and *D. flavus* were significantly more active during the day than night in transect observations but not in nest observations (Fig. 5). *Solenopsis invicta* did not differ significantly in activity

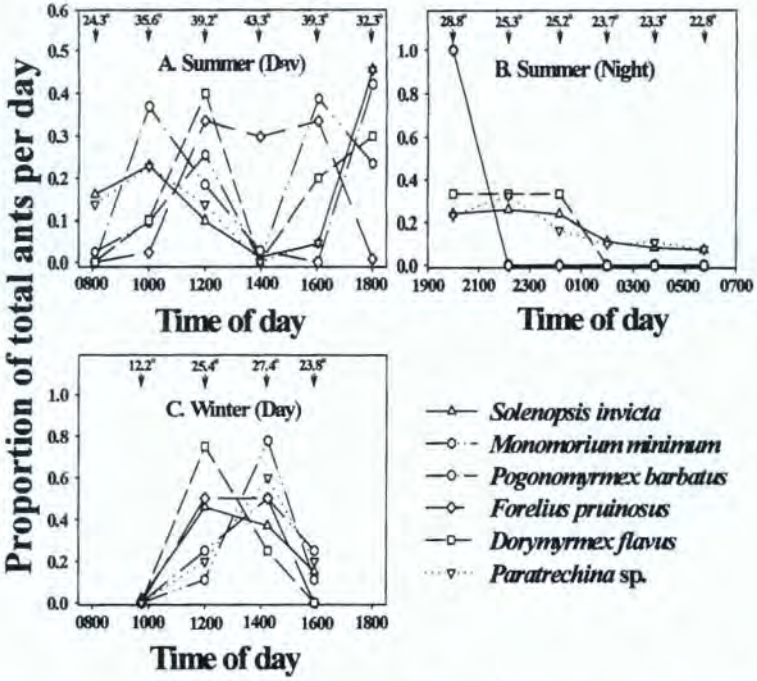

FIG. 4. Representative patterns of surface activity for *Solenopsis invicta* and the five native ant species on transects according to time of day and surface temperatures. A: July 16 1999, B: 4 August 1999, C: 19 February 1999.

between day and night in either location (Fig. 5). However, when we controlled for differences in surface temperature between day and night observations by analyzing surface activity within the range of temperatures that day and night observations share in common (where any ants were observed), only *Paratrechina* sp. and *S. invicta* differed significantly; both species were significantly more active at night than during the day (Fig. 5C). The absolute difference in surface activity between day and night was small in *S. invicta* while more substantial in *Paratrechina* sp. (Fig. 5C).

Our regression analyses revealed significant relationships between surface activity and surface temperature for all six species (Fig. 6, Table 1). In four, *S. invicta*, *P. barbatus*, *M. minimum*, and *Paratrechina* sp., strong concordances between peak surface activity temperature predicted from transect and nest data occurred (within 2° C; Fig. 6, Table 1). With the exception of *P. barbatus*, the range of temperatures where these species were predicted to be active on the surface was also very similar between transects and nest data. The nest data on *P. barbatus* predicted a substantially broader activity temperature range than did transect data (Fig. 6, Table 1). In *F. pruinosus*, nest data predicted a peak temperature 9°C higher than transect data and an proportionate increase in the upper and lower ranges of temperatures where it was predicted to be active, although both transect and nest data predicted peak surface activity temperatures and ranges substantially higher than for any other species (Fig. 6B, Table 1). While a significant peak in surface activity was predicted by transect data for *D. flavus*, no significant peak was predicted from nest data, presumably because of the scarcity of low surface temperatures during nest observations

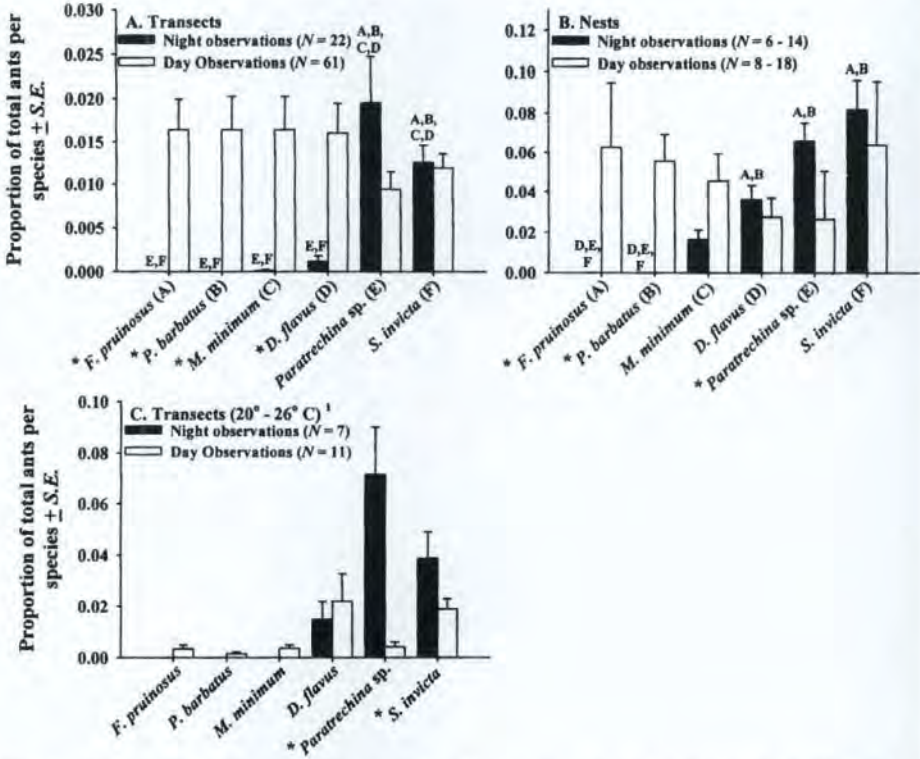

FIG. 5. Diurnal and nocturnal surface activity of *Solenopsis invicta* and the five native ant species. Asterisks preceding species names indicate significant differences between day and night activity for that species ($P < 0.05$, All Pairwise Multiple Comparisons, Dunn's Method). Letters above the bars indicate which species differ significantly from one another in day and night surface activity ($P < 0.05$, All Pairwise Multiple Comparisons, Dunn's Method).

[a] Mean temperatures: Day = 22.85 $\pm$ 0.44, Night = 23.19 $\pm$ 0.68

(Fig. 6E, Table 1). Temperature-dependent surface activity was very similar in three species, *S. invicta*, *D. flavus*, and *Paratrechina* sp., while the remaining three, *P. barbatus*, *M. minimum*, and *F. pruinosus*, were more active at higher temperatures (Fig. 7).

Unlike surface temperature, time of day was a very poor predictor of surface activity. Significant peak regressions occurred for three species (*D. flavus*, *F. pruinosus*, *M. minimum*); however, the r^2-values of regressions of surface activity as a function of time of day for these as well as the remaining species were very small (range = 0.000, 0.198). The mean r^2 across species was 0.069 $\pm$ 0.030, while the mean r^2-value with surface temperature as the independent variable was 0.476 $\pm$ 0.081. Values of r^2 were substantially and significantly greater for all species when temperature was the independent variable (paired *t*-test: $t = -5.017$, $P = 0.004$).

In addition to differences among species in surface activity according to surface temperature, differences also occurred in their above ground location during the time when these observations were conducted. *Forelius pruinosus*, *M. minimum*, and *Paratrechina* sp.

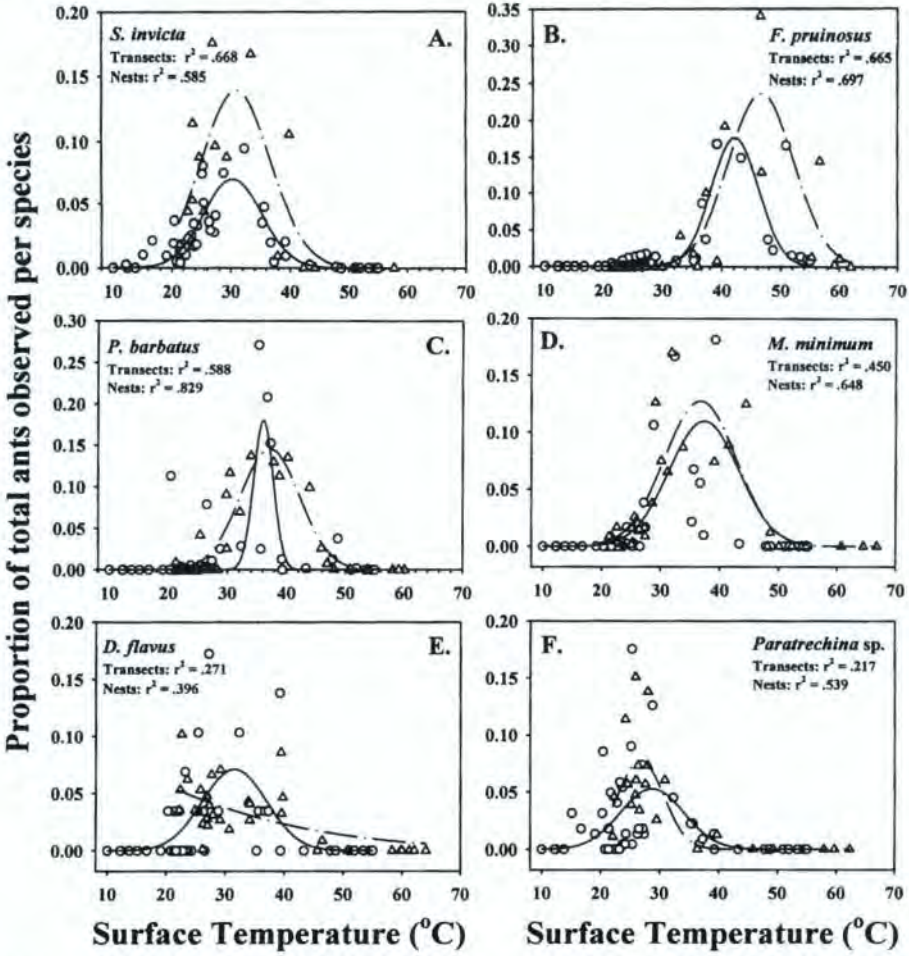

FIG. 6. Surface activity of *Solenopsis invicta* and the five native ant species according to surface temperature. Curves are peak (normal, gaussian) functions fitted to the data by regression. Triangular symbols indicate data from nest observations, whereas circles indicate data collected from transect observations. Solid lines are regression curves based on transect data, and broken lines are regression curves based on nest data. Regression statistics are shown in Table 1.

often occurred above the ground surface on plants, whereas *P. barbatus*, *D. flavus*, and *S. invicta* occurred on plants less frequently. Both *F. pruinosus* and *Paratrechina* sp. were significantly more likely to occur above ground on plants than were all other species (Fig. 8).

