Species Composition, Monitoring, and Feeding Injury of Stink Bugs (Heteroptera: Pentatomidae) in Blackberry

Author(s): S. A. Brennan , O. E. Liburd , J. E. Eger , and E. M. Rhodes

Source: Journal of Economic Entomology, 106(2):912-923. 2013.
Published By: Entomological Society of America
URL: http://www.bioone.org/doi/full/10.1603/EC12415

Species Composition, Monitoring, and Feeding Injury of Stink Bugs (Heteroptera: Pentatomidae) in Blackberry

S. A. Brennan,[1,2] O. E. Liburd,[1] J. E. Eger,[3] and E. M. Rhodes[1]

J. Econ. Entomol. 106(2): 912–923 (2013); DOI: http://dx.doi.org/10.1603/EC12415

ABSTRACT Blackberry (*Rubus* spp.) production in Florida has increased >100% within the past two decades. and several insect pests, including stink bugs (Heteroptera: Pentatomidae), have been observed feeding on this crop. The objectives for this study were to determine the stink bug species present in blackberry; to develop monitoring tools for stink bugs in blackberry; and to describe feeding injury to blackberries by *Euschistus quadrator* Rolston, a relatively new stink bug pest to Florida, that has spread throughout the state. In a field survey, *E. quadrator* was the most abundant stink bug species, followed by *Euschistus servus* Say, *Euschistus obscurus* (Palisot de Beauvois), *Thyanta custator* (F.), *Proxys punctulatus* (Palisot de Beauvois), and *Podisus maculiventris* Say. Yellow pyramid traps caught more stink bugs than tube traps with or without the addition of *Euschistus* spp. pheromone lures. There were no statistical differences between traps baited with a Trécé Pherocon Centrum lure, a Suterra Scenturion lure, and an unbaited trap. These results were supported by Y-tube olfactometer assays with *E. quadrator* where there were no differences between pheromone baited lures and a control. Injury to berries caused by *E. quadrator* adults and third instars was similar, and both adults and third instars fed more on green berries compared with turning berries. In addition, adults fed more on green berries compared with ripe fruit. The most common injury to green berries was discoloration. In contrast, misshapen drupelets were commonly seen on turning and ripe berries. The potential for managing stink bugs in blackberries to prevent them from reaching damaging levels is discussed.

KEY WORDS blackberry, stink bug, *Euschistus* spp., injury, monitoring

Several blackberry (*Rubus* spp.) cultivars have recently been developed to be more tolerant to the climatic conditions of the southern United Sates, possibly opening this region to further commercial blackberry production (Jennings et al. 1991, Moore 1997). Georgia has tripled its production in the past 10 yr, with 122 ha of blackberries harvested in 2007 (Strik et al. 2007, USDA–NASS 2009). Similarly, Florida blackberry production has increased fivefold within the past 5 yr (O.E.L., unpublished data). Earlier ripening of blackberries in Florida may enable growers to meet a market window not currently being filled by other southern states.

Many pest species present in other geographical areas also affect blackberries in the southeast. Southern green stink bug, *Nezara viridula* (L.); green stink bug, *Acrosternum hilare* (Say); and *Euschistus* spp. have been reported as pests of blackberries in the southeastern United States (Johnson and Lewis 2003, Anonymous 2008, Mizell 2008). The brown stink bug, *Euschistus servus* (Say), is also known to cause injury to other *Rubus* spp. (Maxey 2011).

The genus *Euschistus* not only contains *E. servus* but also other pest species in the continental United States, including *Euschistus variolarius* (Palisot de Beauvois), *Euschistus tristigmus* (Say), *Euschistus ictericus* (L.), *Euschistus quadrator* Rolston, and *Euschistus conspersus* (Uhler). *E. quadrator* and smaller species in the genus, such as *Euschistus obscurus* (Palisot de Beauvois), are sometimes referred to as the "lesser brown stink bug complex" due to their increasing pest status (Hopkins et al. 2005). Members of the genus *Euschistus* have been observed feeding on many cultivated crops, including corn, *Zea mays* L.; cotton, *Gossypium hirsutum* (L.); alfalfa, *Medicago sativa* L.; soybean, *Glycine max* (L.) Merr.; and various fruits (McPherson and McPherson 2000). These species have migrated south, and increased in pest status in the southeastern United States in recent years. The introduction of crops such as Bt cotton in the south, which does not control stink bugs, and reduced application of broad-spectrum insecticides in favor of more selective pesticides may have contributed to the increase in numbers of *Euschistus* spp. in southeastern plantings. In addition to stink bugs, thrips (*Frankliniella* spp.); twospotted spider mites, *Tetranychus urticae* Koch; and gall midges (*Dasineura* spp.) have been listed as pests of concern for Florida (Ellis et al. 1991, Mizell 2007; O.E.L. et al., unpublished data).

[1] Department of Entomology and Nematology, University of Florida, Bldg. 970 Natural Area Dr., Gainesville, FL 32611.
[2] Corresponding author, e-mail: sbrennan@ufl.edu.
[3] Dow AgroSciences LLC, 2606 S. Dundee St., Tampa, FL 33629.

Stink bugs are highly mobile and polyphagous pests, thereby making monitoring difficult (McPherson and McPherson 2000). Visual searches, ground cloths, blacklight traps, fruit injury, beating, and sweep-net samples have typically been used to determine stink bug infestation levels in various crops (Todd and Herzog 1980). Many of these methods are labor-intensive and can be biased against catching nymphs or adults (Toews and Shurley 2009).

Several species of stink bugs produce pheromones that aid in the aggregation of stink bugs in the field (Todd and Herzog 1980, Krupke et al. 2001). It is believed that these male-produced pheromones attract both sexes (Aldrich et al. 1991, Millar et al. 2002, Leskey and Hogmire 2005) and may have potential in improving monitoring efficiency in the field.

Aldrich et al. (1991) trapped volatiles separately from male and female *Euschistus* spp. by using gas chromatography and found a sex-specific component, methyl (2*E*,4*Z*)-decadienoate, produced by males. This pheromone attracts males, females, and nymphs of several *Euschistus* spp. and is a major component for *E. servus, Euschistus politus* (Uhler), *E. ictericus, E. conspersus,* and *E. tristigmus* (Aldrich et al. 1991). Although direct studies of volatile secretions of *E. quadrator* have not been conducted, this species is caught in traps baited with the *Euschistus* spp. pheromone (Tillman and Cottrell 2012; S.A.B., unpublished data).

Mizell and Tedders (1995) modified a pyramid trap initially developed by Tedders and Wood (1994) to monitor for the pecan weevil, *Curculio caryae* (Horn). These pyramid traps are most effective when painted with industrial safety yellow as opposed to other colors, indicating that the color yellow may be an attractive visual stimulus for stink bugs (Mizell and Tedders 1995, Leskey and Hogmire 2005). Yellow pyramid traps have increased attraction when paired with the *Euschistus* spp. pheromone methyl (2*E*,4*Z*)-decadienoate. Hogmire and Leskey (2006) were able to significantly reduce escape of *Euschistus* spp. by reducing the cone opening inside the trap, and increasing jar size, with or without an insecticidal ear tag in apple (*Malus* spp.) and peach (*Prunus* spp.) orchards.

