Lygus spp. (Heteroptera: Miridae) Host-Plant Interactions with Lesquerella fendleri (Brassicaceae), a New Crop in the Arid Southwest

JACQUELYN L. BLACKMER¹ AND JOHN A. BYERS²

U.S. Arid-Land Agricultural Research Center, USDA-ARS, 21881 North Cardon Lane, Maricopa, AZ 85238

Environ. Entomol. 38(1): 159-167 (2009)

ABSTRACT Lesquerella fendleri is a new crop being cultivated in the arid Southwest for the hydroxy fatty acids found in its seed oils; however, little is know about the agricultural pests that will affect the seed production of this crop or the role the crop may have as a possible source or sink for current agricultural pests in the Arizona landscape. In the early spring, lygus bugs, some of our most important agricultural pests, are normally found in relatively small numbers on weeds; however, as lesquerella production increases, it may serve as an important early-season host for lygus. Here we present results from olfactometer bioassays that showed a significant attraction by Lygus hesperus females to volatiles associated with flowering lesquerella. Headspace volatiles of flowering lesquerella were collected and identified by gas chromatography-mass spectrometry, and phenylacetaldehyde (PAA) was found to be the major component, followed by benzaldehyde and Z-3- hexenyl acetate. In the field, we examined the *Lygus* spp. complex, as well as the prevalence of other herbivores and select predators, and monitored their responses to unbaited sticky traps of various colors and PAA-baited green and blue sticky traps. Green, blue, and clear sticky traps captured significantly more Lygus spp. (L. elisus in particular) than opaque yellow and red traps, but PAA-baited blue and green traps did not capture more lygus than unbaited traps. *Collops* spp., however, were collected in higher numbers on PAAbaited traps, suggesting that this compound might provide a means of recruiting and/or retaining this particular natural enemy.

KEY WORDS western tarnished plant bug, visual cues, olfactometer, natural enemies, phenylacetaldehyde

Lesquerella fendleri (A. Gray) S. Wats is a member of Brassicaceae and can be found growing naturally in the southwestern United States and northern Mexico. Its seeds contain oils rich in hydroxy fatty acids, which are important raw materials used in cosmetics, adhesives, waxes, lubricants, and pharmaceutical and medical products (Roetheli et al. 1991, Dierig et al. 1992, Thompson and Dierig 1994). Currently, these oils are obtained from castor oil; however, potent allergenic compounds on the plant surface make harvest risky, and the seed meal has large amounts of the highly lethal ricin protein. Consequently, a search for an alternate domestic source began in the 1960s. Although there are >90 species in the *Lesquerella* genus, only L. fendleri is currently being domesticated (Dierig 2002). The main hydroxy fatty acid in L. fendleri (14-hydroxy-cis-11-eicosenoic acid) could serve as an important partial replacement for castor oil. Additionally, the seed coat of lesquerella contains

a natural gum that could be as valuable as the oil as a component in cosmetics, plastics, lubricants, and coatings, for thickening agents for foods, and for crude oil recovery. The seed meal also has promise as a protein supplement for livestock because it contains 30–35% crude protein (Dierig 1995).

Lesquerella is a perennial plant that is currently being cultivated as a winter annual in Arizona. Seeds are planted in late autumn, and the plants begin to flower in mid-February when daytime temperatures average 22°C and nighttime temperatures average 4°C. Throughout the flowering period, which continues until late May, the plant produces a bright yellow indeterminate flower along an inflorescence. The color yellow is known to be highly attractive to a number of insects, including lygus bugs (Landis and Fox 1972), which are some of our most important economic pests in the Southwest (Ellsworth and Barkley 2001). Normally, at this time of the year, lygus are found in relatively small numbers on weeds such as wild sunflower, Helianthus annuus; alkali heliotrope, Heliotropium curassavicum variety oculatum; London rocket, Sisymbrium irio; and various other mustards (J.B., personal observation). However, as production of lesquerella increases, it may serve as a suitable host

Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the USDA for its use. Purchased by the U.S. Department of Agriculture for official use only. ¹ Deceased.

² Correspondending author, e-mail: John.Byers@ars.usda.gov.

where lygus populations build up before moving into other crops.

Currently, nothing is known about the role lesquerella will play in the seasonal dynamics of *Lygus* spp. in Arizona, but other Brassicaceae (i.e., broccoli and canola) are known to be good hosts for lygus bugs (Snodgrass and McWilliams 1992, Cárcamo et al. 2003). Because lesquerella is positioned to become an important new crop in the Arizona landscape, it will be critical to understand its potential as a source of lygus bugs. Here we examined the *Lygus* spp. complex occurring in lesquerella and the insect-plant interactions during the flowering period. Previously, we determined that, in alfalfa, Lygus spp., primarily Lygus hesperus Knight and Lygus lineolaris (Palisot de Beauvois), responded to a broad range of trap colors (=hue) but were generally collected in higher numbers on blue, green, yellow, and clear sticky traps (Blackmer et al. 2008). Other investigators have found that these species respond best to other colors in different settings; L. hesperus to yellow pan traps in Washington State (Landis and Fox 1972) and L. line*olaris* to white, yellow, or clear traps in apple orchards (Prokopy et al. 1979) or pink traps in peach orchards (Legrand and Los 2003). In the latter two studies, it was proposed that the response was related to the flower color of the host crop. If lygus respond to the color of the flower resource in the crop in which they occur, we would speculate that, in lesquerella, they would show a preference for yellow sticky traps. These variations in response to color would be important to understand as we attempt to develop a mass trapping and/or monitoring system for Lygus spp. (Byers 2007). In addition to examining lygus response to colored sticky traps, we studied the plant factors that could influence host location and acceptance of our most important lygus bug, L. hesperus, by examining their response to lesquerella volatiles in a Y-tube olfactometer. Collection and identification of headspace volatiles of L. fendleri were carried out using Porapak-Q absorbent traps and gas chromatographymass spectrometry (GC-MS), and the major component was tested in the field as a potential attractant.