The abundance of native species was negatively and significantly correlated with the percentage overlap in temperature-dependent surface activity with *S. invicta* (Fig. 9A). This remained the case when we incorporated surface location into overlap in surface activity to arrive at an overall estimate of overlap in surface activity (Fig. 9B). Only overlap in surface activity with *S. invicta* predicted species abundance; overlap of any other group of five

TABLE 1. Regression statistics and estimated peak and range in surface activity for *Solenopsis invicta* and the five co-existing ant species.

Ant Species	Where Measured	Regression ANOVA		Temperature At Peak Surface Activity (x_0) ± S.E. (°C)	Surface Activity Range (°C)[a]
Solenopsis invicta	Transects: ($N = 47$)	$F = 44.32$ $P < 0.0001$	0.67	30.3 ± 0.4 $t = 71.9$, $P < 0.0001$	18.1 – 42.5
	Nests: ($N = 16$)	$F = 9.15$ $P = 0.0033$	0.59	30.9 ± 1.1 $t = 28.8$, $P < 0.0001$	15.8 – 46.0
Forelius pruinousus	Transects: ($N = 47$)	$F = 43.58$ $P < 0.0001$	0.67	42.3 ± 0.5 $t = 92.7$, $P < 0.0001$	32.4 – 52.1
	Nests: ($N = 31$)	$F = 32.24$ $P < 0.0001$	0.70	46.8 ± 0.9 $t = 51.3$, $P < 0.0001$	32.4 – 61.1
Pogonomyrmex barbatus	Transects: ($N = 47$)	$F = 31.38$ $P < 0.0001$	0.59	36.1 ± 0.4 $t = 86.8$, $P < 0.0001$	32.2 – 40.0
	Nests: ($N = 32$)	$F = 70.32$ $P < 0.0001$	0.83	37.2 ± 0.5 $t = 79.40$, $P < 0.0001$	24.1 – 50.3
Monomorium minimum	Transects: ($N = 47$)	$F = 17.98$ $P < 0.0001$	0.45	37.3 ± 1.5 $t = 24.6$, $P < 0.0001$	22.6 – 52.1
	Nests: ($N = 31$)	$F = 25.77$ $P < 0.0001$	0.65	36.7 ± 0.8 $t = 48.2$, $P < 0.001$	21.1 – 52.3
Dorymyrmex flavus	Transects: ($N = 47$)	$F = 8.18$ $P = 0.001$	0.27	31.6 ± 1.1 $t = 27.8$, $P < 0.0001$	17.9 – 45.3
	Nests: ($N = 32$)	$F = 9.52$ $P = 0.0007$	0.40	-61.2 ± 666.2 $t = 0.09$ (N.S.)	N/A
Paratrechina sp.	Transects: ($N = 47$)	$F = 6.08$ $P = 0.0047$	0.22	28.7 ± 1.5 $t = 19.2$, $P < 0.0001$	15.3 – 42.2
	Nests: ($N = 22$)	$F = 11.11$ $P = 0.0006$	0.54	26.8 ± 1.1 $t = 25.3$, $P < 0.0001$	21.1 – 52.3

[a] Range encompassing 99% of the area under the surface activity as a function of surface temperature curves

species with any other remaining sixth species failed to result in a significant correlation ($P > 0.40$ in all cases).

DISCUSSION

Our study showed that native ant species exhibited varying degrees of differences from *S. invicta* in surface activity. The surface activity of relatively abundant native species overlapped less with the surface activity of *S. invicta*, while relatively less abundant native species overlapped more (Fig. 9). This is consistent with overlap in surface activity as a factor important in coexistence. Also consistent with our results is the close similarity in thermal preferences between *S. invicta* and two species of *Solenopsis* it has largely displaced in the southeast United States, *S. geminata* and *S. xyloni* (Cokendolpher and Francke 1985). Because these species are closely related and quite similar in other aspects of their ecology as well (e.g., Potts et al. 1984, Cokendolpher and Franke 1985, Braulick et al. 1988, Tennant and Porter 1991, Trager 1991, Hooper and Rust 1997), resource competition resulting from a

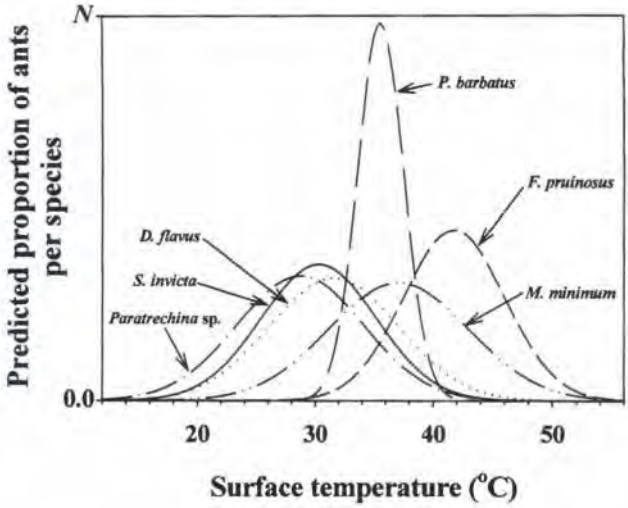

FIG. 7. Estimated overlap in temperature-dependent surface activity between *Solenopsis invicta* and the five native ant species. Curves are based on regressions of transect data. Area under the curves was equalized as described in Methods.

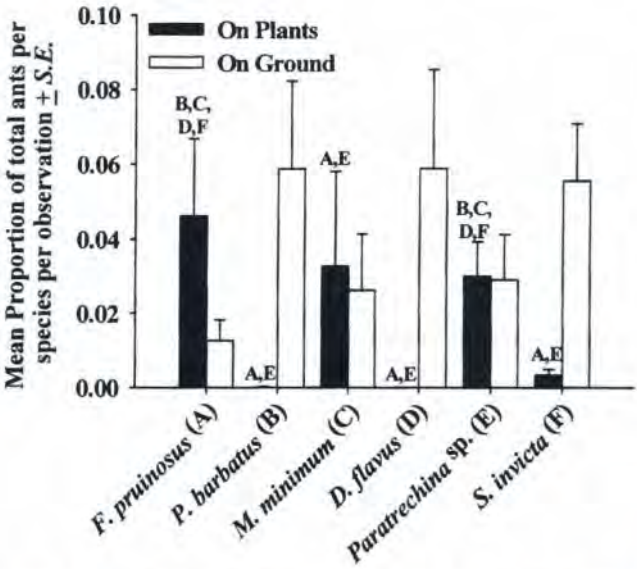

FIG. 8. Location of surface activity of *Solenopsis invicta* and the five native species. Location differs significantly among species (Kruskal-Wallis One Way ANOVA: $H = 29.923$, $P < 0.001$). Letters above the bars indicate which species differ significantly from one another ($P < 0.05$, All Pairwise Multiple Comparisons, Student-Newman-Keuls Method).

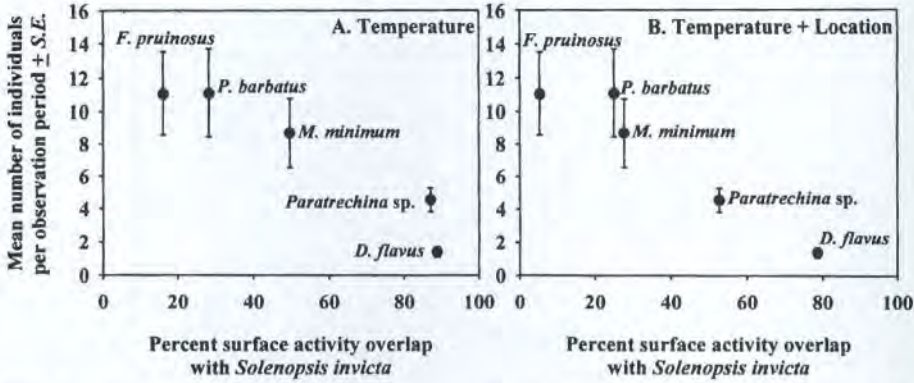

FIG. 9. Correlation between the abundance of native ant species and their degree of similarity with *Solenopsis invicta* in surface activity. A. According to the percent total overlap in temperature-dependent surface activity (r = - 0.956, P = 0.011, Pearson product moment correlation). B. According to the percent total overlap in temperature-dependent surface activity and location above ground (r = - 0.965, P = 0.008, Pearson product moment correlation).

close match in thermal preferences may be one important reason why *S. geminata* and *S. xyloni* often fail to coexist with *S. invicta*. However, additional studies will be valuable in determining the generality of our results, and it is important to note that factors in addition to surface activity patterns are also likely to be important in the impact that *S. invicta* has on native ant species (Hook and Porter 1990, Tennant and Porter 1991, Human et al. 1998, Morrison 2000).

Our study suggests that differences in surface activity appear largely dependent upon differences in surface temperature (Fig. 6, Table 1). While somewhat consistent temporal activity patterns occurred for some species, those patterns may occur because temperature can vary somewhat predictably with time. For example, *P. barbatus* and *F. pruinosus* appeared strictly diurnal; however, their surface activity did not differ significantly between day and night when analyzed over the same range of temperatures (Fig. 5). Similarly, *M. minimum* appeared largely diurnal; however, over the same range of day and night temperatures, no significant difference between diurnal and nocturnal activity occurred (Fig. 5). In fact, these three species are surface-active at higher temperatures, which rarely occur at night (the mean daytime surface temperature per observation period in our study was 32.97 ± 2.16, while the mean nighttime surface temperature was 21.32 ± 1.10).