Tube traps made from clear plastic tubes with wire mesh cones on the ends have been used in several studies with varying results, and they are still commercially available for monitoring stink bugs. Krupke et al. (2001) caught very few stink bugs by using two variations of the tube trap in mullein (*Verbascum thapsus* L.), whereas Aldrich et al. (1991) used these traps to evaluate the *Euschistus* spp. pheromone in weedy areas or blackberry patches, and these traps caught many stink bugs.

Very little research has been conducted on stink bugs in blackberries, despite their potential to cause fruit injury. Feeding injury in blackberries results in collapsed or leaky drupelets that render the fruit unmarketable. Moreover, stink bug odor can alter the taste of the fruit and thereby also negatively affect its marketability. This is especially important in mechanically harvested berries where stink bugs are continuously disturbed with the mechanical harvester (DeFrancesco et al. 2002).

With the changing pest complex and potential future increases in blackberry production in the southeast, it is necessary to identify and survey any previously undocumented pest species for blackberry in Florida that may pose a threat to increasing production. The specific objectives for this study were to identify key stink bug species present in blackberry, to compare commercial traps and lures for monitoring stink bugs in blackberries, and to conduct feeding assays to determine injury caused by *E. quadrator* on blackberry.

Materials and Methods

Study Site. Research was conducted at the University of Florida, Plant Science Research and Education Unit (PSREU) in Citra, FL, and at the Small Fruit and Vegetable integrated pest management (SFVIPM) laboratory at the University of Florida in Gainesville, FL. The experimental site in Citra consists of two 0.10-ha sites, each composed of six blackberry cultivars, one cultivar per row: 'Kiowa', 'Ouachita', 'Arapaho', 'Chickasaw', 'Choctaw', and 'Natchez'. Each row was ≈38.1 m in length, with 30 blackberry plants. Rows were spaced ≈4.5 m apart, with the plot ≈27.5 m in width. Half of the site was managed as a traditional conventional site, and the other was managed as an organic site. Blackberry plants were ≈3 yr old and 1.5 m in height during experimentation.

Blackberry plots were watered using a hard line with in-line emitters as needed, not exceeding 23,385–28,062 liters/ha/d. The organic plot received half the amount of water as the conventional plot. Organic plots were covered with DeWitt landscaping tarp (DeWitt Company, Sikeston, MO). Hydrosource water-gathering crystals (Castle International Resources, Sedona, AZ) in various sizes (medium and standard) were added to the soil at a rate of 4,882.59 kg/ha in the organic plots for water retention. Blackberries were grown on a moving arm shift trellis (Stiles 1999). This trellis is shifted during bloom stages to allow flowers to open toward the sky and after fruiting to ease in berry harvest. After fruiting, floricanes were pruned to allow room for the primocanes.

All fungicides were applied biweekly at manufacturer's recommended rates. Both conventional and organic plots were sprayed with copper sulfate (Lowe's, Gainesville, FL) as a fungicide. Conventional plots also received the fungicide Pristine (BASF, Co., Research Triangle Park, NC), whereas organic plots were sprayed with the fungicide Serenade (AgraQuest, Inc., Davis, CA). With respect to nutrition, in March and June conventional plots were fertilized with 10–10–10 (Lowe's) and the organic plots were fertilized with Nature Safe 10–2–8 (Nature Safe Cold Spring, KY) at a rate of 224.17 kg/ha over a 1-m band. Weed control in the conventional plots was accomplished manually or with the herbicide Round Up (Monsanto, St. Louis, MO) at the recommended rate, whereas weeds were hand-pulled weekly from the organic plot.

Species Composition. Sampling was conducted in 2010 on four randomly chosen blackberry plants of each cultivar in conventional and organic blackberry plots. A harvest tray (≈1 by 0.6 m) (Wal-Mart, Gainesville, FL) was placed on the ground at the base of the blackberry bush. Bushes were shaken vigorously three to four times over the tray, and the cover of the tray was replaced immediately. All stink bug species that fell into the tray were collected. Sampling occurred once every 2 wk for 8 wk from 8 May to 19 June when blooms were >50% fruiting. Stink bugs from each sample were transferred to collecting jars and labeled by date, sample, variety, and plot. All jars were brought back to the SFVIPM laboratory. All stink bugs were counted, pinned and identified to species using a key to the Florida families of Pentatomidae (J.E.E., unpublished; Rolston 1974, McPherson 1989, Rider and Chapin 1992). Weekly observations on percent of berries in various ripening stages were recorded.

Trap Comparison. Two different types of commercially available stink bug traps were compared with and without pheromone lures: 1) Yellow pyramid trap (R. Mizell, Quincy, FL) and 2) Trécé tube trap (Great Lakes IPM, Vestaburg, MI). Pyramid traps were constructed as recommended by Mizell (2008). Both the Trécé Pherocon Centrum lure (Trécé Inc., Adair, OK) and Suterra Scenturion lure (Suterra Corporate, Bend, OR) were used in each baited trap because information on the lure that performs the best in blackberry was unavailable.

Four treatments were evaluated: 1) Pyramid trap baited, 2) Pyramid trap unbaited, 3) Trécé trap baited, and 4) Trécé trap unbaited. The experiment was a randomized complete block design with three replicates. The yellow pyramid trap was placed east of the row ≈0.5 m from the bushes (Fig. 1). The Trécé tube trap was hung from the trellis inside the bush ≈1 m from the ground (Fig. 2). Traps were spaced a minimum of 15 m apart and were blocked by cultivar (row). Traps were placed in the organic side of the planting. Trap contents were emptied into collecting jars, and treatments were rotated weekly for 3 wk. All stink bug species were brought back to the SFVIPM laboratory to be counted, pinned, and identified.

Pheromone Comparison. *Insect Source.* A laboratory colony was established and maintained for several months from wild-caught *E. quadrator* adults from a variety of host plants. Adults were caged in groups of 8–10 males and females. Cages (15 by 15 by 18 cm) were made from plastic food storage containers (Target, Minneapolis, MN) with holes in the lids (≈ 4 by 5 cm) and sides covered with 0.3-mm mesh screens for aeration. Each cage contained a 59.2-ml Solo soufflé cup with lid (Solo Cup Company, Lake Forest, IL) with a cotton roll (1 cm in diameter, cut to 5-cm length; Richmond Dental, Charlotte, NC) inserted into a hole cut into the lid for a water source. Adults were reared on organic green beans, *Solanum lycopersicum* var. *cerasiforme*, organic roma beans, *Phaseolus vulgaris* L., and organic cherry tomatoes, *Phaseolus vulgaris* "Roma II" L. A strip of cheesecloth was taped to the inside of the cage for oviposition. Cages were kept in a rearing incubator at 25 ± 0.5°C, 55 ± 5% RH, and a photoperiod of 16:8 (L:D) h.

Fig. 1. Yellow pyramid trap with screen top. (Online figure in color.)