Materials and Methods

Insect Rearing and Maintenance. Olfactometer bioassays were conducted in the spring of 2006 using L. hesperus that had been maintained in culture on green beans, carrots, pink bollworm eggs [Pectinophora gossypiella (Saunders)], and 10% sucrose solution. To maintain genetic diversity, feral individuals were added to our colony three to four times per year. Food was changed every other day, and the previously used beans and carrots, which also served as oviposition substrates, were placed in 2 by 14-cm-diameter petri dishes lined with filter paper. When nymphs emerged, they were placed in 8.5 by 12.5-cm-diameter paper cartons with nylon organdy lids and maintained in an incubator at $24 \pm 2^{\circ}C$ (SEM), $40 \pm 10\%$ RH, and under a light-dark regimen of 14:10 (L:D) h until needed for bioassays.

Y-tube Olfactometer Studies. The response of male and female *L. hesperus* to flowering *L. fendleri* plants was examined in a Y-tube olfactometer previously described in Blackmer et al. (2004). Filtered air was split between two 2-liter holding chambers: one chamber served as a control (clean air) and the other held the plant material. Airflow through the system was maintained at 4.0 liters/min (3.2 m/min inside the tube) by an inline flow meter (Gilmont Instruments; Barnant, Barrington, IL). Before each trial, light intensity was measured with a light meter (ExTech Instruments Model 401025; Zefon International, St. Petersburg, FL), and it averaged 620.5 \pm 3.4 lux during the bioassays.

Approximately 30 min to 1 h before trials began, 7to 10-d-old, mated adult *L. hesperus* were placed into individual holding/release tubes as described in Blackmer et al. (2004). Tubes containing bugs were placed into a separate holding container, so they would not be exposed to treatments before their release. At the onset of the experiment, each insect was given 5 min to respond, and a choice for the left or right arm of the olfactometer was noted when the insect was 1 cm past the Y junction. The variables recorded were percentage of bugs exiting the holding tube, percentage of bugs walking upwind, time needed to choose between arms, percentage of bugs responding to treatment, and percentage of bugs walking to the extreme end of the Y tube.

In early May, when flowers were in full bloom, intact flowering L. fendleri plants were randomly harvested from the edge of the field by means of a small hand trowel immediately before the bioassay. Their root system was wrapped with moist paper towels and enclosed inside a plastic sleeve before being placed into the holding chamber. These plant samples were most similar, in terms of handling and environmental conditions, to in situ whole plant samples that are mentioned below (see Identification of Headspace Volatiles of L. fendleri). Plant material was replaced every hour, and treatment presentation was switched to the opposite arm of the Y tube every 10 runs to eliminate potential bias caused by odor source location. Bioassays were conducted between 1000 and 1400 hours, a time when floral volatiles from fieldcollected lesquerella are abundant. Each insect was tested only once using a clean Y tube for each trial. On any given day, 20 individuals were tested. Tests continued until the response of 40 male and 40 female bugs had been evaluated. Mean temperature and relative humidity during the assays were $23.8 \pm 0.1^{\circ}$ C and $25.7 \pm 0.8\%$, respectively.

Lygus spp. Composition and Color Preference in L. fendleri. In 2007, trials were conducted in May (7, 17, and 23) when flowers were in full bloom and lygus populations had reached adequate levels for our tests. Lesquerella fields were located on the University of Arizona, Maricopa Agricultural Center, Maricopa, AZ. Experiments were set up in a 2-m-wide cleared area that extended the length of the field and ran between two large (1.9 acres each) benches of lesquerella. Before each trial, four sets of 25 sweeps using a 38cm-diameter by 84-cm heavy duty sweep net (7212CM; BioQuip Products, Gardena, CA) were made to determine species composition, density, and sex ratios of resident *Lygus* spp.

Response to visual cues (hue) was examined in lesquerella using sticky traps that were fabricated from 0.02-mm-thick rigid vinyl plastic sheeting purchased from GE Polymershapes (Phoenix, AZ). Trap colors tested included yellow (translucent and opaque), red (as an unattractive visual control for lygus, see Blackmer et al. 2008), blue, green, or clear. Reflectance spectra, hue, saturation, and luminosity values of these colors were previously described in Blackmer et al. (2008). All traps were translucent, except for the opaque yellow traps, which were cut from high-density plastic sheeting (cat. no. 01-4000-1; Hummert International, Earth City, MO). This particular color was included in the test because it has been shown to be highly attractive to several homopteran groups (Hardie 1989, Todd et al. 1990). Traps were cut into 60 by 30-cm rectangles and hand rolled on one side with a thick coating of Pestick adhesive (Hummert International).