Other than *P. barbatus* and *F. pruinosus*, the species in our study were active on the surface both day and night. However, when differences between day and night temperatures were controlled, only two species, *S. invicta* and *Paratrechina* sp., exhibited significant preferences. Both were significantly more likely to be active at night. This apparent preference was slight for *S. invicta* while more substantial for *Paratrechina* sp. (Fig. 5 C). The reasons are unclear, but could be related to differences in light intensity or in relative humidity, which is consistently greater at night than during the day (nighttime relative humidity during our study averaged 96.7 ± 1.3 % (N = 17) while daytime was 66.3 ± 2.2 %, N = 50 (Helms and Vinson, Unpublished Data).

Interestingly, our study showed that *S. invicta* was active on the surface over a range of temperatures that allowed it to be surface-active during much of the year. During summer and fall, *S. invicta* can be active throughout the night and much of the day, while during

winter and other relatively cold periods, daytime activity is often possible (Fig. 3, Fig. 4). When we analyze the variance in the proportion of all ants of each species that occurred per observation period, *S. invicta* varied little in surface activity when compared to the native species. Median surface activity differed significantly among species ($H = 36.84$, $d.f. = 5$, $P < 0.001$, Kruskal-Wallis ANOVA) and was significantly greater for *S. invicta* than any of the native species ($P < 0.05$, All Pairwise Multiple Comparisons, Student-Newman-Keuls Method). In fact, the median value for all native species other than *Paratrechina* sp. was zero (see Fig. 3). Because the most common native species in our study were surface-active at higher surface temperatures than *S. invicta* (Fig. 4, Fig. 7), this result would be deceptive if temperatures during our observations were significantly lower than normal; however, a comparison of ambient temperatures at a weather station 8 km from our study site showed that this is not the case. The mean daily ambient temperature ($\pm$ *SD*) during the course of an average year (1910 – 2002) was 19.8 $\pm$ 7.0°C, while the mean ambient temperature on the days of our observations was 24.51 $\pm$ 5.5°C (NOAA 2002).

In eastern Texas, relatively warm surface temperatures (> 35° C) generally occur over a limited period of warm and sunny days, and rarely occur at all during cool seasons or at night. As a result, surface temperatures in the preferred range of a number of the coexisting native species in our study occur during short time intervals relative to those preferred by *S. invicta*. Consistent with extensive activity as a contributing factor in the success and effects of invasive ants, a study of the Argentine ant, *Linepithema humile* (Mayr), has shown that this invasive species also exhibits less variability in surface activity than do coexisting native species (Human et al. 1998). Matching the local temperature regime to a degree that allows for extensive activity may be one factor important in the invasion and success of *S. invicta* by allowing it to discover and acquire the resources necessary to achieve and maintain large colonies and populations (e.g., Korzukhin et al. 2001). Because *S. invicta* populations are larger in the southeastern United States than in their native range in South America (Porter et al. 1997), it would be noteworthy to know if the thermal environment in the southeastern United States allows for more extensive surface activity than within its native range. This is not to suggest that *S. invicta* is not well adapted to its native environment; rather, it is possible that evolution resulting from the competitive environment in its native range could have resulted in more restricted activity than occurs within the southeastern United States. If so, this could provide additional insight into the success of *S. invicta* in the United States.

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ECTOPARASITE FLEAS[1] OF COTTONTAIL RABBITS[2] AND BLACK-TAILED PRAIRIE DOGS[3] INHABITING THE HIGH PLAINS OF WEST TEXAS

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ABSTRACT

A survey of fleas parasitizing prairie dogs, *Cynomys ludovicianus,* and cottontail rabbits, *Sylvilagus* spp., was conducted from August 2003 to January 2004 in Lubbock, Texas. This study involved the sampling of fleas from active prairie dog burrows as well as the capture of other small mammals inhabiting prairie dog towns. Of the 174 active burrows that were sampled, only five yielded fleas. The sampled rabbits had a mean number of 7.9 (SEM 1.32) fleas each (range of zero to 30), while the prairie dogs had a minimum of one and a maximum of 22 fleas each, with the average prairie dog having a flea burden of 6.9 (SEM 1.52). Identification of the fleas revealed three species: *Oropsylla hirsuta* Baker (formerly *Opisocrostis hirsutus*), *Pulex spp.,* and *Euhoplopsyllus glacialis* Taschenburg. The majority of fleas (99%) parasitizing the rabbits were *E. glacialis.* The majority of fleas (74%) on prairie dogs were *Pulex* spp., with the remaining fleas *O. hirsuta.* All of the fleas collected from within prairie dog burrows were identified as *O. hirsuta.*

INTRODUCTION

Plague is a zoonotic disease that principally affects wild rodents. The infection cycle is maintained in wild rodent colonies through the transfer of the plague bacillus, *Yersinia pestis,* by certain species of fleas (Pollitzer 1954). The black-tailed prairie dog, *Cynomys ludovicianus*, is a member of the rodent order harboring ectoparasite fleas of particular importance in the cycle of this disease. This large, social, ground-dwelling squirrel (Schmidley 2003) is considered an amplifying host for *Y. pestis*, with fleas that contribute to the proliferation of the disease among many other species of small mammals (Barnes 1993).

As part of an ongoing effort to better characterize the epidemiology of plague epizootics within prairie dog colonies, a survey of fleas parasitizing black-tailed prairie dogs and the cohabitating cottontail rabbit, *Sylvilagus spp.*, was accomplished. The survey was divided into two separate efforts, with the first portion focused on the sampling of fleas from prairie dog burrows, and the second part involving the removal of fleas from both rabbit and prairie dog pelage.

[1] Siphonaptera: Pulicidae
[2] Lagomorpha: Leporidae
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MATERIALS AND METHODS

This study was conducted in and around the city of Lubbock, TX (33° 33'N, 101° 55'W). A total of 174 prairie dog burrows, representative of 15 geographically unique, active prairie dog colonies were selected for inclusion in the burrow survey. Prairie dog colonies were observed for activity prior to sampling, and the burrows included in this study were based on recent activity. For the animal pelage survey, both rabbits (*S. audobonii* and/or *S. floridanus*) as well as black-tailed prairie dogs were trapped at two geographically isolated prairie dog colonies, separated by approximately 25 kilometers.

Fleas were collected from a prairie dog burrow by the "flagging" method of swabbing an individual burrow with cloth (Barnes et al. 1972). Briefly, this method involved using a cloth attached to a 3-m flexible metal cable that was pushed as far as possible into the burrow, held there for 30 s, and then removed. Upon removing the swab from a burrow, the cloth was placed in a plastic bag and then sealed. Upon arriving in the laboratory, plastic bags containing the swab were placed in a freezer (-20°C) and held overnight to kill all of the insects on the cloth. The fleas were later removed from the swabs, counted, and placed in 70% ethanol. The fleas were identified using published taxonomic references (Hubbard 1947, Eads 1950, Pollitzer 1954, Stark 1958, 1970, Furman and Catts 1982). Following identification, fleas were triturated in preparation for a polymerase chain reaction (PCR) assay for the direct detection of *Y. pestis*-infection. Therefore these fleas were not available for further identification using morphological keys.

Cottontail rabbits (n = 37) as well as black-tailed prairie dogs (n = 15) were trapped from August 2003 throughout January 2004. Animals were trapped using live traps, either Havahart (Model 1030, Havahart, Lititz, PA) or Tomahawk (48 x 16 x 16 cm, Tomahawk Live Trap Company, Tomahawk, WI) randomly set at each location. The traps were baited with either a sweet-mix horse feed (Purina Mills, St Louis, MO) for rabbits, or a peanut butter, oats, and sweet-mix combination for prairie dogs. Traps that were baited for rabbits were set in the evening (approximately 1700 hours) and checked the next morning (approximately 0900 hours). Trapping periods for rabbits were separated by at least 7 days. Prairie dog traps were set in the morning and checked approximately seven hours later, with trapping days separated by at least 24 h. When trapping for prairie dogs, the animals were first acclimated to the traps by baiting them for a minimum of three days and allowing the prairie dogs to remove the bait without being trapped. This acclimation period was not necessary for rabbits. Prairie dog pelage was marked using livestock marking paint (All Weather Paintstik, LA-CO Industries Inc., Elk Grove, Village, IL) to identify recaptures. Rabbits were not marked.

After the rabbits and prairie dogs had been trapped they were anesthetized prior to flea removal. Anesthesia was accomplished by placing the animal, contained within the trap, inside of a transparent container that was slightly larger than the trap. Located inside the transparent container was a jar containing cotton saturated with either chloroform (Sigma, St. Louis, MO) or halothane (Halocarbon Labs, River Edge, NJ). Once the trap (containing an animal) was placed inside the transparent container, the container was covered and the animal was monitored until rendered unconscious. Fleas were removed by vigorously combing through the pelage with a fine toothed flea comb over the transparent container, collected from the container, placed into host specific vials, and stored overnight at -20°C. The fleas were then placed in 70% ethanol and processed as previously described. All anesthesia and animal care and use procedures were approved by the Texas Tech University Animal Care and Use Committee.

RESULTS AND DISCUSSION

Of the active burrows that were sampled, only five (approximately 3%) yielded fleas. Of those five sites, one had 13 fleas and the remaining four had only one flea each. The sampled rabbits (n = 37) yielded a mean count of 7.9 (SEM 1.32) with a range from zero to 30. The prairie dogs (n = 15) each had a minimum of one flea, and a maximum of 22 fleas, and a mean flea burden of 6.9 (SEM 1.52). No prairie dogs were identified as recaptures.

The identification of the fleas revealed three species: *Oropsylla hirsuta* Baker (formerly *Opisocrostis hirsutus*), *Pulex spp.*, and *Euhoplopsyllus glacialis* Taschenberg (Table 1). The overwhelming majority of fleas (99%, n = 286) parasitizing the rabbits were *E. glacialis*. The majority of the fleas (74%, n = 75) on the prairie dogs were *Pulex* spp., with the remaining fleas (n = 27) being *O. hirsuta*. All of the fleas (n = 17) collected from the prairie dog burrows were *O. hirsuta*. All of the identified fleas are considered effective vectors of the plague (Pollitzer 1954).