Laboratory Study. Y-tube methodology was modified from Borges and Aldrich (1994) and Borges et al. (2011). Bioassays were conducted in a glass Y-tube olfactometer (Chemglass Life Sciences, Vineland, NJ). Humidified airflow was maintained at 1,000 ml/min by using a two-channel air delivery system, with two glass flowmeters, an acrylic chassis, two charcoal filters, and two gas bubblers (Analytical Research Systems, Gainesville, FL). The glass Y-tube (150-mm main tube, 80-mm arms, 35-mm internal diameter, 40/35 joints) was held at a 30° angle inside a cardboard box (44 by 30 by 23 cm) on a piece of foam core set at a 5% incline (Fig. 3). Preliminary studies indicated that *E. quadrator* responded better at a 5% incline as opposed to a horizontal surface. The foam core and interior of the box were white. A hole was cut into the side of the box at the end of the Y-tube so that pheromones did not accumulate inside the box. Two mason jars, located outside of the box, were modified to house the lures during the assays (Fig. 3). Holes were drilled into the lids, and valves were secured to the lids that were connected to the corrugated plastic tubing (5.55-mm interior diameter and 6.35-mm outer diameter) (Cole Parmer, Vernon Hills, IL) attached to the filtration system.

Two commercially available aggregation pheromone lures, Trécé Pherocon Centrum lure and Suterra

Fig. 2. Tube trap. (Online figure in color.)

Scenturion lure, for monitoring *Euschistus* spp. were compared in the Y-tube bioassay. Treatments were compared as follows: 1) Trécé Pherocon Centrum lure against a blank control, 2) Suterra Scenturion lure against a blank control, and 3) Trécé Pherocon Centrum lure versus Suterra Scenturion lure. Stink bugs were placed in the Y-tube base and allowed to acclimate for 3 min before attaching the Y-tube arms and airflow. Stink bugs were evaluated to determine their preference, and the time it took them to make a decision was recorded. A choice was considered made after the insect remained in one of the arms for 1 min. Stink bugs were considered unresponsive after staying in the Y-tube for 15 min without making a choice. Adult stink bugs >1 mo old, from the colony, were used in the assay. In total, 20 responding stink bugs per treatment (10 males and 10 females) were evaluated, with the jars being rotated after 10 responding stink bugs to prevent positional bias. Glassware and tubing were rinsed with deionized water, methanol, and ethanol and dried in an oven at 160°C after each test.

Field Study. The aggregation pheromone lures tested in the laboratory were evaluated in the field. Yellow pyramid traps were used because our preliminary trap comparison research indicated that these traps performed better than tube traps. Three treatments were compared: 1) Trécé Pherocon Centrum lure, 2) Suterra Scenturion lure, and 3) unbaited trap. Experimental design was a randomized complete block with four replicates. Traps were placed east of the row ≈0.5 m from the blackberry bushes. Traps were spaced a minimum of 15 m apart within rows, ≈5 m between rows, and were blocked by cultivar (row). Traps were placed in the first, third, fifth, and sixth rows of the organic side of the planting. Trap contents were emptied into collecting jars, and traps were rotated weekly. All stink bug species were brought back to the SFVIPM laboratory to be counted, pinned, and identified.

Feeding Injury. No-choice experiments to evaluate *E. quadrator* feeding injury on blackberries were conducted in the SFVIPM laboratory. Experimental layout consisted of three treatments in a completely randomized design with four replicates. Treatments included 1) green fruit, 2) turning fruit (red stage), and 3) ripe fruit (black berries). Blackberry shoots with berries used in assays were harvested from the blackberry planting located at PSREU. Bushes were

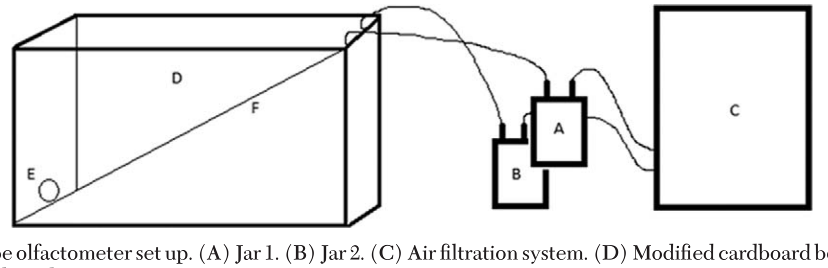

Fig. 3. Y-tube olfactometer set up. (A) Jar 1. (B) Jar 2. (C) Air filtration system. (D) Modified cardboard box. (E) Aeration hole. (F) Foam board.

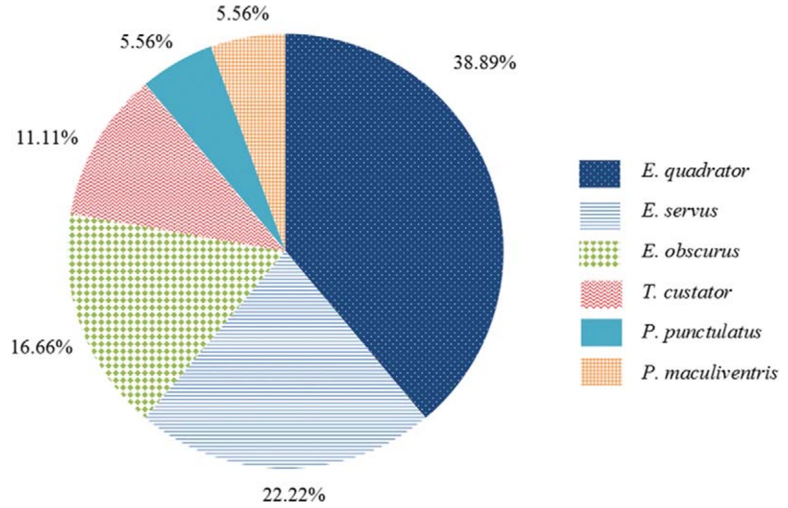

Fig. 4. Species complex in blackberries by using the beating tray method. (Online figure in color.)

managed as discussed under Study Site. Blackberry shoots containing at least two berries were collected from Kiowa blackberries. After shoots with berries were cut from blackberry canes, they were immediately placed into a foam cooler with ice packs to maintain freshness until they were placed in bioassay chambers. Each shoot was surrounded with a foam stopper and placed into a 50-ml vial, containing 35 ml of tap water before being placed into a bioassay chamber. Bioassay chambers consisted of 32-oz plastic containers with lids that had a mesh covered hole.

E. quadrator used for fruit injury assays were obtained from the colony discussed under *Insect Source.* The experiment was conducted with *E. quadrator* adults and with third instars. Adults and nymphs used in the experiment ranged between 20 (nymphs) and 50 (adults) days old. Before conducting assays, all stink bugs were starved for a 24-h period. Three stink bugs were released into each container. In experiments with adult *E. quadrator,* females were released into half of the containers, and males were released into the other half. The adult experiment was conducted in two trials of four replicates to produce a total of eight replicates, four with females and four with males. Stink bugs were allowed to feed for 72 h. Containers were checked daily and the location of each stink bug (berry, leaf, or other [e.g., container, stem]), was recorded. After 72 h, injury descriptions were made by examining each berry for visible signs of injury and recording them.