To facilitate the hanging of traps, a 1-cm-diameter hole was punched into each of the four corners of the rectangles. Traps were hung from 1-cm-diameter screws that had been screwed into 90-cm-tall wooden stakes. The top screw was placed 11 cm from the top of the stake, and the bottom screw was placed 25 cm below the top screw. The stake was pounded into the ground, and a sticky trap was placed onto the screws and wrapped around the stake to form an 18-cmdiameter cylinder, where the bottom of the cylinder was 20 cm above the ground. At this height, the trap bottom was slightly above canopy height. In all trials, traps were placed 10 m apart. Traps were set out in a randomized complete block design (RCB; N = 4) and collected after 5 d. Lygus bugs were counted, sexed, and identified using the diagnostic keys provided in Mueller et al. 2003. To evaluate nontarget effects, the number of green lacewing, Chrysoperla carnea (Stephans) (Neuroptera: Chrysopidae), Hippodamia spp. (Coleoptera: Coccinellidae), Collops spp. (Coleoptera: Melyridae), and false chinch bugs, Nysius raphanus Howard (Heteroptera: Lygaeidae), were also determined. False chinch bugs were extremely common during this study, and consequently, their numbers were estimated by counting the number found on three randomly selected 4-cm² sections of each trap. These counts were averaged and multiplied by 112.5 (trap area/sample area or 1,800/16 cm²).

Identification of Headspace Volatiles of *L. fendleri*. Flowering *L. fendleri* plants were sampled for volatiles. Collections from production fields located in Maricopa, AZ, were made between 0830 and 1600 hours from 26 April to 10 May 2006. For in situ whole plant sampling, an entire plant was enclosed inside a clear plastic oven bag (482 by 596 mm; Reynolds, Richmond, VA) with the opening wrapped tightly about the stems and fastened with wire. A typical plant at this time of year would contain ~150 inflorescents. For the cut plant material, a set of stems from about eight

plants or stems from one whole plant were cut near ground level and taken to the laboratory and similarly enclosed within an oven bag so that any cut ends were exposed outside the bag. In some cut material, the stems were deflowered and enclosed inside oven bags. Pressurized air from a vacuum pump was filtered by a 120-ml activated charcoal trap (Alltech Associates, Deerfield, IL) and passed through 0.125-in OD Teflon tubing to empty inside the oven bag containing the stems. Volatiles released in the interior of the bag were transferred by Teflon tubing to a 20 by 2.3-mm ID plug of Porapak Q (80/100 mesh; Alltech Associates) held between plugs of glass wool inside the Teflon tubing connected on both sides by Parker brass fittings (Alltech Associates). The air exiting the Porapak adsorbent passed through a flow meter at 950 ml/min to a vacuum pump. Before volatile collections, the air was passed through the system for 15 min to remove background organics. After this, the Porapak plug was installed, and volatiles were collected for 30 min. In preliminary work, no breakthrough of volatiles was observed with a second Porapak plug after 60-min collections. The Porapak plug, with the "downstream" Parker nut, was removed from the system, and a glass tube (25 mm by 0.25 in OD) was fitted to the Parker nut/plug to allow two washings of 100 μ l of hexane with ethyl heptanoate and/or (+)-carvone internal standards $(1 \text{ ng}/\mu l)$ to pass through the adsorbent and wash volatiles into a vial for subsequent GC-MS analysis. The Porapak Q traps were cleaned by passing three 200- μ l volumes of pentane through the plugs and heated at 150°C while passing hydrocarbon-trapped N₂ through the plugs for 5 min. Repeated cleaning of the Porapak plugs reduced levels of a few trace organics that were ignored in analyses.

Chemicals in the hexane washes of the Porapak plugs were separated on a Varian 3800 GC (Varian, Palo Alto, CA) with a fused-silica capillary column (Cyclodex-B; JW Scientific) of 60 m by 0.25 mm ID coated with 0.25 µm permethylated B-cyclodextrin and He carrier gas at a flow rate of 1.2 ml/min. The injector was split-less for 0.75 min and then 60:1 split for 5 min and thereafter 20:1. A Varian CP-8400 autosampler was used for $1-\mu$ l sample injections at 250°C. The temperature program was 40°C for 2 min, then 30°/min to 98°C and held for 15 min, then 3°/min to 125°C, and then 10°/min to 250°C and held 10 min. Chemical standards as shown in Table 1 were purchased from Sigma-Aldrich (St. Louis, MO), and all were >98% purity. Chemical identification of the GC effluents was performed by a Varian Saturn 2000 MS using the NIST02 (National Institute of Standards) spectral database and spectra of the standards. For quantification of volatile solvent washes, the total ion chromatogram (TIC) peak areas at the respective retention times were compared with areas of the internal standard (ethyl heptanoate) and adjusted for the MS response factor sensitivities of identified compounds.

Color Plus Major Volatile Component from *L. fendleri*. Previously, we determined that visual cues in combination with volatile plant cues often act in an