TABLE 1. Species Composition of the 410 Fleas Collected from Prairie Dogs, Prairie Dog Burrows, and Cottontail Rabbits in Lubbock, Texas.

Location (sample size)	*E. glacialis*	*O. hirsuta*	*Pulex* spp.	Unknown
Prairie Dogs (15)	-	27	75	-
Prairie Dog Burrows (174)	-	17	-	-
Rabbits(37)	286	-	4	1[a]
Total	286	44	79	1[a]

[a]Flea was in poor condition and unable to be identified.

Using morphological keys to identify species within the genus *Pulex* (Siphonaptera: Pulicidae) is problematic, as species determination is dependent upon the observation of slight differences in male genitalia (Smit 1958). There is no current method to separate the females of the species *P. irritans* and *P. simulans*, and no consensus to either acknowledge the two species separately or simply merge them into one (Dittmar et al. 2003, de la Cruz and Whiting 2003). All male *Pulex* specimens collected in this survey were identified as *P. simulans* Baker.

The study area is located in an area where the historical range of the eastern cottontail (*S. floridanus*) and the desert cottontail (*S. audobonii)* overlap (Schmidly 2003), making the proper identification of rabbit species equally problematic. Differentiating between these two *Sylvilagus* species is dependent upon the relative differences in anatomical features such as smoothly rounded auditory bullae (as opposed to large and rough) and relatively shorter ears (Schmidly 2003). Using catch and release trapping, the accurate measurement of skeletal features such as bullae was not possible, and the recognition of relative ear length was subject to significant error. Therefore, no definitive classification of cottontail species is offered.

It was observed that 26% of the fleas collected from prairie dogs, and all of the fleas collected from their burrows were *O. hirsuta*. The exclusive presence of *O. hirsuta* on prairie dogs and in prairie dog burrows, and not on any of the cottontails is common. This singularity of host preference by *O. hirsuta* has previously been documented (Wilcomb 1954, Pfaffenberger and Wilson 1985). It has also been noted that *O. hirsuta* may be the only flea species recovered from prairie dog burrows (Wilcomb 1954).

Although *Pulex* species may be found on cottontails, they show greater host-preference for the prairie dog. The prevelance of *Pulex* spp. as the dominant flea taxa parasitizing prairie dogs in western Texas is consistent with the literature (Miles 1952, Roberts 2001). Additionally, similar reports of ectoparasites of cottontails cohabitating black tailed prairie dogs in nearby Portales, NM, have also been published (Pfaffenberger and Wilson 1985). This classification of *Pulex* is also in agreement with the previous identification of *Pulex* spp. fleas collected from prairie dogs in Lubbock, Texas (Roberts 2001).

The *E. glacialis* collected from rabbits were not found on either the prairie dog or the burrow swabs. The ectoparasite-host preference between *E. glacialis* and the cottontail is, in fact, a true host-association (Marshall 1981, Pfaffenberger and Wilson 1985), and the absence of this flea from the prairie dog, despite the cohabiatation of these two mammals, was expected. This flea has previously been found in robust numbers on cottontails living within prairie dog towns (Pfaffenberger and Wilson 1985).

Despite the preference of *E. glacialis* for the cottontail, some faunal overlap did occur with respect to four *Pulex* spp. fleas. Given the nature of cottontail rabbits to rear three to five litters per year within prairie dog burrows, as well as a tendency to rest in the burrows during daylight hours (Schmidly 2003), the opportunity for ectoparasite transfer is certainly present.

The "flagging" method of flea collection from prairie dog burrows has previously been described as an effective methodology for the field collection of fleas (Barnes et al. 1972, Gage 1999). However, "flagging" did not prove to be effective in this study, yielding fleas in only five (less than 3%) out of the 174 burrows that were sampled. Animal combing following the burrow swabbing revealed that the animals occupying those same burrows did harbor fleas. The time of our collection as it related to seasonal factors (i.e., temperature and humidity) or interepizootic periods (Gage 1999) may explain the low numbers of fleas collected. However, burrow swabbing may simply be an ineffective method of flea collection (Karhu and Anderson 2000).

It is of interest to note that every flea collected from a prairie dog burrow (n = 17) was *O. hirsuta.* This is contrary to the dominant species of the pelage survey (*Pulex* spp.). These data are consistent with reports that *O. hirsuta* shows exclusive host preference and perhaps are more likely than *Pulex* spp. to be obtained from burrow swabbing. These results also suggest caution when evaluating the risk of zoonotic disease outbreaks by interpreting flea survey data (Gage 1999) based on the taxonomy of fleas collected from burrow swabbing.

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EFFECT OF REPEATED SPINOSAD TREATMENTS ON CATTLE AGAINST *BOOPHILUS ANNULATUS* [1] UNDER SOUTH TEXAS FIELD CONDITIONS

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ABSTRACT

Repeated treatments of spinosad applied as a whole-body spray to calves infested with *Boophilus annulatus* (Say) were evaluated in two separate but continuous trials to determine whether a field population of ticks could be eradicated. Calves were treated at 3-wk intervals at 0.05% active ingredient (AI) for 21 wk (seven treatments), followed by 0.08% AI during the subsequent 18-wk (six treatments). While neither concentration resulted in eradication, fewer ticks were observed when the concentration was 0.08% AI. At 1 wk following each spinosad treatment, there were significantly fewer ticks on the treated (TG) calves on 10 of the 13 counting periods (76.9%). However, at 2 wk following each of the 13 treatments, tick counts on TG calves increased substantially compared to counts obtained at either 1 or 3 wk after each treatment, particularly when the spinosad concentration was 0.05% AI. At 3 wk following each of the 13 spinosad treatments, significant reductions in tick numbers on TG calves were observed on 61.2% of the 13 counting periods. Tick numbers obtained from six different sentinel groups of calves placed in pastures with the TG and untreated (UG) calves at various intervals indicated that spinosad treatments had an adverse impact on the field tick population. After the initial sentinel group was placed with the TG cattle, dramatic declines were observed in tick numbers on four of the five subsequent groups of sentinel cattle. Conversely, after the initial sentinel group was placed with the UG calves, steady increases in tick numbers were observed on four of the five subsequent groups of sentinel cattle.

This article reports the results of research only. Mention of a proprietary product does not constitute an endorsement or a recommendation by the USDA for its use.
In conducting the research described in this report, the investigators adhered to a protocol approved by the USDA, ARS, Animal Welfare Committee. The protocol is on file at the Knipling-Bushland U.S. Livestock Insects Laboratory, Tick Research Unit, USDA, ARS, Kerrville, TX.

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INTRODUCTION

The United States eradication effort against *Boophilus annulatus* (Say) and *B. microplus* (Canestrini), which are the primary vectors of bovine babesiosis (Smith and Kilborne 1893), prevents re-infestation into the U.S. principally by the systematic treatment of all livestock with an approved acaricide. Since 1968 the keystone acaricide used in the Cattle Fever Tick Eradication Program (CFTEP) has been the organophosphate (OP) agent, coumaphos (Graham and Hourrigan 1977). When cattle are held on infested premises, the Animal and Plant Health Inspection Service (APHIS), Veterinary Services (VS) branch of the USDA requires that all cattle be treated every 14 d for a period of 6-9 mo (14-21 total treatments) to ensure the eradication of the ticks. While the long-term success of the CFTEP is based on the high efficacy level of coumaphos, the emergence of OP-resistant ticks or the removal of coumaphos from the marketplace could jeopardize the success of the program. Consequently, identification and evaluation of alternative classes of chemicals is critical to maintaining a successful eradication effort.

Chemicals referred to as endectocides, such as ivermectin, moxidectin, eprinomectin, and dormectin, have shown promise as potential alternatives to the use of coumaphos in the CFTEP (Davey et al. 2001a, Miller et al. 2002, Davey and George 2002, George and Davey 2004). However, aside from endectocides, the development of spinosad by Elanco Animal Health (Greenfield, IN) is perhaps the only other compound that has potential for use in the CFTEP at the present time. Spinosad is a natural product produced under fermentation conditions of the actinomycete, *Saccharopolyspora spinosa* (Thompson et al. 1995). It is commercially available in the United States under the tradename Elector™ for use on cattle to control horn fly and lice, and stall trials have shown that it provides ≈85-90% control against *Boophilus* ticks (Davey et al. 2001b).

The purpose of this field study was to determine whether the required procedures of the CFTEP for eradicating *Boophilus* spp. on tick-infested premises would result in the eradication of a natural field population of *B. annulatus* ticks using repeated applications of spinosad at 3-wk treatment intervals. If eradication is achievable with spinosad using this repeated treatment regime it would provide the CFTEP with a viable alternative to the use of coumaphos, while reducing the number of required treatments.

MATERIALS AND METHODS

The study site is located in the vegetative zone referred to as the Rio Grande Plains (Hatch et al. 1990), consisting of ≈65% open buffel grass pasture, *Cenchurus ciliarus* L. and ≈35% woody species. The climate is characterized by short, mild winters and relatively long summers with temperatures of ≥32° C on 130 d during the year, with an average annual rainfall of 550 cm (Everitt and Alaniz 1982; National Oceanic and Atmospheric Administration 1983).

Evaluations were conducted in two separate and individually maintained pastures, each with an area of ≈6.9 ha. The study was divided into two separate but continuous phases (Phase I and Phase II). Phase I was conducted during the first 21 consecutive weeks, consisting of seven individual acaricide treatments, whereas Phase II was conducted during the last 18 consecutive weeks, consisting of six individual acaricide treatments at a different acaricide concentration. At the time the study began both pastures were infested with *B. annulatus* ticks that had been maintained under natural field conditions for a period of eight years.

Sixteen Hereford heifer calves, 6-9 mo of age and weighing ≈200 kg, were used in the study. The calves had no prior exposure to *Boophilus* spp. ticks. At 11 wk prior to

initiation of acaricide treatments, calves were randomly assigned to one of two groups, each consisting of eight calves. One group of calves, hereafter designated as the untreated group (UG), was released into one pasture where no acaricide treatments were applied during the study. The other group of calves, hereafter designated as the treated group (TG), was placed in the remaining pasture where the calves were repeatedly treated with acaricide at predetermined intervals throughout the study. Six wk prior to the initiation of acaricide treatments (designated as wk -6), calves in each pasture were gathered in penning facilities that were erected in each individual pasture. Each calf within each pasture was placed individually in a squeeze chute and all female ticks ≥5 mm in size were counted on the entire left half of the animal's body using the procedure described by Wharton and Utech (1970). This procedure provided an estimate of the number of nearly engorged ticks that would be returned to the pasture to sustain or increase the natural population. This tick count procedure was conducted for the six consecutive weeks prior to initiation of acaricide treatments (wk -5 thru wk 0). These pretreatment tick counts provided an assessment of the relative abundance of ticks on the UG and TG calves prior to initiating acaricide treatments.