After the injury assessment, berries and leaves were removed from stems and stored in small plastic containers in the freezer. A double stain solution, a combination of Acid Fuchsin and Lignin Pink, (BioQuip Products, Rancho Dominguez, CA) was used to stain stink bug stylet sheaths on the leaves and berries (Bowling 1979). The solution was diluted using the ratio 1 ml of stain:10 of ml water. Berries and leaves were left in the stain for 1 h and then rinsed with tap water. Leaves and berries were examined under a dissecting microscope. Leaves were examined only for the presence of stylet sheaths to determine whether *E. quadrator* feeds on leaves as well as fruit. In contrast, the number of stylet sheaths on each berry was counted, and these counts were used to calculate the average number of sheaths per berry.

Data Analysis. Species survey information is presented as total counts from organic and conventional due to low numbers of stink bugs found in the field. Data on trap comparison and pheromone comparison were analyzed using analysis of variance (ANOVA), and differences among means were determined using the least significant difference (LSD) mean separation test (0.05) (PROC GLM, SAS Institute 2008).

The Y-tube assay data were analyzed using a chi-square analysis with an expected probability of 0.5. A *t*-test was used to determine any significant differences between sexes of *Euschistus* spp. (0.05) (PROC T-TEST, SAS Institute 2008).

For the feeding injury study, the number of *E. quadrator* found on berries was square root–transformed to fit the assumptions of the analysis and compared among berry stage (green, turning fruit, and ripe fruit) and sex by using a two-way ANOVA (SAS Institute 2008) for the adult data. A one-way ANOVA (SAS Institute 2008) was used to compare the number of *E. quadrator* found on berries for the third-instar data. Means were separated using the LSD test (SAS Institute 2008).

For the adult stink bug data, numbers of stylet sheaths per berry were $\log_{10}$-transformed to fit the assumptions of the analysis and compared using a two-way ANOVA (SAS Institute 2008), with berry stage and sex as factors. For the third instars, numbers of stylet sheaths per berry also were $\log_{10}$-transformed to fit the assumptions of the analysis and were compared using a one-way ANOVA (SAS Institute 2008).

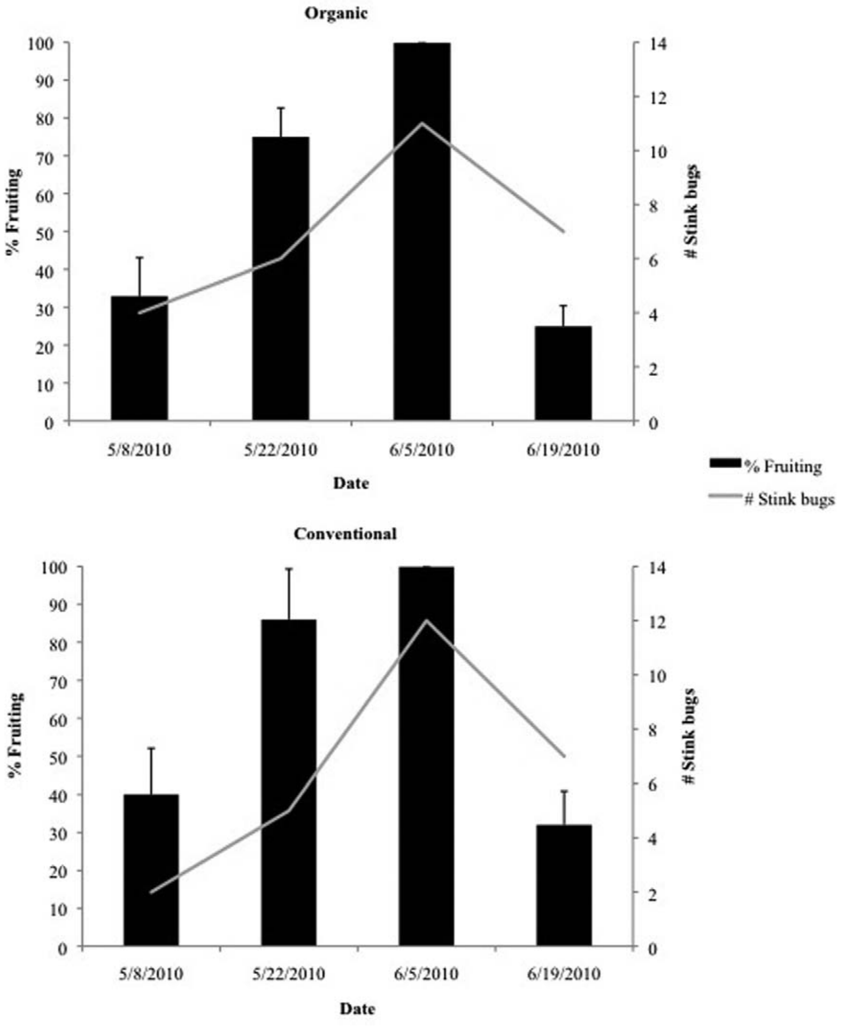

Fig. 5. Total stink bug numbers correlated with mean percent fruit development in organic and conventional blackberry plots.

Means were separated using the LSD test (SAS Institute 2008).

Results

Species Composition. In total, 54 stink bugs were collected in both conventional and organic blackberry plots throughout the sampling period. Figure 4 provides the percentages of species collected, including *E. quadrator*, the most abundant species, followed by *E. servus; E. obscurus; Thyanta custator* (F.); *Proxys punctulatus* (Palisot de Beauvois); and the spined soldier bug, *Podisus maculiventris* (Say). Both males and females were found in the field, with the exception of *P. punctulatus* and *P. maculiventris*, for which no females were recorded. Sex ratios between species were not statistically different, averaging 1:1 (male:female) among most species. Also, there was no difference in species composition between conventional and organic plots. As the percentage of ripe fruit increased in both the organic and conventional plots, the number of stink bugs also increased (Fig. 5).

Other insects collected in the beating tray included ants (Hymenoptera: Formicidae), spiders (Arachnida: Araneae: Salticidae, Oxyopidae), grasshoppers (Orthoptera: Acrididae), plant bugs (Hemiptera: Miridae), katydids (Orthoptera: Tettigoniidae), leaffooted bugs (Hemiptera: Coreidae), lady beetles (Coleoptera: Coccinellidae), and flower beetles [Coleoptera: Scarabaeidae; *Euphoria sepulcralis* (F.)]. There was no difference in insect species composition between conventional and organic plots.