Compound	$\frac{\text{Whole plant}^{a}}{(\text{ng/plant/h})}$ Flowering (N = 5)	Multiple plant stems b (ng/g tissue/h)		Cut plant ^c (ng/g tissue/h)
		Flowering $(N = 3)$	De-flowered $(N = 3)$	Flowering $(N = 6)$
Isopropyl isothiocyanate	138 ± 61	2.84 ± 0.39	59.37 ± 15.17	2.22 ± 0.7
E-2-hexenal	0	0.37 ± 0.19	4.84 ± 1.48	1.29 ± 0.4
α-pinene	58 ± 15	0.08 ± 0.04	0.11 ± 0.08	0.06 ± 0.02
Methoxybenzene	20 ± 5	0.07 ± 0.04	0.16 ± 0.08	0.11 ± 0.04
Z-3-hexenol	210 ± 12	1.10 ± 0.07	14.17 ± 2.02	1.88 ± 0.76
Butyl isothiocyanate	0	0	2.4 ± 2.4	0.09 ± 0.05
E-β-ocimene	69 ± 32	0.12 ± 0.02	0	0.66 ± 0.15
Z-3-hexenyl acetate	332 ± 223	1.31 ± 0.37	10.52 ± 4.64	2.9 ± 1.24
6-methyl-5-hepten-2-one	21 ± 11	0.15 ± 0.07	0.12 ± 0.12	0.87 ± 0.28
Benzaldehyde	801 ± 221	2.26 ± 0.92	0.14 ± 0.14	7.19 ± 1.04
Phenylacetaldehyde	3632 ± 1165	23.13 ± 14.9	2.11 ± 1.03	108.23 ± 20.32
Linalool	169 ± 133	0.07 ± 0.04	0	0.24 ± 0.09
Z-3-hexenyl butyrate	12 ± 10	0.03 ± 0	0.17 ± 0.04	0.03 ± 0.02
Benzyl alcohol	61 ± 28	0.21 ± 0.12	0.07 ± 0.07	1.37 ± 0.13
Methyl salicylate	88 ± 26	0.2 ± 0.03	0.02 ± 0.02	0.49 ± 0.06
Benzothiozole	37 ± 12	0.06 ± 0.03	0.03 ± 0.01	0.14 ± 0.06

Table 1. Release rates of volatiles from whole plants in field or cut stems with flowers or flowers removed (de-flowered) in laboratory (ng \pm SEM per plant or gram fresh weight)

^a Volatiles collected from intact plant in field.

^b Each sample (N = 3) consisted of about eight plants.

^c Whole plant taken to laboratory for volatile collection.

additive or synergistic manner to attract L. hesperus (Blackmer and Cañas 2005). To examine this in lesquerella, we used the two most attractive colored sticky traps identified from the color preference trial listed above and combined them with the dominant plant compound identified in the study listed above. Blue and green traps, with or without slow-release dispensers, were hung as described above on 90-cmtall stakes and tested in the field during May 2007. The slow-release dispensers consisted of 0.3-ml polypropylene high-performance liquid chromatography (HPLC) vials (Dionex no. 055428), with 300 µl of neat PAA (95% purity, Cas. #122-78-1; ICN Biomedicals, Aurora, OH). The vials were covered with parafilm and had a 5-µl micropipette (Drummond Scientific Company, Broomall, PA) inserted into the center. The micropipette reached the bottom of the centrifuge tube and extended 2 mm above the top edge of the tube. These dispensers were attached with twist ties to the center of the sticky traps on the downwind side, so that insects could more easily orient upwind to the potential attractant. Traps were placed in the cleared area between two 1.9-acre lesquerella benches. Previously, Blackmer et al. (2008) determined that lygus were trapped in higher numbers when traps were placed over bare ground between benches of alfalfa. In all trials, traps were placed 10 m apart. Traps were set out in a randomized complete block design (RCB; N = 12) and collected after 6 (green traps) or 7 d (blue traps). Lygus bugs were counted, sexed, and identified using the diagnostic keys provided in Mueller et al. 2003. To evaluate nontarget effects, the number of C. carnea, Hippodamia spp., and Collops spp. (Coleoptera: Melyridae) were also counted. False chinch bugs were not counted during these trials because numbers had declined.

Statistical Analysis. For olfactometer bioassays, the null hypothesis that *L. hesperus* showed no preference

for either olfactometer arm (a response equal to 50:50) was analyzed with a χ^2 goodness-of-fit test. For experiments on colored traps and PAA-baited green or blue traps, counts were analyzed by analysis of variance (ANOVA) using SigmaStat Software (version 3.5). Counts for *Lygus* spp. (lygus males and females, *L. elisus*, *L. lineolaris*, and *L. hesperus*), predators, and false chinch bugs were analyzed separately by ANOVA. Counts were transformed by $\sqrt{(y + 0.5)}$ when needed to meet the assumptions of normality and homogeneity of variance. When *F*-statistics were significant, means were separated by the Fisher least significant difference (LSD) method.

Results

Y-tube Olfactometer Studies. Females showed a significant preference to lesquerella volatiles over humidified blank air controls ($\chi^2 = 3.98, P < 0.05$). Within the 5 min allowed for a response, 88% of the females exited the holding tube, 97% of these individuals walked upwind, and 74% of these chose the arm with the flowering lesquerella. This choice was made fairly soon after beginning the trial $(90.6 \pm 11.8 \text{ s})$. Of those individuals that chose the side with plant volatiles, 42% went all the way to the end of the tube, whereas only 5% went all the way to the end on the control side of the Y tube. Males, however, did not exhibit a preference to lesquerella volatiles ($\chi^2 = 0.53$, P > 0.05). This is despite the fact that 98% exited the holding tube and 90% walked upwind, choosing an arm in about the same amount of time $(100.7 \pm 13.2 \text{ s})$ as females. Approximately 25% of the males that made a choice went all the way to the end of the tube on the side with lesquerella volatiles, whereas 42% went all the way to the end on the control side.

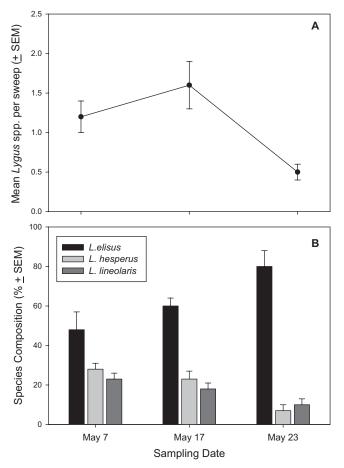


Fig. 1. (A) Mean number (\pm SEM) of *Lygus* spp. per sweep in *Lesquerella fendleri* and (B) percentage (\pm SEM) of *L. elisus*, *L. hesperus*, and *L. lineolaris* in sweeps in May 2007.