The acaricide used in all treatments (13 total) was a 44% aqueous suspension concentrate of spinosad. The treatment method for each calf in both phases of the study was the same, with the only difference being that in Phase I the spinosad concentrate was diluted in water to 0.05% active ingredient (AI), whereas the concentration used in Phase II was 0.08% AI. Each treatment consisted of transporting the TG animals from the pasture to a holding area where each calf was individually restrained in a chute located in a 3.3 x 3.3 m cinder block room with a concrete floor and drain. A total of 10 liters of the appropriate concentration of spinosad was applied to the animal using a model 61 Bean® electric powered sprayer calibrated at 827 Kpa to deliver 7.125 liters per minute. Once each calf was treated individually, the group was returned to the pasture and released.

During Phase I the TG calves were treated at 0.05% AI a total of seven times at 3 wk (21 d) intervals. Following the initial spinosad treatment at wk 0, calves in both pastures (UG and TG) were gathered at weekly intervals beginning at wk 1 post-treatment and continuing through wk 21 (3 wk after the seventh spinosad treatment), to assess tick numbers as described above. Once tick counts were completed on all cattle within each group, the calves were released back into the pasture.

Implementation of Phase II was based on data collected during Phase I, which showed that the 0.05% AI concentration was not providing the desired level of control on the natural tick population. Consequently, following the seventh treatment at 0.05% AI spinosad, the concentration was increased to 0.08% AI for the last six treatments to determine whether a substantially greater adverse impact could be achieved. Both groups of calves (UG and TG) used in Phase I were also used in Phase II. Aside from the increase in spinosad concentration, all other variables remained the same as in Phase I. Following the initial treatment at 0.08% AI spinosad in Phase II, both groups of calves (UG and TG) were gathered weekly beginning at 1-wk post-treatment and continuing through wk 18 post-treatment (3 wk after the sixth treatment), to assess the tick numbers on each calf as previously described.

To further assess the density level of the tick population in the two pastures, untreated Hereford heifer calves were used as sentinel cattle by placing a new set of sentinel cattle in each pasture at regular intervals throughout the study. Three groups of sentinel calves (designated Group 1, 2, and 3) were placed in each pasture during Phase I and another three groups of sentinel calves (designated Group 4, 5, and 6) were used during Phase II (total of six groups of sentinel calves). Each set of sentinel calves consisted of three animals per pasture per interval which were pastured with the other calves (UG or TG) for a period of 2 wk (14 d). After 2 wk, the sentinel calves were removed (before detachment of engorged female ticks) and individually stanchioned

inside 3.3 x 3.3 m stalls in an open-sided barn. All engorged females that detached from each calf for a 4-wk period were collected, counted, and recorded on a daily basis. During Phase I the first set of sentinel calves was placed in each of the two pastures at 4-6 wk after the TG calves were initially treated with 0.05% AI spinosad (wk 0). Subsequently, a new set of sentinel calves was placed in the two pastures at 6 wk intervals throughout the remainder of Phase I of the study. Thus, in Phase I the second and third sets of sentinel calves were placed in each pasture at 10-12 and 16-18 wk, respectively, after the initial 0.05% AI treatment. Using the same 6-wk interval as a model, the fourth set of sentinel calves used in Phase II was placed in each pasture at 1-3 wk after the TG calves were initially treated at the 0.08% AI spinosad concentration, and the fifth and sixth sets of sentinel calves (Phase II) were placed in each pasture at 7-9 and 13-15 wk, respectively, after the initial 0.08% AI treatment.

Data obtained from the UG and TG calves during each weekly tick count and from the six sets of sentinel calves were subjected to an Analysis of Variance (ANOVA) using a Mixed Model Procedure (SAS 2002). The fixed variables used in the analyzing weekly tick counts were the treatment group (UG and TG calves) and the week number on which the tick count was conducted, while the random variable was the number of ticks counted on the calves. In the analysis of the sentinel calf groups the fixed variables were the treatment group (held with UG or TG calves) and the sentinel group number (Group 1-6), whereas the random variable was the number of ticks recovered from the sentinel animals. Differences were determined by comparing arithmetic means of UG and TG calves at each weekly counting interval, or by comparing arithmetic means of sentinel calves held with either UG or TG calves.

RESULTS

On each of the weekly pretreatment counting intervals, the TG calves harbored more ticks per calf than the UG calves, although differences were not significant (range: t=0.0-1.8; df=14; n=16; $P>0.05$) on five of the seven counting intervals (Fig. 1). However, on two occasions (wk -2 and wk 0), TG calves harbored significantly more ticks (wk -2: t=3.7; wk 0: t=3.0; df=14; n=16; $P<0.05$) than UG calves.

There was considerable week-to-week variation in the number of ticks counted on both groups of calves (UG and TG) during the 21 wk following initiation of spinosad treatments at 0.05% AI in Phase I (Fig. 1). At 1 wk following each spinosad treatment the TG calves had significantly fewer ticks (range: t=3.0-6.2; df=294; n=352; $P<0.05$) than the UG on five of the seven counting intervals (71.4%) (Fig. 1). On the remaining two counting intervals (28.6%), conducted at 1 wk following the first and third treatment, the TG calves had more ticks, but differences were not significant in either instance (1^{st} trt: t=0.7; 3^{rd} trt: t=0.9; df=294; n=352; $P>0.05$). At 2 wk following each spinosad treatment, the TG calves had more ticks than the UG calves on five of the seven weekly counting intervals (71.4%), although differences were significant only at 2 wk following the first and third spinosad treatment (1^{st} trt: t=2.9; 3^{rd} trt: t=3.5; df=294; n=352; $P<0.05$). At 2 wk following the sixth and seventh spinosad treatments (28.6%), the UG calves had more ticks than the TG calves, but differences were not significant in either instance (6^{th} trt: t=1.6; 7^{th} trt: t=0.5; df=294; n=352; $P>0.05$). At 3 wk following each spinosad treatment, TG calves had fewer ticks than UG calves on all seven of the weekly counting intervals (100%), but differences were significant only after the first, fourth, and fifth spinosad treatments (1^{st} trt: t=3.2; 4^{th} trt: t=2.3; 5^{th} trt: t=3.9; df=294; n=352; $P<0.05$).

The mean number of ticks from each of the three sentinel calf groups placed in each pasture during Phase I is presented in Table 1. The tick numbers recovered from the first group of sentinel calves, placed in each pasture at 4-6 wk after initiation of spinosad

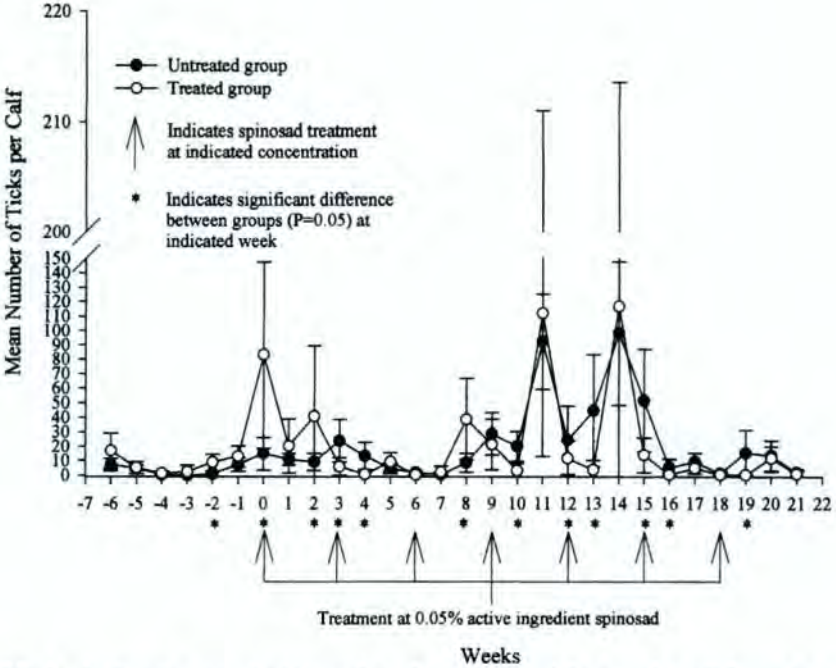

FIG. 1. Arithmetic mean (±SD) number of female *Boophilus annulatus* (≥5 mm in size) counted at weekly intervals on untreated calves and calves treated repeatedly at 0.05% active ingredient (AI) spinosad during Phase I of the study.

TABLE 1. Arithmetic Mean Number (±SD) of Engorged Female *Boophilus annulatus* per Calf Recovered from Sentinel Calves Placed in Pastures with Untreated Calves and Calves Treated Repeatedly at 0.05% Active Ingredient (AI) spinosad during Phase I of the Study.

Sentinel Group Number	Weeks calves were in the pasture after spinosad treatments were initiated	No. of spinosad treatments at time calves were in pasture	Mean no. of female ticks recovered from calves held with indicated treatment group	
			Untreated	Treated
1[a]	4-6	2	918 ± 67	17878 ± 1165
2[a]	10-12	4	2717 ± 997	13664 ± 4309
3[b]	16-18	6	3387 ± 183	2190 ± 745

[a] Indicates there was a significant difference ($P<0.05$) between the treatment groups for the indicated sentinel group of calves.