Trap Comparison. In field tests, there was a highly significant difference between trap types ($F = 17.79$; df = 3, 11; $P = < 0.0001$), with the yellow pyramid trap catching more stink bugs than the tube trap (Fig. 6). However, there were no significant differences be-

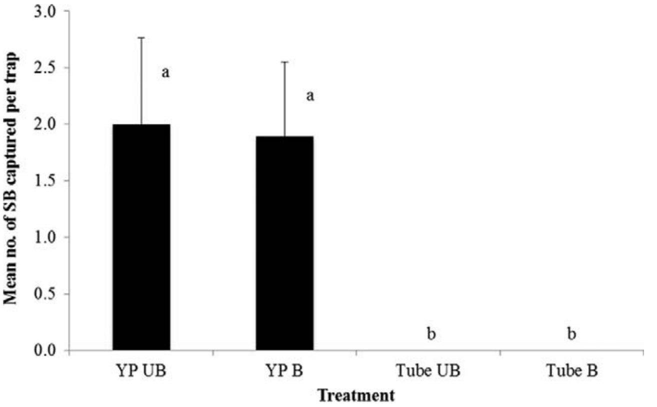

Fig. 6. Mean number of stink bugs captured in yellow pyramid traps (YP) or tube traps with (B) or without (UB) pheromone lures. Means followed by the same letter were not significantly different at $P < 0.05$ level.

tween baited and unbaited pyramid or tube traps. Both males and females were caught in the traps, but there were no statistical differences between sexes ($t = 0.49$, $P = 0.6526$).

The species recorded for this experiment, in order of abundance, consisted of *E. servus, E. quadrator, P. maculiventris, E. obscurus, Euschistus ictericus* (L.), and *T. custator* (Table 1).

Pheromone Comparison. In the Y-tube assays, there were no statistical differences between any of the treatments. In the Trécé lure versus a blank control, 12 stink bugs chose the lure and eight chose the blank control ($\chi^2 = 0.8$, df = 1, $P = 0.3711$). In the Suterra lure versus a blank control, 10 stink bugs each chose the lure or the blank control ($\chi^2 = 0.0$, df = 1, $P = 1.00$). In the Trécé lure versus the Suterra lure, 13 stink bugs chose the Trécé lure and seven chose the Suterra lure ($\chi^2 = 1.8$, df = 1, $P = 0.1797$). There were 12 nonresponders in total, distributed fairly evenly across all treatments.

Table 1. Mean species composition for trap comparison study (n = 35)

Species	Avg. density[a]	% total[b]
E. servus	5.00 ± 3.05	42.86
Males	1.67 ± 0.67	
Females	3.33 ± 2.40	
E. quadrator	2.00 ± 0.00	17.14
Males	1.67 ± 0.33	
Females	0.33 ± 0.33	
T. custator	0.33 ± 0.33	2.86
Males	0.33 ± 0.33	
Females	0.00	
E. ictericus	1.33 ± 0.88	11.43
Males	0.33 ± 0.33	
Females	1.00 ± 0.58	
P. maculiventris	1.67 ± 0.67	14.29
Males	0.00	
Females	1.67 ± 0.67	
E. obscurus	1.33 ± 0.33	11.43
Males	0.33 ± 0.33	
Females	1.00 ± 0.58	

[a] Average density (mean ± SE) of each category per replicate.
[b] Percentage of total stink bugs collected.

Similar to our Y-tube assays, no significant differences were found in the field tests that compared pheromone-baited pyramid traps with unbaited pyramid traps ($F = 0.85$; df = 2, 11; $P = 0.4354$) (Fig. 6). Numerically, traps baited with the Trécé lure caught more stink bugs ($n = 10$). The species composition for this experiment in order of abundance consisted of *E. servus, T. custator, E. quadrator,* and *E. ictericus.* Both males and females were caught in the traps, but there were no statistical differences between sexes ($t = 0.18$, $P = 0.8648$).

Feeding Injury. Injury to berries caused by *E. quadrator* adults and third instars was similar. The most common injury to green berries was discoloration (Fig. 7A). There were a few green berries with misshapen drupelets (Fig. 7B). In contrast, misshapen drupelets were commonly seen on turning and ripe berries (Fig. 7B), with only a few berries showing discoloration (Fig. 7C).

In the comparison of numbers of *E. quadrator* on berries, there was no interaction between berry stage and sex on any sampling day in the adult data (all $F \leq 1.54$; df = 2, 23; $P \geq 0.24$). There were no differences between males and females on any sampling day in either adult experiment (all $F \leq 1.39$; df = 1, 23; $P \geq 0.25$). There were no differences among treatments (Fig. 8A) on any sampling day (all $F \leq 1.24$; df = 2, 23; $P \geq 0.31$).

In the experiment with third instars, 72 h after release, there were significantly higher numbers of nymphs on the ripe berries compared with the green berries ($F = 4.5$; df = 2, 11; $P = 0.04$) (Fig. 8B). There were no differences on any other day (both $F \leq 1.46$; df = 2, 11; $P \geq 0.28$).

In comparing the number of stylet sheaths per berry, there was no interaction between berry stage and sex in the adult ($F = 0.27$; df = 2, 23; $P = 0.77$). There was also no difference between males and females ($F = 2.81$; df = 2, 23; $P = 0.11$). For the adults, there were significantly more stylet sheaths found in green berries compared with both of the other berry stages ($F = 16.04$; df = 2, 23; $P = 0.0001$) (Fig. 9).

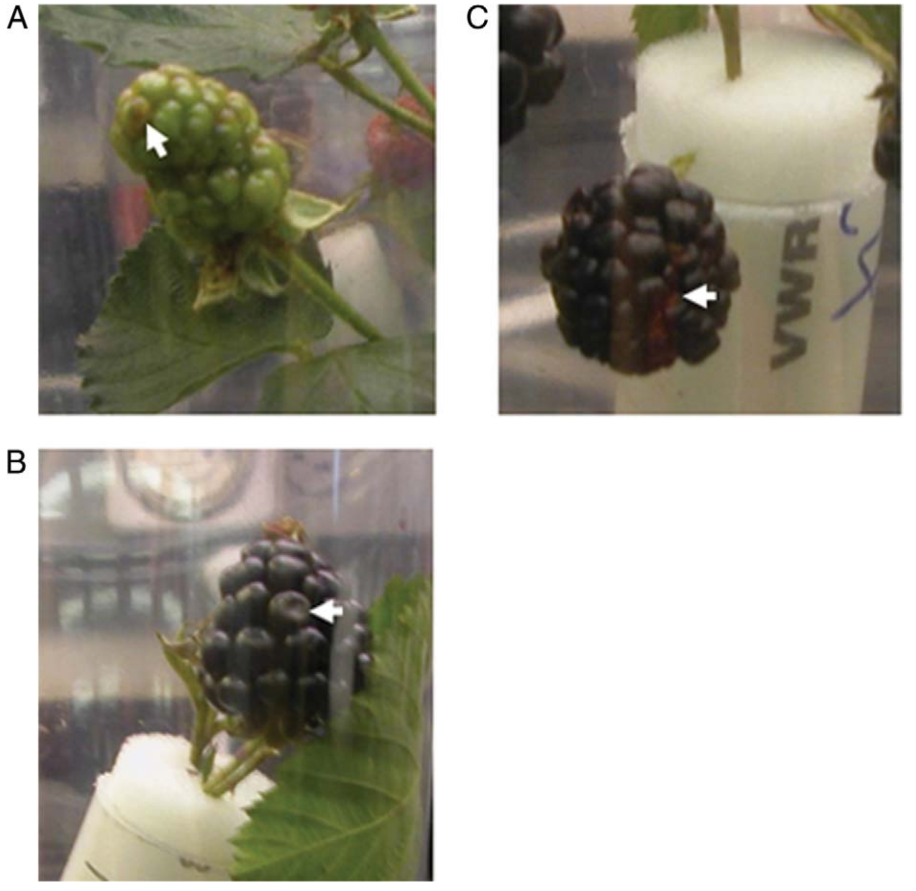

Fig. 7. Stink bug injury to berries. (A) Discoloration of green berry. (B) Ripe berry with misshapen drupelet. (C) Discoloration of ripe berry. (Online figure in color.)