Lygus spp. Composition and Color Preference in L. fendleri. Three species of lygus bugs were common in lesquerella: Lygus elisus Van Duzee, L. lineolaris, and *L. hesperus*. When plants were flowering in early to mid-May, the number of Lygus spp. per sweep was 1.47 ± 0.20 , and the sex ratio was 1.3:1.0 (male:female), but as plants began to senesce in late May, numbers decreased to 0.45 ± 0.09 per sweep, and the sex ratio was 1.0:1.0 (Fig. 1A). L. elisus was the dominant species throughout these trials, representing between 49 and 81% of the individuals collected. L. lineolaris and L. hesperus each accounted for $\approx 18\%$ of the individuals collected by sweep net (Fig. 1B); however, as plants began to senesce in late May, L. elisus represented a higher percentage of the Lygus spp. collected. Although their numbers decreased at the end of the growing season, there was only a 57% reduction in L. elisus numbers, whereas L. hesperus and L. lineolaris experienced a 91 and 79% reduction, respectively; perhaps indicating an earlier emigration out of the crop to other suitable hosts. At this time, both L. *lineolaris* and *L. hesperus* were found in fairly high numbers on wild sunflowers located on the edges of the lesquerella field (J.L.B., personal observation).

Trap color played a significant role in the catch of *Lygus* spp. (F = 2.82; df = 5.15; P = 0.05). More *Lygus* spp. were trapped on blue and green traps than on red traps. Seventy percent of the bugs trapped were L. elisus, 21% were L. lineolaris, and only 9% were L. hesperus. When species and sex were analyzed separately, responses to trap color varied. L. elisus was captured more often on blue, green, and clear traps than on red traps (F = 3.75; df = 5,15; P = 0.02; Fig. 2A). L. hesperus was captured more often on green and clear traps than on yellow (opaque) and red traps (F = 3.26; df = 5,15; P = 0.03). L. lineolaris was captured more often on yellow (opaque and translucent) and green traps than on clear or red traps (F = 3.14; df = 5,15; P =0.04). Although 69% of the bugs captured were males, high variability in response led to a lack of separation relative to color (Fig. 2B; F = 2.50; df = 5,15; P = 0.08), whereas females were captured more often on green, clear, yellow (translucent), and blue traps than on red traps (F = 3.96; df = 5,15; P = 0.02).

False chinch bugs exhibited a significant attraction to trap color (Fig. 3A; F = 2.99, df = 5,15, P = 0.05),

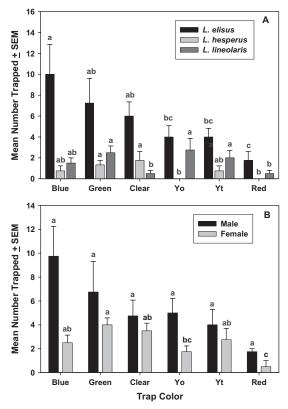


Fig. 2. (A) Mean number (\pm SEM) of *L. elisus*, *L. lineolaris*, and *L. hesperus* and (B) mean number (\pm SEM) of female and male *Lygus* spp. captured on colored sticky traps in *L. fendleri*. Means within a species with the same lowercase letters are not significantly different (Fisher LSD at *P* > 0.05). Yo, yellow opaque; Yt, yellow translucent.

with more captured on clear and blue than on yellow (opaque) and red traps. Of the beneficial insects, *Hippodamia* spp. showed a preference to green, clear, yellow (translucent), and blue over yellow (opaque) and red traps (Fig. 3B; F = 9.60, df = 5,15, P < 0.001), but *C. carnea* showed no significant preference to trap color (F = 2.06, df = 5, 15, P = 0.13). *Collops* spp. numbers were too low to analyze during this particular trial.

Identification of Headspace Volatiles of *L. fendleri*. All plants and stems had numerous yellow flowers (RGB: 243, 255, 95; Byers 2006) that released phenylacetaldehyde (PAA) in the largest amounts of any volatile (Table 1). Other flower-derived volatiles, in order of amount, were benzaldehyde, benzyl alcohol, linalool, methyl salicylate, and *E*- β -ocimene. When flowers on stems were removed, there was a drastic reduction in amounts of flower volatiles, with the exception of linalool, whereas some compounds such as isopropyl isothiocyanate, *E*-2-hexenal, *Z*-3hexenol, butyl isothiocyanate, *Z*-3-hexenyl acetate, and *Z*-3-hexenyl butyrate increased in amounts and may have been induced by the damage associated with flower removal (Table 1). Large differences in release

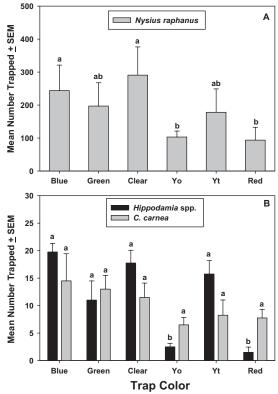


Fig. 3. (A) Mean number $(\pm \text{SEM})$ of *N. raphanus* and (B) mean number $(\pm \text{SEM})$ of *Hippodamia* spp. captured on colored sticky traps in *L. fendleri*. Means within a species with the same lowercase letters are not significantly different (Fisher LSD at P > 0.05). Yo, yellow opaque; Yt, yellow translucent.

rates between whole plant collections in the field and whole plant cut collections in the laboratory were also noted, but the three major components, phenylacetaldehyde, benzaldehyde and Z-3-hexenyl acetate, always accounted for 84–92% of the headspace volatiles.