[b] Indicates there was no significant difference ($P>0.05$) between the treatment groups for the indicated sentinel group of calves.

treatments at 0.05% AI, showed that sentinel animals in the TG pasture had dramatically more (t=13.2; df=7; n=18; $P<0.05$) ticks than sentinel calves in the UG pasture during the same period. Tick counts obtained from the second group of sentinel calves, placed in each pasture at 10-12 wk following the initiation of spinosad treatments, again showed significantly more ticks (t=8.0; df=7; n=18; $P<0.05$) on sentinel animals in the TG pasture than were recovered from animals in the UG pasture. Nevertheless, the ticks recovered from the second sentinel group held in the TG pasture represented a 23.6% decline compared to the number of ticks recovered from the first sentinel group held in the TG pasture. Conversely, the mean number of ticks recovered from the second sentinel group held in the UG pasture reflected a three-fold increase compared to the first group of sentinel animals. Tick counts obtained from the third set of sentinel calves, placed in each pasture at 16-18 wk following the onset of 0.05% AI spinosad treatments, showed that sentinel calves in the UG pasture at this time period had more ticks than the group held in the TG pasture, but the difference was not significant (t=2.3; df=7; n=18; $P>0.05$). Tick numbers obtained from the third group of sentinel calves held in the TG pasture represented an 84.1% decrease compared to the second sentinel group; whereas, the tick numbers obtained from sentinel calves held in the UG pasture at this time represented a 24.7% increase compared to the second group of sentinel calves.

Week-to-week variations in tick counts still occurred in both groups (UG and TG) during Phase II, but variations were not as pronounced as in Phase I (Fig. 2). Results at 1

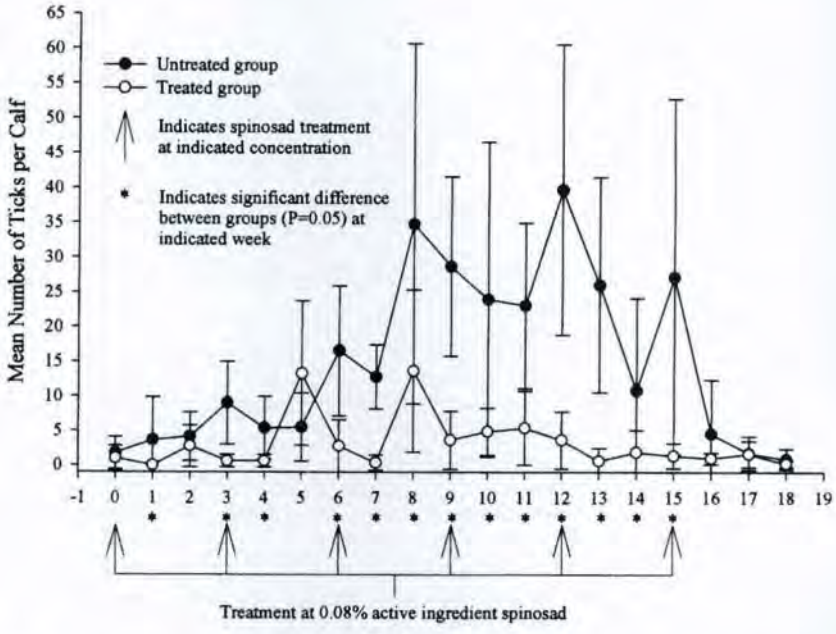

FIG. 2. Arithmetic mean (±SD) number of female *Boophilus annulatus* (≥5 mm in size) counted at weekly intervals on untreated calves and calves treated repeatedly at 0.08% active ingredient (AI) spinosad during Phase II of the study.

wk following each spinosad treatment at 0.08% AI showed that TG calves had fewer ticks than UG calves on all six counting intervals (100%); differences were significant on five of six occasions (range: t=2.3-7.5; df=252; n=304; $P<0.05$). The only exception was at 1 wk following the sixth treatment where there was no difference (t=1.4; df=252; n=304; $P>0.05$). At 2 wk following each spinosad treatment at 0.08% AI, there were fewer ticks on the TG calves on five of the six counting periods (83.3%) than were observed on the UG calves, but differences were significant only at 2 wk following the third, fourth, and fifth spinosad treatments (3^{rd} trt: t=2.4; 4^{th} trt: t=4.0; 5^{th} trt: t=3.2; df=252; n=304; $P<0.05$). At 2 wk following the second treatment (16.7%), the number of ticks on the TG calves was higher than that of the UG calves, but the differences was not significant (t=2.0; df=252; n=304; $P>0.05$). At 3 wk following each spinosad treatment, the TG calves had fewer ticks than the UG calves on all counting intervals (100%), and the differences were significant on five of the six counting periods (range: t=4.3-6.2; df=252; n=304; $P<0.05$). The only interval where no difference was observed was at 3 wk following the sixth treatment (t=0.7; df=252; n=304; $P>0.05$).

The mean number of ticks recovered from each of the three sentinel groups of calves placed in each pasture in Phase II of the study is shown in Table 2. The number of

TABLE 2. Mean Number (±SD) of Engorged Female *Boophilus annulatus* per Calf Recovered from Sentinel Cattle Placed in Pastures with Untreated Calves and Calves Treated Repeatedly at 0.08% Active Ingredient (AI) Spinosad during Phase II of the Study.

Sentinel Group Number	Weeks cattle were in the pasture after spinosad treatments at 0.08% AI were initiated	No. of spinosad treatments at time cattle were in pasture		Mean no. of female ticks recovered from cattle held with indicated group	
		0.05%	0.08%	Untreated	Treated
4[a]	1-3	7	1	3945 ± 923	10545 ± 1599
5[b]	7-9	7	3	5457 ± 1521	8544 ± 135
6[b]	13-15	7	5	4924 ± 1323	4344 ± 1478

[a] Indicates there was a significant difference ($P<0.05$) between the treatment groups for the indicated sentinel group of cattle.

[b] Indicates there was no significant difference ($P>0.05$) between the treatment groups for the indicated sentinel group of cattle.

ticks recovered from the fourth group of sentinel calves, placed in each pasture at 1-3 wk after the initiation of spinosad treatments at 0.08% AI (Phase II), showed there were significantly more ticks (t=4.9; df=7; n=18; $P<0.05$) on calves held in the TG pasture than were recovered from calves held with the UG calves. The tick numbers obtained from the fourth group of sentinel cattle held with the TG calves reflected a five-fold increase compared to the third set of sentinel calves held with the TG calves during Phase I, when the treatment concentration of spinosad was 0.05% AI. Likewise, the fourth sentinel group held in the UG pasture represented a 16.5% increase in tick numbers compared to the third set of sentinel calves held in the UG pasture during Phase I. Tick counts obtained from the fifth set of sentinel calves, placed in each pasture at 7-9 wk after the spinosad concentration was increased to 0.08% AI, showed that although cattle held in the TG pasture had more ticks than the sentinel group held in the UG pasture at this time interval, the difference was not significant (t=2.4; df=7; n=18; $P>0.05$). While the number of ticks obtained from the fifth sentinel group held in the TG pasture represented a 19.0% decrease compared to the fourth sentinel group held with TG calves, the tick numbers obtained from the fifth sentinel group of calves held in the UG pasture

represented a 38.2% increase compared to the fourth group of sentinel calves held with UG calves. Results obtained from the sixth set of sentinel cattle, placed in each pasture at 13-15 wk following the initiation of spinosad treatments at 0.08% AI, produced more ticks on the sentinel calves held in the UG pasture than were recovered from calves held in the TG pasture, although the difference was not significant ($t=0.7$; $df=7$; $n=9$; $P>0.05$). Tick numbers recovered from sentinel calves held in both pastures at this time showed a decline compared to the fifth set of sentinel calves. However, the decline in tick numbers obtained from the group of sentinel calves held in the TG pasture was dramatically greater (49.2%) than the decline in the number of ticks obtained from the sentinel group held in the UG pasture (9.8%).

DISCUSSION

Even though a prior study (Davey et al. 2001b) provided some indication that repeated applications of spinosad at 0.05% AI might result in a population collapse, this long-term study demonstrated that under the conditions tested, repeated whole-body spray treatments with spinosad were not sufficient to achieve eradication. While a number of factors probably prevented eradication, three specific factors appeared to be evident. First, the 3-wk interval between treatments used throughout the study was too long, allowing females to survive to repletion between treatments, thereby enabling the field tick population to be sustained. Second, the concentrations of spinosad used in the study (0.05 and 0.08% AI) were not high enough to kill a sufficient proportion of the ticks in the field to cause a collapse in the natural population. Although the 0.08% AI treatments (Phase II) resulted in fewer ticks on the animals than the 0.05% AI treatments (Phase I), neither concentration was high enough to produce the dramatic decline necessary to cause a population collapse. Third, the tick population in the pasture where the TG calves were held was at such a high density level that even though spinosad treatments appeared to produce substantial tick reductions, they were not sufficient to cause a collapse in the field population within the duration of the study.

The weekly pretreatment tick counts, as well as tick counts obtained from a majority of the sentinel calf groups, indicated that the tick population in the pasture where the TG calves were held was dramatically higher than the pasture where the UG calves were held, though we have no explanation for this disparity. Nevertheless, the importance of this factor in assessing the level of control seems worthy of note. Because of the extremely high density level of ticks in the TG pasture, whenever tick counts showed that TG calves had significantly fewer ticks than UG calves, it is probable that proportionally greater numbers of ticks were eliminated (killed) on the TG calves than on UG calves in order for the difference in tick numbers to be significant between the two groups of calves. Thus, the level of control on the TG calves was probably substantially greater than the data indicated from just comparing mean differences in tick numbers between the two groups.

Another factor that provided insight into the level of control was associated with analysis of data on the basis of the number of weeks following a given spinosad treatment that the tick counts were conducted (1, 2, or 3 wk post-treatment). The fact that at 1 wk following each of the 13 spinosad treatments (Phase I and II) the TG calves had significantly fewer ticks than the UG calves on 10 of the 13 counting periods strongly indicated that a substantial level of control was being achieved at this time. On the other hand, tick counts obtained 2 wk after each of the 13 spinosad treatments showed that on every counting interval during this time there was an increase in tick numbers on the TG calves compared to the tick counts conducted on the TG calves at 1 wk following each corresponding treatment. This provided a strong indication that by 2 wk following a given spinosad treatment the level of control had declined substantially, allowing females

to survive to repletion and provide a source of replenishment for ticks in the field. At 3 wk following each of the 13 spinosad treatments (Phase I and II) results were similar to those obtained at 1 wk following each treatment, showing that TG calves had significantly fewer ticks than UG calves on 8 of the 13 counting intervals at this time. These results indicated that while the level of spinosad may not have been high enough to kill ticks in the latter stages of development at 3 wk following a treatment, the activity level was still high enough to protect treated calves to some degree against larval re-infestation.