In the experiment with third instars, there were significantly more stylet sheaths found in green berries compared with turning berries ($F = 5.34$; df = 2, 11; $P = 0.03$).

Stylet sheaths were found in leaves from all berry stages in all three experiments. Sheaths were found in leaf veins and were concentrated on the main leaf vein.

Discussion

Several new stink bug records for blackberries were found. This report is the first known record of *E. quadrator, E. obscurus, T. custator, P. punctulatus,* and *P. maculiventris* in blackberry. Little, if any, information has been published in journals on the stink bug complex in blackberries. The limited information available is on websites states that green stink bugs, southern green stink bugs, and brown stink bugs are most prominent.

The first visual sighting of stink bugs was of *E. servus* on 28 April 2010 in the organic plot that began fruiting after the conventional plot. Blackberries ripened faster in the organic plot, perhaps making the organic plot more attractive to stink bugs. Although overall stink bug numbers were low, there was a general correlation between the number of stink bugs found on each sampling date and the amount of ripe fruit in each plot. As the percentage of ripe fruit increased in both the organic and conventional plots, the number of stink bugs also increased. During sampling, most stink bugs were found on the third sampling date (5 June 2010), when most berries were ripe. The Chickasaw cultivar produced the most stink bugs across both the conventional and organic plots. This variety ripens ≈1 wk later than Kiowa and Natchez cultivars.

The majority of stink bug information in Florida, especially for *Euschistus* spp., is from the early 20th century. A review of the literature shows that, historically, the pest status of stink bugs in Florida is debatable. The most commonly mentioned stink bugs in the state are *N. viridula, E. servus,* and *E. ictericus. N. viridula* is commonly found in rice (*Oryza sativa* L.), soybean, faba bean (*Vicia faba* L.), and various weeds in southern Florida (Buschman and Whitcomb 1980, Temerak and Whitcomb 1984, Jones and Cherry 1986, Nuessly et al. 2004, Cherry and Wilson 2011). Given this information, we expected to find mostly green stink bugs and *E. servus* in our blackberry planting. *E. servus* is usually seen in pecan, *Carya* spp., soybean,

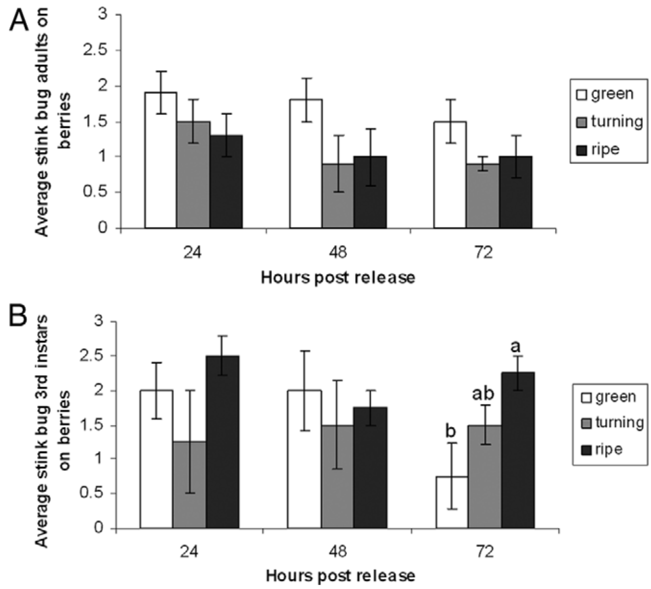

Fig. 8. Number of stink bug adults (A) and third instars (B) on berries at each sampling time. Means with the same letter are not significantly different from each other at $P \leq 0.05$. Error bars represent SEM.

elderberry, *Sambucus* spp., and goldenrod *Solidago* spp., in Florida (Hill 1938, Frost 1979, Fontes et al. 1994). *E. ictericus* is found in rice and faba bean (Temerak and Whitcomb 1984, Jones and Cherry 1986, Nuessly et al. 2004, Cherry and Wilson 2011). There is no doubt that these pests also occur in other Florida crops, but literature with this information was not found.

When using any monitoring device, care should be taken in identifying the stink bug species present. *E. servus* is relatively easy to identify versus other *Euschistus* spp., but *E. ictericus, E. tristigmus,* and *P. maculiventris* look very similar, and the smaller brown stink bugs of the "lesser brown stink bug complex" are easily confused (Hopkins et al. 2005). Predatory stink bugs, such as the spined soldier bug, may be mistaken as pests. *P. maculiventris* is a beneficial predatory stink bug that mostly feeds on lepidopteran and coleopteran larvae, but it has been shown to feed on phytophagous stink bugs (McPherson et al. 1980, McPherson and McPherson 2000).

The yellow pyramid traps captured both predatory and pest species by using the *Euschistus* spp. pheromone (Mizell 2008). In the trap comparison experiment, the yellow pyramid trap was clearly superior. In fact, no stink bugs were caught in the tube traps. This is similar to the observations of Krupke et al. (2001), where very low numbers of stink bugs were captured when comparing two different sizes of tube traps. However, it should be noted that Aldrich et al. (1991) caught a number of *Euschistus* spp. by using the tube trap in a deciduous forest. This indicates that tube traps may not be effective in blackberry plantings but may have potential uses in other crops. The pyramid traps were effective in catching stink bugs, either with or without the addition of the *Euschistus* spp. pheromone, and there were no differences between baited and unbaited traps. Most studies using the pyramid trap find that the addition of the *Euschistus* spp. pheromone increases the efficacy of the trap (Leskey and Hogmire 2005, Cottrell and Horton 2011). It is possible that volatiles

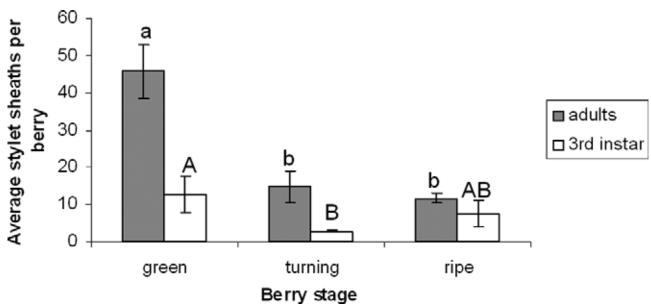

Fig. 9. Average sheaths per berry recorded from each berry stage from the first and second adult and third-instar larvae experiments. Means with the same letter are not significantly different from each other at $P \leq 0.05$. Error bars represent SEM.

emitted from developing blackberries may have lessened the effect of the pheromone, making it less attractive to *Euschistus* spp.