Color Plus Major Volatile Component from L. fendleri. On green traps, the total Lygus spp. captured was not significantly different between unbaited and PAA-baited traps (F = 1.05, df = 1,22, P = 0.32). The numbers of Hippodamia spp. and C. carnea were also not significantly different on baited and unbaited traps (F = 2.03, df = 1,22, P = 0.17 and F = 0.03, df = 1,22,P = 0.87, respectively). However, the number of Collops spp. were significantly higher on green traps baited with PAA versus unbaited green traps (3.92 \pm 0.84 versus 1.33 ± 0.33 ; F = 11.32, df = 1,22, P = 0.003). On blue traps, which were tested subsequent to green traps when plants were beginning to senesce and populations of all insects had decreased, the total Lygus spp. captured was not significantly different between unbaited and PAA-baited traps (F = 0.76, df = 1,22, P = 0.39), nor were the numbers of *Hippodamia* spp., C. carnea, and Collops spp. different on baited and unbaited traps (P > 0.05 for all three groups).

Discussion

Lesquerella fendleri represents a promising new agricultural crop in the Southwest (Dierig et al. 1992, Dierig 1995). Its bright yellow flowers may be highly attractive to a number of herbivores (Prokopy and Owens 1983), some of which are economic pests. Yellow is known to be attractive to some of our most important cotton pests, lygus bugs (Landis and Fox 1972); however, because lesquerella is currently grown early in the season, well before cotton emerges, the lygus bugs emigrating from lesquerella would most likely be immigrating to alfalfa or weed hosts and not directly to cotton. Regardless of which crop they move to, as lesquerella begins to increase in acreage, the potential for larger numbers of lygus bugs early in the season could add to management problems of these pests.

Here we determined that three species of lygus were commonly found in L. fendleri; however, the most prevalent species in 2007, L. elisus, is not usually found in large numbers in cotton during the growing season. In canola, a crop closely related to lesquerella, L. elisus was found to be the dominant Lygus spp. (Demirel and Cranshaw 2006). Our two most important economic species, L. hesperus and L. lineolaris, were much less common in lesquerella in 2007. However, in a similar survey conducted in May 2008, whereas overall numbers of lygus bugs were similar to numbers found in 2007, L. hesperus and L. lineolaris accounted for >80% of the individuals collected (J.L.B., unpublished data). All three of these species of lygus bugs damage their hosts by feeding on meristematic tissue and represent a potential threat to this new crop. Whether these lygus bugs will impact lesquerella production is an open question.

Similar to a previous study (Blackmer et al. 2008), we found that lygus exhibit a fairly broad response to trap color. All three species were captured less frequently on red traps than on other colored traps. L. elisus and L. hesperus generally preferred green, blue, and clear traps, whereas L. lineolaris showed a preference for yellow and green traps. This response to yellow by L. lineolaris was not observed in a previous trial in alfalfa (Blackmer et al. 2008). Other studies with plant bugs have shown attraction to various colors, but the response was species specific, and in some cases, habitat dependent. Landis and Fox (1972) found that significantly more L. hesperus and L. elisus were trapped in light orange-yellow and deep chrome-yellow pans than in green, red, or pink water trap pans. For L. lineolaris, Prokopy et al. (1979) found that non-UV-reflecting gloss white captured significantly greater numbers than yellow, green, orange, blue, red, aluminum foil, black, and lead-white rectangles. This response was thought to be because of the color of the flowers in the apple orchard. In peach orchards, Legrand and Los (2003) found that L. line*olaris* was captured in higher numbers on pink sticky traps than on white traps. The pink trap closely mimicked the color of peach flower petals, and this was thought to play a role in their preference for pink traps. We initially speculated that if lygus bugs responded to flower-specific colors then yellow would be the preferred color in lesquerella. Only *L. lineolaris* showed a response that may indicate a flower-specific response, but this would need to be verified under controlled laboratory conditions. Such a response could enhance foraging behaviors, but of the species examined here, only *L. lineolaris* appeared to exhibit such behaviors.

In laboratory studies, we determined that female *L*. *hesperus*, our main cotton pest, responded to plant volatiles associated with flowering lesquerella by moving upwind and selecting the treatment arm of the Y tube. Males did not exhibit a significant attraction to lesquerella volatiles. This response is similar to previous studies, where Blackmer et al. (2004) showed that female *L. hesperus* were more likely to show a preference to alfalfa volatiles in a Y-tube olfactometer than were males. In previous studies where the antennal responses of male and female bugs to plant compounds were examined using the electroantennagram (EAG) technique, the EAG responses of female *L. lineolaris* and Lygocoris pabulinus L. were far more pronounced to plant compounds (green leaf volatiles and monoterpenes) than the antennal responses of male bugs (Chinta et al. 1994, Groot et al. 1999).

Many flowering plants have evolved complex floral bouquets with the most obvious purpose being to attract pollinators; however, detrimental herbivores may also exploit these communication systems (Theis 2006). Specialist herbivores, such as those that feed on Brassicaceae, exploit scents to locate their host (Renwick 2002). For example, the specialist pollen beetle, *Meligethes aeneus*, cues into isothiocyanates to locate its cruciferous host plant, *Brassica napus* (Smart and Blight 2000). Although it is thought to be less common, attraction to plant volatiles by generalist insects has also been shown in Coleoptera and Lepidoptera (Bernays and Chapman 1994), and it is likely to exist in other orders of insects as well.