The tick counts obtained from the different groups of sentinel cattle also provided some insight into the level of control that was achieved during the study. After the introduction of the initial group of sentinel calves into the TG pasture (Group 1), there was a decrease in tick numbers on each subsequent group of sentinel animals (Group 2, 3, 5, and 6) in relation to the tick numbers obtained on the immediately preceding group, except for the fourth group of sentinel calves (Group 4). Conversely, following the introduction of the initial group of sentinel calves into the UG pasture (Group 1), there was an increase in tick numbers on each subsequent group of sentinel calves (Group 2, 3, 4, and 5) in relation to the tick numbers obtained on the immediately preceding group, except for the last group (Group 6). These results indicated that the repeated application of spinosad on the TG calves was, for the most part, causing a decline (control) in the tick population in the pasture where treatment of cattle was occurring, otherwise the tick numbers on sentinel calves held in the TG pasture would have been expected to increase, as was the case on most groups of sentinel cattle held in the UG pasture.

Results of this study showed both favorable and contrasting comparisons with other investigations conducted with endectocides that have similar acaricidal properties to spinosad. While exact control percentages were not determined in this study, it could be assumed from weekly tick count comparisons that the levels of control obtained at 1 and 2 wk following each treatment were similar to the reported 83.2% control obtained at 12 d post-treatment using a subcutaneous injection of ivermectin against *B. microplus* (Caproni et al. 1998). In two other studies using a pour-on formulation of moxidectin, investigators concluded that 21-d treatment intervals would be necessary to provide an adequate level of control against *B. microplus* (Sibson 1994, Remington et al. 1997). However, results of this study indicated that although a 21-d treatment interval with spinosad would provide some control, an even shorter treatment interval would be necessary to substantially impact the tick population, particularly when the density level of ticks in the field was at a high level. The results of this study showing a substantial decline in spinosad activity at 2-wk post-treatment appeared to contrast with an earlier study conducted in a stall setting using spinosad, where it was demonstrated that at 8-13 d (≈2 wk) after a single treatment at 0.05% AI the level of control averaged ≈90.0% (Davey et al. 2001b). However, the previous study was conducted in a barn setting where the cattle were not exposed to direct sunlight or rainfall; whereas, in this study cattle were exposed to the natural elements. Thus, it is possible that in this study the reduction in activity of the spinosad at 2 wk following treatment may have been caused by a substantial degree of photo-degradation that occurred from exposure to UV light, a phenomenon which has been reported by Cleveland et al. (2002). It is also possible that since the spinosad formulation used in the study was an aqueous based suspension, the reduction in efficacy at 2-wk post-treatment may have been caused by natural rainfall that occurred at times, which prematurely diluted the chemical to a sub-lethal level.

While the treatment regime and spinosad concentrations evaluated in this study would not be suitable for use in an eradication program, nevertheless, they could have applicability for use in a program aimed at reducing the tick population to an acceptable level (control program). Results indicated that a reasonable level of control could be achieved using the regime and concentrations tested in this study, particularly at the

higher spinosad concentration (0.08% AI) used in Phase II. It is possible that a different treatment interval might substantially improve the level of control. If intervals between repeated treatments with spinosad were reduced to 2 wk, it is possible that considerably fewer females would survive to replenish the field tick population. Additionally, if the spinosad concentration was increased to 0.15% AI, a greater adverse affect on the ticks might be expected. Previous research showed that while a 0.15% AI spinosad treatment did not substantially increase the level of control as compared to a 0.05% AI treatment; nevertheless, there was a significant decrease in the weight of egg masses produced by surviving females as compared to females treated at 0.05% AI (Davey et al. 2001b). Therefore, it could be speculated that repeated treatments at 0.15% AI might result in a decline in the field tick population because of considerably lower egg production that would be expected from surviving females. Finally, if treatments were applied at times of the year when natural tick populations were low (summer and winter), repeated spinosad treatments could cause a substantial decline in the existing population to the point that the population would collapse, perhaps leading to eradication. Obviously, further research using such treatment scenarios would need to be confirmed. However, the presence of OP-resistant ticks and/or the removal of coumaphos from the market certainly make further research into the use of spinosad seem warranted, particularly if eradication appears to be an achievable goal.

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FEEDING PREFERENCE, FECUNDITY AND EGG HATCH OF RICE STINK BUG[1] ON ARTIFICIAL DIET, RICE AND ALTERNATE HOST GRASSES

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ABSTRACT

Feeding preference, mating frequency, fecundity and egg hatch of rice stink bug, *Oebalus pugnax* (F.) were examined in the laboratory. Adult rice stink bugs were allowed to feed on one of five different food sources including rice, *Oryza sativa* L.; barnyardgrass, *Echinochloa crusgalli* (L.); dallisgrass, *Paspalum dilatatum* Poir.; ryegrass, *Lolium* spp; and a new artificial rice flour diet. Initially, a higher percentage of females fed on all food sources than did males. Ryegrass and dallisgrass were fed on less frequently than rice or barnyardgrass. The insects mated twice as frequently and laid more eggs on rice panicles than on rice flour diet. However, rice flour diet was comparable to ryegrass and dallisgrass both in mating frequency and number of eggs laid per female. Egg hatch was higher from females reared on natural hosts than on rice flour diet. However, percentage egg hatch was similar for females fed rice flour diet or ryegrass. Rice flour diet may provide an acceptable alternate food source for rice stink bug adults in the laboratory but needs to be improved to make it as acceptable as barnyardgrass or rice.

INTRODUCTION

Rice stink bug, *Oebalus pugnax* (F.), is an important pest of rice, *Oryza sativa* L., in the southern United States. In addition to rice, it has been reported to have a wide host range of gramineous plants (Douglas 1939, Odglen and Warren 1962). Rice stink bug exhibits variable feeding responses to different grass species with rice being the most favorable (Naresh and Smith 1984). The average number of eggs laid by a rice stink bug female during its lifetime ranged from 73 to 491 on barnyardgrass, *Echinochloa crusgalli* (L.) (Odglen and Warren 1962, Nilakhe 1976) and as high as 915 on rice (Nilakhe 1976).

Artificial diets are important in laboratory cultured insect colonies. Brewer and Jones (1985) have reported failure of a meridic diet for rearing rice stink bug. Mittler and Dadd (1963) reared aphids on artificial diets fed through parafilm M® membranes. Landes and Strong (1965) and Raulston and Auclair (1968) successfully reared *Lygus hesperus* (Knight) on bean juices or chemically defined diet fed through stretched parafilm membranes in sachet form.

The objective of this experiment was to compare rice stink bug mating frequency, fecundity potential and egg hatch among three wild host grasses and a new rice flour diet.

[1] Hemiptera: Pentatomidae

MATERIALS AND METHODS

Adult rice stink bugs and late instar nymphs were collected during May 2000 from rice fields and grass hosts at Stuttgart, AR, and taken to the University of Arkansas Agricultural Experiment Station (AES), Fayetteville in 19-liter plastic pails covered with 64-mesh nylon cloth. Combinations of excised panicles of different grasses including ryegrass, *Lolium* spp, barnyardgrass and dallisgrass, *Paspalum dilatatum* Poir., collected from fields at AES were provided as a food source to adults and late instar nymphs. The colony was held at the AES at room temperature of 25.6±2°C and 14:10 L:D photoperiod.

Ten grams stone ground white rice flour (Bob's Red Mill Natural Foods, Milwaukie, OR) and 5g sucrose (granulated sugar) were added to 100ml of boiling water with continuous stirring. The solution was removed from the hot plate when it became viscous. When the temperature dropped to 50°C, 1.2g Vanderzant-Adkisson® vitamin mix (Bio-Serv, Frenchtown, NJ) was incorporated into the diet solution. The diet was held at room temperature until cooled.

PM-992 parafilm M® (American National Can, Menasha, WI) was cut into sections 12.9cm^2 in size and then stretched. Approximately 5ml of diet was poured onto each stretched parafilm piece. Both sides of the parafilm were joined, pressed firmly together and then the ends were twisted. Stretched parafilm packets containing the rice flour diet were punctured with #7 insect pins mounted on a cork to induce and stimulate rice stink bug feeding. A 20-cm long straw was vertically attached to a plastic bottle lid which served as a platform. A hole was drilled through the lid to insert one end of the straw. Two parafilm packets containing diet were tied together facing each other on the straw with 5cm distance between each pair of the packets. Three pairs were tied to each straw.

Panicles and stems of barnyardgrass, ryegrass and dallisgrass collected from AES were cut to the same length as that of the straws used for rice flour diet packets. The panicles were placed in glass vials containing water and plugged with cotton.

Vertically held straws with rice flour diet packets and panicles of all three grasses in glass vials were randomly placed equidistant in a (50cm × 25cm × 30cm) glass arena and covered with metal screen. Rice stink bug adults were held without food on water-moistened cotton for 8h prior to the experiment (Naresh and Smith 1984). Twenty adult rice stink bugs were used in each of eight replications. Males and females were tested separately. The insects were released into the arena and observed for feeding 1, 3, 6, 9, 12 and 24h post-introduction, respectively at 25.6±2°C and 14:10 L:D photoperiod. An individual was considered 'feeding' when observed for at least one minute with mouthparts inserted into the food source.

The rice stink bugs were collected and held according to the protocol described previously. The experiment was conducted at a room temperature of 25.6±2°C, 14:10 L:D photoperiod. The panicles of rice from plants grown in the greenhouse and grasses (barnyardgrass, ryegrass and dallisgrass) that were collected from AES were cut 20-30cm below the panicle bases. The bases of the panicles were merged into water in glass bottles and secured with paper towels to prevent entry of the rice stink bug individuals into the bottles. Rice diet was prepared according to procedures described previously and diet packets were tied on straws. The straws and bottles holding the food source were placed on the bottom of the cage. Late fifth-instar male and female rice stink bug nymphs were identified by the presence or absence, respectively, of red testes visible through the abdominal sterna (Nilakhe 1976) and separated into cages on rice diet or one of the host grass panicles. After the molt to adult, 15 pairs of rice stink bugs (24-h old) were randomly selected and transferred into the cages consisting of 19-liter plastic pails containing one of the food sources and covered with 64-mesh nylon cloth. The panicles and the rice diet were replaced every 2 days. The cages were observed at 0800 and 2000h CDT for eggs and any

mating pairs. Eggs were collected, counted and incubated. The number of days required for at least 90% hatch from each egg mass (one egg mass for each of three replicates given > 25 eggs per mass) was recorded. Data were analyzed with ANOVA and means separated with student's *t*-test (SAS Institute 2002).