Similarly, in the pheromone comparison experiment, there were no statistical differences between the Trécé Pherocon Centrum lure, the Suterra Scenturion lure, and an unbaited trap. Overall, stink bug numbers were low, which may partially account for some of the observed nonsignificant differences and the obvious similarities in effectiveness. However, these results were supported by the Y-tube olfactometer assays where there were no statistical differences between the lures and a control, although higher numerical values were recorded with the Trécé lure when using *E. quadrator.* The composition of these commercial lures was unknown because this was proprietary information that the companies did not want to disclose. Because there were no statistical differences between lures, both in the field and in the Y-tube bioassay, there is strong evidence to indicate that these lures may not be effective for monitoring stink bugs in blackberry plantings. Also, that there were no differences between baited and unbaited traps may indicate that the primary attraction to pyramid traps in blackberry crops may be a response to the yellow color and probably trap type (architecture).

Species found in field experiments included *E. quadrator, E. servus, E. obscurus, T. custator, E. ictericus,* and *P. maculiventris.* Only adult stink bugs were caught in monitoring traps. The reason why only adults were caught is most likely due to adult stink bugs flying to the traps, and nymphs were not likely to crawl to the traps from blackberry bushes. Because no statistical differences were found between sexes, it can be determined that this trap attracts both males and females. Leskey and Hogmire (2005) captured both sexes of *E. servus* in a similar pyramid trap; however they found a higher percentage of females than males in both baited and unbaited traps. Stink bugs seem to colonize blackberry when berries are mid-ripe to fully ripe. In the trap comparison field experiment the majority of stink bugs were found in traps located in the Kiowa variety. Kiowa ripens earlier than most varieties, in early June, and fruiting extends for 6 wk (Moore and Clark 1996). The peak in stink bug numbers occurred during the week of 5 May 2010, when berries were mid-ripe. In the pheromone comparison experiment, most stink bugs were found during the week of 26 May 2010 in Kiowa and Natchez when berries were beginning to ripen. Natchez also ripens early in June, and fruiting extends for 4 wk (Clark and Moore 2008).

P. punctulatus was not caught in traps during field studies but was found in the species survey experiment. This stink bug has been found previously in the pyramid trap (Mizell 2008). It has been suspected of being predaceous but is also known to feed on plants and weeds, such as cotton and zigzag spiderwort, *Tradescantia subaspera* Ker (Vangeison and McPherson 1975, Gomez and Mizell 2009). These stink bugs are not thought to cause significant economic damage (Schaefer and Panizzi 2000).

E. quadrator feeding injury to green blackberries produced discoloration but little deformation. This was probably due, in part, to berries not continuing to develop after the shoots are clipped from the bushes. Deformation injuries to turning and ripe berries were more obvious.

Berry stage had little effect on the location of *E. quadrator* within the containers. This may be partly due to both adults and third instars feeding on the leaves as well as the berries. Most of the adults and all of the nymphs were found on some part of the blackberry shoot.

The mean number of stylet sheaths found in the different berry stages indicates that both adult and third-instar *E. quadrator* fed more on green berries compared with turning fruit and adults fed more on green berries compared with ripe fruit. Therefore, monitoring for *E. quadrator* should begin as soon as the blackberry plants begin to set fruit. Adults and nymphs also fed on both turning and ripe berries. In the first trial of the adult experiment and the third-instar experiment, the turning fruit was in a later stage of ripening compared with the turning fruit in the second trial of the adult experiment. This may explain the lack of differences between turning and ripe fruit.

Overall, blackberries in Florida seem to have a different stink bug complex than that of other production areas. Although we found several *Euschistus* spp. that are commonly mentioned in other areas, we did not find either the green stink bug or the southern green stink bug. More extensive sampling may be needed to capture additional species. It is also interesting to note that so many different *Euschistus* spp. were found on the blackberries at the same time and that *E. quadrator* was the dominant species. Our findings also indicate that the yellow pyramid trap was more effective than the tube trap for monitoring stink bugs in blackberry. Furthermore, our findings indicate that the addition of pheromone lures to pyramid traps may not increase their attractiveness to stink bugs in blackberry. Both adults and nymphs of *E. quadrator* feed on green fruit, and the species seem to favor this stage of ripening. Therefore, we recommend the implementation of pest management strategies, including regular monitoring, when fruit is young and stink bug nymphs begin to appear in the planting.

Acknowledgments

We thank employees of the Plant Science Research and Education Unit in Citra, FL, for assisting with plot maintenance. Special thanks to the SFVIPM laboratory at the University of Florida for field assistance throughout the duration of the project. Special thanks to Nicole Benda for assistance with the Y-tube bioassay. We thank Eileen Buss for revisions on an earlier draft of this article. This project was funded by a sustainable SHARE grant from the University of Florida.

References Cited

Aldrich, J. R., M. P. Hoffmann, J. P. Kochansky, W. R. Lusby, J. E. Eger, and J. A. Payne. 1991. Identification and attractiveness of a major pheromone component for Nearctic *Euschistus* spp. stink bugs (Heteroptera: Pentatomidae). Environ. Entomol. 20: 477–483.

Anonymous. 2008. Berries Northwest. (http://www.berriesnw.com/BerryDisordersList.asp).

Borges, M., and J. R. Aldrich. 1994. Attractant pheromone for Nearctic stink bug, *Euschistus obscurus* (Heteroptera: Pentatomidae): insight into a Neotropical relative. J. Chem. Ecol. 20: 1095–1102.

Borges, M., M.C.B. Moraes, M. F. Peixoto, C.S.S. Pires, E. R. Sujii, and R. A. Laumann. 2011. Monitoring the Neotropical brown stink bug *Euschistus heros* (F.) (Hemiptera: Pentatomidae) with pheromone-baited traps in soybean fields. J. Appl. Entomol. 135: 68–80.

Bowling, C. C. 1979. The stylet sheath as an indicator of feeding activity of the rice stink bug. J. Econ. Entomol. 72: 259–260.

Buschman, L. L., and W. H. Whitcomb. 1980. Parasites of *Nezara viridula* (Hemiptera: Pentatomidae) and other Hemiptera in Florida. Fla. Entomol. 63: 154–162.

Cherry, R. H., and A. Wilson. 2011. Flight activity of stink bug (Hemiptera: Pentatomidae) pests of Florida rice. Fla. Entomol. 94: 359–360.

Clark, J. R., and J. N. Moore. 2008. 'Natchez' thornless blackberry. HortScience. 43: 1897–1899.

Cottrell, T. E., and D. L. Horton. 2011. Trap capture of brown and dusky stink bugs (Hemiptera: Pentatomidae) as affected by pheromone dosage in dispensers and dispenser source. J. Entomol. Sci. 46: 135–147.

DeFrancesco, J., W. Parrott, and J. Jenkins. 2002. Crop profile for blackberry in Oregon. (http://www.ipmcenters.org/cropprofiles/docs/ORblackberries.pdf).

Ellis, M. A., R. H. Converse, R. N. Williams, and B. Williamson. 1991. Compendium of raspberry and blackberry diseases and insects. APS Press, St. Paul, MN.