The majority of plant volatiles that are involved in insect attraction fall within one of the following categories: terpenoids, fatty acid derivatives, benzenoids, and nitrogen-containing compounds, and although each plant species has a unique volatile profile, there is often considerable overlap between species (Jönsson 2005). The volatile profile of lesquerella included a blend of 16 compounds that included all of the categories mentioned above. Of the flower-specific compounds, PAA was dominant. This particular flowerspecific compound has been found to enhance upwind flights of fall armyworm adults, Spodoptera frugiperda, to its sex pheromone (Meagher and Mitchell 1998), was found to be highly attractive to a number of pollinators and folivores of Canada thistle, Cirsium arvense (Theis 2006), and increased trap capture of C. carnea by 10–100 times that of unbaited traps (Tóth et al. 2006). Here we could find no significant attraction to this single dominant component of lesquerella by *Lygus* spp. Because we used whole flowering lesquerella that was harvested immediately before our olfactometer bioassays, the upwind response of female lygus bugs is likely caused by the flower-specific plant components, but additional studies will be needed to determine which components are most important. It is likely to be a more complex blend of the 16 identified compounds.

Of the natural enemies examined here only *Collops* spp. on green traps exhibited a significant attraction to PAA. In Italy and Hungary, C. carnea exhibited a significant attraction to PAA (Tóth et al. 2006), but we found no such response here. The attraction of *Collops* spp. to PAA that was observed here might allow us to recruit and/or better retain this beneficial insect and has the potential to improve biological control and integrated pest management (IPM) programs. Previously, Flint et al. (1981) found that male Collops vittatus beetles were attracted to the plant volatile, caryophyllene alcohol, and that C. carnea was attracted to caryophyllene (Flint et al. 1979). Studies using induced plant volatiles, such as methyl salicylate, have shown significantly enhanced populations of a number of natural enemies (James and Price 2004, James 2005, 2006, Zhu and Park 2005). These findings taken together suggest a tremendous potential for using plant volatiles to enhance biological control in lesquerella and potentially other crops.

This study represents the first study to examine the *Lygus* spp. complex of a new and potentially important seed oil crop for the state of Arizona. *L. fendleri* was a good reproductive and food host for *Lygus* spp., with adult and nymphal populations reaching ≈ 1.5 and 1.3 per sweep sample, respectively. As this crop increases in acreage, it will likely serve as an important source of lygus bugs and may make this pest even more difficult to manage in crops grown later in the season. A study of the potential of lesquerella as a source of lygus bugs for crops grown later in the season is needed to provide us with a better idea as to just how important its role will be in the Arizona landscape.

Acknowledgments

We thank T. Steele for help with the Y-tube olfactometer setup, L. Jech, L. Elhoff, and L. Lee for maintaining plant and insect cultures, L. Jech, M. Noble, L. Lee, and W. Price for help with behavioral studies, L. Elhoff for assistance in collecting plant volatiles, and the University of Arizona at Maricopa Agricultural Center and Dave Dierig for providing study sites. For reviews of previous versions of this manuscript, we thank D. Dierig, L. Williams III, L. Jech, S. Naranjo, and two anonymous reviewers for helpful suggestions.

References Cited

- Bernays, E. A., and R. F. Chapman. 1994. Host-plant selection by phytophagous insects. Chapman & Hall, London, United Kingdom.
- Blackmer, J. L., and L. A. Cañas. 2005. Visual cues enhance the response of *Lygus hesperus* (Heteroptera: Miridae) to volatiles from host plants. Environ. Entomol. 34: 1524– 1533.
- Blackmer, J. L., C. Rodriguez-Saona, J. A. Byers, K. L. Shope, and J. P. Smith. 2004. Behavioral response of Lygus hes-

perus to conspecific and headspace volatiles of alfalfa in a Y-tube olfactometer. J. Chem. Ecol. 30: 1529–1546.