RESULTS AND DISCUSSION

Numbers of male and female rice stink bugs observed feeding were significantly greater (F = 130.29; df = 3, 383; $P < 0.0001$) on rice flour diet and barnyardgrass than on either ryegrass or dallisgrass throughout the observation period (Table 1). One hour after release, overall male feeding (32.5%) was significantly less than that observed for females (55.5%) (F = 14.24; df = 1,63; P = 0.0004) (Table 1). However, at 3 h or more post-introduction, male feeding (> 66%) was comparable to female feeding (> 74%) on all hosts. Overall, significantly more females than males fed (F = 6.65; df = 1, 383; P = 0.01) during a 24-h period. This study agrees with the previous host preference studies of rice stink bugs where the initial female response to feeding stimuli was more rapid than that of males (Naresh and Smith 1984).

TABLE 1. Feeding Preference (±SE) of 20 Male or Female Rice Stink Bugs, *Oebalus pugnax* (F.), on Different Food Sources in Laboratory.

	Time (CDT)					
Food Source	0900	1100	1400	1700	2000	0800
			Males			
Barnyardgrass	2.8±0.4a	4.8±0.3a	4.5±0.5a	5.5±0.3a	4.9±0.6a	5.1±0.4a
Rice Flour Diet	1.8±0.5ab	3.8±0.7ab	4.5±0.6a	4.5±0.7a	4.6±0.6a	4.0±0.5b
Dallisgrass	0.9±0.2b	2.6±0.2b	2.5±0.3b	2.5±0.3b	1.9±0.4b	2.3±0.3c
Ryegrass	1.0±0.3b	2.1±0.4c	1.6±0.3b	1.9±0.2b	1.8±0.3b	2.1±0.2c
Mean Response	1.6	3.3	3.3	3.6	3.3	3.4
% Feeding	32.5	66.5	65.5	72.0	66.0	67.5
			Females			
Barnyardgrass	4.4±0.5a	5.9±0.5a	5.8±0.6a	5.8±0.5a	5.9±0.4a	6.1±0.5a
Rice Flour Diet	2.9±0.4b	4.6±0.5a	4.9±0.6a	5.1±0.5a	5.0±0.5a	5.1±0.7a
Dallisgrass	2.0±0.4b	2.4±0.4b	2.3±0.3b	2.5±0.3b	2.0±0.4b	2.0±0.4b
Ryegrass	1.8±0.4b	2.3±0.4b	2.3±0.3b	2.0±0.4b	1.9±0.2±b	2.3±0.4b
Mean Response	2.8	3.8	3.8	3.9	3.7	3.9
% Feeding	55.5	76.0	76.5	77.0	74.0	77.5

[a]Means in same column with different letters are significantly different (P<0.05, Student's *t*-test)

Overall mean number of eggs increased with days after eclosion (Fig. 1). Significantly more (F = 76.36; df = 1, 629; $P < 0.0001$) eggs were collected at 0800h compared to those collected at 2000h CDT. Mean number of eggs laid per day significantly differed (F = 4.8; df = 4, 314; P = 0.0008) among food sources (Table 2).

Significantly more mating (F = 115.85; df = 1, 629; $P < 0.0001$) occurred at 2000 than at 0800h CDT. Overall, mating frequency was significantly higher (F = 11.4; df = 4, 314; $P <$ 0.0001) on rice than on barnyardgrass. Mating on barnyardgrass was greater than on ryegrass, dallisgrass or rice diet, which were all similar (Table 2).

FIG. 1. Mean Number of Egg Masses Laid per Day by Female Rice Stink Bug, *Oebalus pugnax* (F.) (n = 15, Each with 15 Females) During a 21-Day Period While Maintained in the Laboratory on *Oryza sativa* L., *Echinochloa crusgalli* (L.), *Paspalum dilatatum* Poir., *Lolium* spp and a new artificial rice flour diet at 25.6°C, 14:10 L:D Photophase.

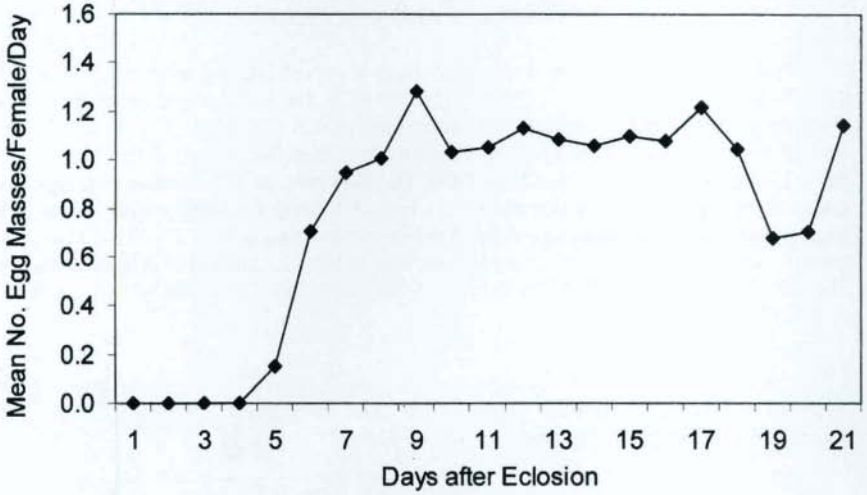


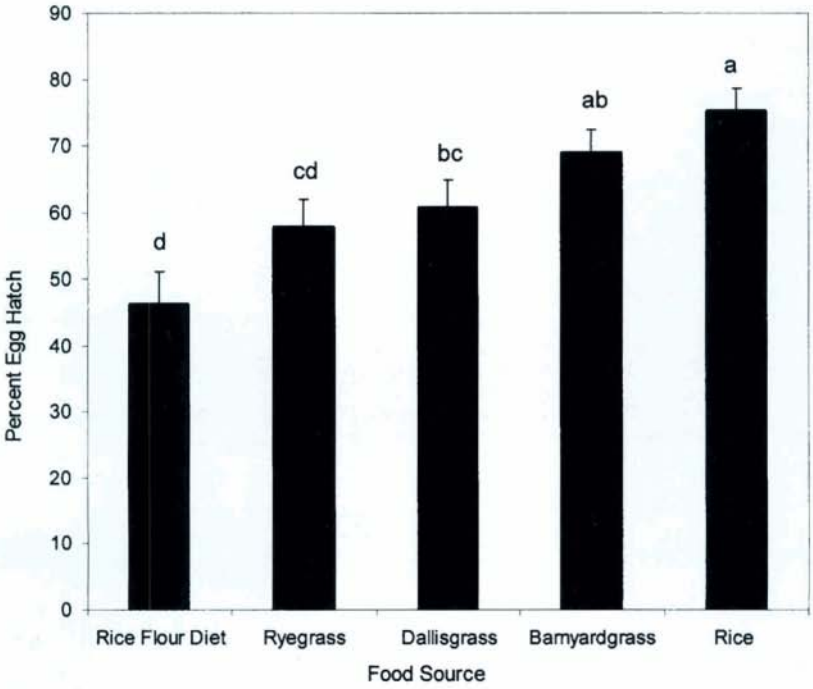
TABLE 2. Mean Number (±SE) of Matings and Eggs Laid per Day by 15 Pairs of Rice Stink Bugs, *Oebalus pugnax* (F.), on Different Food Sources in Laboratory During a Period of 21 d.

Food Source	Time (CDT) 0800	Time (CDT) 2000	Total
		No. of Matings	
Rice	1.6±0.1a	2.6±0.16a	4.2±0.2a
Barnyardgrass	1.4±0.09ab	2.2±0.14b	3.6±0.2b
Dallisgrass	1.2±0.1bc	1.9±0.14bc	3.1±0.2bc
Ryegrass	0.9±0.07c	1.8±0.13c	2.7±0.18c
Rice Flour Diet	0.9±0.07c	1.6±0.13c	2.5±0.18c
		No. of Eggs	
Rice	9.1±1.2a	3.6±0.6a	12.7±1.5a
Barnyardgrass	7.8±0.9ab	2.8±0.5ab	10.6±1.2ab
Dallisgrass	6.5±0.8b	2.8±0.6ab	9.3±1.1b
Ryegrass	5.8±0.8bc	2.3±0.4ab	8.1±1.0bc
Rice Flour Diet	4.4±0.8c	1.6±0.4b	6.0±0.9c

[a]Means in same column with different letters are significantly different (P<0.05, Student's t-test)

Egg hatch also significantly differed (F= 7.1; df = 4, 201; $P < 0.0001$) among females reared on different food sources with rice being the most efficient food source (Fig. 2). The 42.2% egg hatch recorded for females reared on rice flour diet was significantly less than that of females reared on rice (75.2%), barnyardgrass (68.9%) or dallisgrass (60.6%) but was not significantly different from ryegrass (57.7%) (Fig. 2).

FIG. 2. Percent Hatch of Rice Stink Bug, *Oebalus pugnax* (F.) eggs (n = 3 Egg Masses; Given > 25 eggs per Mass) as Affected by Adult Feeding on Different Food Sources While Maintained in the Laboratory at 25.6°C, 14:10 L:D Photophase.



Rice flour diet may be an acceptable alternate food source for adult rice stink bugs in the laboratory comparable to natural hosts such as ryegrass. Efforts to rear early instar rice stink bug nymphs on rice flour diet were unsuccessful. The early instar nymphs fed on diet packets for an extended period of time (2-3 weeks) but no growth was observed and additional molts did not occur. Further studies are necessary to determine the dietary requirements of the early instar rice stink bug nymphs to overcome the nutritional or feeding stimulant deficiencies in the diet presented here.

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