Fontes, E.M.G., D. H. Habeck, and F. Slansky, Jr. 1994. Phytophagous insects associated with goldenrods (*Solidago* spp.) in Gainesville, Florida. Fla. Entomol. 77: 209–221.

Frost, S. W. 1979. A preliminary study of North American insects associated with elderberry flowers. Fla. Entomol. 62: 341–355.

Gomez, C., and R. F. Mizell, III. 2009. Black stink bug *Proxys punctulatus* (Palisot) (Insecta: Hemiptera: Pentatomidae). EENY-432. University of Florida IFAS Extension. (http://edis.ifas.ufl.edu/in795).

Hill, S. O. 1938. Important pecan insects of northern Florida. Fla. Entomol. 21: 9–13.

Hogmire, H. W., and T. C. Leskey. 2006. An improved trap for monitoring stink bugs (Heteroptera: Pentatomidae) in apple and peach orchards. J. Entomol. Sci. 41: 9–21.

Hopkins, B. W., J. S. Bernal, and A. E. Knutson. 2005. *Euschistus quadrator* (Hemiptera: Pentatomidae): a new pest in lower Texas Gulf coast cotton. Proc. Beltwide Cotton Conf. 1480–1485.

Jennings, D. L., H. A. Daubeny, and J. N. Moore. 1991. Blackberries and raspberries (*Rubus*). Acta Hortic. 290: 331–392.

Johnson, D., and B. A. Lewis. 2003. Crop profile for blackberries in Arkansas. (http://www.ipmcenters.org/CropProfiles/docs/ARblackberry.pdf).

Jones, D. B., and R. H. Cherry. 1986. Species composition and seasonal abundance of stink bugs (Heteroptera: Pentatomidae) in southern Florida rice. J. Econ. Entomol. 79: 1226–1229.

Krupke, C. H., J. F. Brunner, M. D. Doerr, and A. D. Kahn. 2001. Field attraction of the stink bug *Euschistus conspersus* (Hemiptera: Pentatomidae) to synthetic pheromone-baited host plants. J. Econ. Entomol. 94: 1500–1505.

Leskey, T. C., and H. W. Hogmire. 2005. Monitoring stink bugs (Hemiptera: Pentatomidae) in mid-Atlantic apple and peach orchards. J. Econ. Entomol. 98: 143–153.

Maxey, L. M. 2011. Pest management of Japanese Beetle (Coleoptera: Scarabaeidae) and a study of stink bug (Hemiptera: Pentatomidae) injury on primocane-bearing caneberries in southwest Virginia. M.S. thesis, Virginia Polytechnic Institute and State University, Blacksburg.

McPherson, J. E. 1989. An overview of the Heteroptera of Illinois. Great Lakes Entomol. 22: 177–198.

McPherson, J. E., and R. M. McPherson. 2000. Stink bugs of economic importance in America north of Mexico. CRC, Boca Raton, FL.

McPherson, J. E., M. K. Mukerji, and E. J. LeRoux. 1980. A list of the prey species of *Podisus maculiventris* (Hemiptera: Pentatomidae). Great Lakes Entomol. 13: 17–24.

Millar, J. G., H. L. McBrien, H. Y. Ho, R. E. Rice, E. Cullen, F. G. Zalom, and A. Üokl. 2002. Pentatomid bug pheromones in IPM: possible applications and limitations. Int. Org. Biol. Control Bull. 25: 241–250.

Mizell, III, R. F. 2007. Insect management in blackberries. ENY-410. University of Florida IFAS Extension. (http://edis.ifas.ufl.edu/ig069).

Mizell, III, R. F. 2008. Monitoring stink bugs with the Florida stink bug trap. University of Florida IFAS Extension. (http://nfrec.ifas.ufl.edu/MizellRF/stink_bugs/stink_bugs.htm).

Mizell, III, R. F., and W. L. Tedders. 1995. A new monitoring method for detection of the stinkbug complex in pecan orchards. Proc. Southeast. Pecan Growers Assoc. 88: 36–40.

Moore, J. N. 1997. Blackberries and raspberries in the southern United States: yesterday, today and tomorrow. Fruit Varieties J. 51: 148–157.

Moore, J. N., and J. R. Clark. 1996. 'Kiowa' blackberry. HortScience. 31: 286–288.

Nuessly, G. S., M. G. Hentz, R. Beiriger, and B. T. Scully. 2004. Insects associated with faba bean, *Vicia faba* (Fabales: Fabaceae), in southern Florida. Fla. Entomol. 87: 204–211.

Rider, D. A., and J. B. Chapin. 1992. Revision of the genus *Thyanta* Stål, 1862 (Heteroptera: Pentatomidae) II. North America, Central America, and the West Indies. J.N.Y. Entomol. Soc. 100: 42–98.

Rolston, L. H. 1974. Revision of the genus *Euschistus* in Middle America (Hemiptera, Pentatomidae, Pentatomini). Entomol. Am. 48: 1–102.

SAS Institute. 2008. SAS/STAT user's guide, version 9.2, SAS Institute, Cary, NC.

Schaefer, C. W., and A. R. Panizzi. 2000. Heteroptera of economic importance. CRC, Boca Raton, FL.

Stiles, H. D. 1999. Limited arm-rotation shift-trellis (LARS) and primocane management apparatus (PMA) for raspberries and blackberries (*Rubus* cvs. or crops). Va. Agric. Exp. Sta. Bull. 99-1. (http://scholar.lib.vt.edu/ejournals/vaes/vaes99-1.pdf).

Strik, B. C., J. R. Clark, C. E. Finn, and M. Pilar Bañados. 2007. Worldwide production of blackberries. HortTechnology 17: 205–213.

Tedders, W. L., and B. W. Wood. 1994. A new technique for monitoring pecan weevil emergence (Coleoptera: Curculionidae). J. Entomol. Sci. 29: 18–30.

Temerak, S. A., and W. H. Whitcomb. 1984. Parasitoids of predaceous and phytophagous pentatomid bugs in soybean fields at two sites of Alachua County, Florida. Z. Ang. Entomol. 97: 279–282.

Tillman, P. G., and T. E. Cottrell. 2012. Case study: trap crop with pheromone traps for suppressing *Euschistus servus* (Heteroptera: Pentatomidae) in cotton. Psyche. (in press).

Todd, J. W., and D. C. Herzog. 1980. Sampling phytophagous Pentatomidae on soybean, pp. 438–478. *In* M. Kogan and D. C. Herzog (eds.), Sampling methods in soybean entomology. Springer, New York.

Toews, M. D., and W. D. Shurley. 2009. Crop juxtaposition affects cotton fiber quality in Georgia farmscapes. J. Econ. Entomol. 102: 1515–1522.

[USDA–NASS] **U.S. Department of Agriculture–National Agricultural Statistics Survey. 2009.** 2007 Census of agriculture. Volume 1, part 51. United States summary and state data. USDA, Washington, DC.

Vangeison, K. W., and J. E. McPherson. 1975. History and laboratory rearing of *Proxys punctulatus* (Hemiptera: Pentatomidae) with descriptions of immature stages. Ann. Entomol. Soc. Am. 68: 25–30.

Received 1 October 2012; accepted 21 January 2013.