- Blackmer, J. L., J. A. Byers, and C. Rodriguez-Saona. 2008. Evaluation of color traps for monitoring *Lygus* spp.: Design, placement, height, time of day, and non-target effects. Crop Protect. 27: 171–181.
- Byers, J. A. 2006. Analysis of insect and plant colors in digital images using Java software on the Internet. Ann. Entomol. Soc. Am. 99: 865–874.
- Byers, J. A. 2007. Simulation of mating disruption and mass trapping with competitive attraction and camouflage. Environ. Entomol. 36: 1328–1338.
- Cárcamo, H. A., J. Otani, J. Gavloski, M. Dolinski, and J. Soroka. 2003. Abundance of *Lygus* spp. (Heteroptera: Miridae) in canola adjacent to forage and seed alfalfa. J. Entomol. Soc. Br. Columbia 100: 55–63.
- Chinta, S., J. C. Dickens, and J. R. Aldrich. 1994. Olfactory reception of potential pheromones and plant odors by tarnished plant bug, *Lygus lineolaris* (Hemiptera: Miridae). J. Chem. Ecol. 20: 3251–3267.
- Demirel, N., and W. Cranshaw. 2006. Surveys of Lygus spp. and their movement on cultivated crops and non-cultivated habitats throughout growing season in Colorado. Pakis. J. Biol. Sci. 9: 197–200.
- Dierig, D. A. 1995. Lesquerella. New Crop FactSheet. (http://www.hort.purdue.edu/newcrop/cropfactsheets/ Lesquerella.html).
- Dierig, D. A. 2002. Lesquerella. (http://www.nnfcc.co.uk/ cropinfo/lesquerella.pdf).
- Dierig, D. A., A. E., Thompson, and F. S. Nakayama. 1992. Lesquerella commercialization efforts in the United States. Indust. Crop Prod. 1: 289–293.
- Ellsworth, P. C., and V. Barkley. 2001. Cost-effective Lygus management in Arizona cotton, pp. 299–307. In J. C. Silvertooth (ed.), Cotton, a College of Agricultural and Life Sciences Report. University of Arizona, College of Agriculture and Life Sciences, Tucson, AZ.
- Flint, H. M., S. S. Salter, and S. Walters. 1979. Caryophyllene: an attractant for the green lace-wing. Environ. Entomol. 8: 1123–1125.
- Flint, H. M., J. R. Merkle, and M. Sledge. 1981. Attraction of male *Collops vittatus* in the field to caryophyllene alcohol. Environ. Entomol. 10: 301–304.
- Groot, A. T., R. Timmer, G. Gort, G. P. Lelyveld, F. P. Drijfhout, T. A. Van Beek, and J. Hans Visser. 1999. Sex-related perception of insect and plant volatiles in Lygocoris pabulinus. J. Chem. Ecol. 25: 2357–2371.
- Hardie, J. 1989. Spectral specificity for targeted flight in the black bean aphid, *Aphis fabae*. J. Insect Physiol. 35: 619– 626.
- James, D. G. 2005. Further field evaluation of synthetic herbivore-induced plant volatiles as attractants for beneficial insects. J. Chem. Ecol. 31: 481–495.
- James, D. G. 2006. Methyl salicylate is a field attractant for the goldeneyed lacewing, *Chrysopa oculata*. Bioconserv. Sci. Tech. 16: 107–110.
- James, D. G., and T. S. Price. 2004. Field-testing of methyl salicylate for recruitment and retention of beneficial insects in grapes and hops. J. Chem. Ecol. 30: 1613–1628.
- Jönsson, M. 2005. Responses to oilseed rape and cotton volatiles in insect herbivores and parasitoids. PhD thesis, Swedish University of Agricultural Sciences, Alnarp, Sweden.
- Landis, B. J., and L. Fox. 1972. Lygus bugs in eastern Washington: color preference and winter activity. Environ. Entomol. 1: 464–465.

- Legrand, A., and L. Los. 2003. Visual responses of Lygus lineolaris and Lygocoris spp. (Hemiptera: Miridae) on peaches. Fla. Entomol. 86: 424–428.
- Meagher, R. I., Jr., and E. R. Mitchell. 1998. Phenylacetaldehyde enhances upwind flight of male fall armyworm (Lepidoptera: Noctuidae) to its sex pheromone. Fla. Entomol. 8: 556–559.
- Mueller, S. C., C. G. Summers, and P. B. Goodell. 2003. A field key to the most common *Lygus* species found in agronomic crops of the Central San Joaquin Valley of California. University of California, Agriculture & Natural Resources, Oakland, CA.
- Prokopy, R. J., and E. O. Owens. 1983. Visual detection of plants by herbivorous insects. Annu. Rev. Entomol. 28: 337–364.
- Prokopy, R. J., R. G. Adams, and K. J. Hauschild. 1979. Visual responses of tarnished plant bug adults on apple. Environ. Entomol. 8: 202–205.
- Renwick, J.A.A. 2002. The chemical world of crucivores: lures, treats and traps. Entomol. Exp. Appl. 104: 35–42.
- Roetheli, J. C., K. D. Carlson, R. Kleiman, A. E. Thompson, D. A. Dierig, L. K. Glaser, M. G. Blasé, and J. Goodell. 1991. Lesquerella as a source of hydroxy fatty acids for industrial products. An assessment. U.S. Department of Agriculture/CSRS Office of Agricultural Materials, Washington, DC.

- Smart, L. E., and M. M. Blight. 2000. Response of the pollen beetle, *Meligethes aeneus*, to traps baited with volatiles from oilseed rape, *Brassica napus*. J. Chem. Ecol. 26: 1051– 1064.
- Snodgrass, G. L., and J. M. McWilliams. 1992. Rearing the tarnished plant bug (Heteroptera: Miridae) using a tissue paper oviposition site. J. Econ. Entomol. 85: 1162–1166.
- Theis, N. 2006. Fragrance of Canada thistle (*Cirsium arvense*) attracts both floral herbivores and pollinators. J. Chem. Ecol. 32: 917–927.
- Thompson, A. E., and D. A. Dierig. 1994. Initial selection and breeding of *Lesquerella fendleri*, a new industrial oilseed, Industr. Crops Prod. 2: 97–106.
- Todd, J. L., M. O. Harris, and L. R. Nault. 1990. Importance of color stimuli in host-finding by *Dalbulus* leafhoppers. Entomol. Exp. Appl. 84: 245–255.
- Tóth, M., A. Bozsik, F. Szentkiralyi, A. Letardi, M. R. Tabilio, M. Verdinelli, P. Zandigiacomo, J. Jekisa, and L. Szarukan. 2006. Phenylacetaldehyde: a chemical attractant for common green lacewings (*Chrysoperla carnea s.l.*, Neuroptera: Chrysopidae). Eur. J. Entomol. 103: 267–271.
- Zhu, J., and K. C. Park. 2005. Methyl salicylate, a soybean aphid-induced plant volatile attractive to the predator *Coccinella septempunctata*. J. Chem. Ecol. 31: 1733–1746.

Received 30 October 2007; accepted 30 July 2008.