

Monitoring Red Imported Fire Ant (Hymenoptera: Formicidae) Foraging Behavior and Impact on Foliage-Dwelling Arthropods on Soybean Produced Under Three Ant Suppression Regimes¹

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Abstract The red imported fire ant, *Solenopsis invicta* Buren, has been reported to contribute to the biological control of key arthropod pests in soybean, *Glycine max* (L.) Merrill, and other crops. *Solenopsis invicta* also has been implicated as a major disrupter of biological control and a direct crop pest associated with reduced yields. In 2000 and 2001, fire ant foraging, survival of sentinel lepidopteran eggs and pupae, seasonal abundance of common foliage-dwelling soybean arthropods, and crop yield were assessed under three different fire ant suppression treatments at two test sites in Tift Co., GA. The treatments included an untreated control, hydramethylnon ant bait, and hydramethylnon bait in combination with the broad spectrum insecticide chlorpyrifos. Fire ant foraging was reduced in the chemically-treated plots in comparison to the control, based on fire ant numbers on hot dog baits after a 1-h field exposure in the early morning. Survival of soybean looper, *Pseudoplusia includens* Walker, eggs in 2000 and corn earworm, *Helicoverpa zea* Boddie, eggs in 2001 was lower in the untreated plots after a 24-h field exposure, where *S. invicta* foraging rates were high. Soybean looper pupal survival was 34 and 88% lower on foliage and the ground, respectively, in the control compared to those with reduced fire ant foraging. The seasonal abundance of foliage-dwelling soybean arthropods was primarily unaffected by the fire ant suppressing treatments. However, in 2001, green cloverworms, *Hypena scabra* F., and spiders were significantly lower (38.6 and 40.5%, respectively) in the chlorpyrifos plus hydramethylnon bait treatment in comparison to the other treatments. Season-long suppression of fire ants had no effect on soybean yield. The results of this study suggest that *S. invicta*, at naturally-occurring levels, is an active predator on some common soybean lepidopteran pests.

Key Words red imported fire ant, *Solenopsis invicta*, *Glycine max*, hydramethylnon, chlorpyrifos

The red imported fire ant, *Solenopsis invicta* Buren, is a serious pest of humans, wildlife, and structures throughout its North American range (Vinson 1997, Taber 2000). Within the cropping systems of the southeastern United States, *S. invicta* is very abundant and has been reported to be a beneficial predator of crop pests, a disrupter of biological control by other natural enemies, as well as a direct crop pest in a variety of crops. Densities of and damage caused by the boll weevil, *Anthonomus*

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grandis grandis Boheman, are reduced by direct predation from *S. invicta* (Sterling 1978, Sterling et al. 1984). *Solenopsis invicta* also has been observed to be an important predator of pest lepidopteran eggs in Texas cotton (Diaz et al. 2004). All stages of the sugarcane borer, *Diatraea saccharalis* F., are attacked by *S. invicta* (Negm and Hensley 1969). When ant populations were reduced with mirex insecticide, borer infestation and damage increased by 53 and 69% (Reagan et al. 1972). Lepidopteran larval damage to peanuts has been reported to be lower in *S. invicta* infested fields (Vogt et al. 2001).

Predation by *S. invicta* also can have a negative effect on tending and predator interference leading to an increase in cotton aphid, *Aphis gossypii* Glover in cotton (Diaz et al. 2004, Kaplan and Eubanks 2002). Fire ants have been shown to reduce the predatory effectiveness of coccinellids, syrphid larvae, and lacewing larvae on cotton aphid in the laboratory (Vinson and Scarborough 1989). Predation and other interactions with *S. invicta* can negatively impact parasitoids (Lopez 1982, Vinson and Scarborough 1991). *Solenopsis invicta* density was negatively associated with most natural enemy taxa monitored in Alabama cotton (Eubanks 2001, Eubanks et al. 2002). However, Sterling et al. (1979) reported that even at high densities fire ants failed to reduce 47 taxa of predaceous arthropods in cotton. In Oklahoma peanut fields, *S. invicta* preys mostly on pest species and has no negative impact on predators although they do consume some predators such as spiders (Vogt et al. 2001, 2002).

Soybeans in the southeastern United States are heavily infested with *S. invicta*, with 22.2-207.5 active mounds per ha (Banks et al. 1990). Conventional tillage reduces the number of active mounds (Morrill and Greene 1975), thus fire ant abundance is likely to increase in cropping systems that adopt conservation tillage. *Solenopsis invicta* in soybean fields mainly forage on the ground and rarely forage higher than 20 cm on soybean plants (Kidd and Apperson 1984). Despite this, *S. invicta* has been reported to be a major predator of foliage-inhabiting pests. In both open field and field cage studies it has been documented that *S. invicta* has a substantial negative effect on stink bug populations (Krispyn and Todd 1982, Eubanks 2001). Stam et al. (1987) observed *S. invicta* attacking southern green stink bug, *Nezara viridula* L., eggs during soybean vegetative stages but not during reproductive stages. Eggs and small-to-medium sized larvae of the velvetbean caterpillar, *Anticarsa gemmatalis* Hübner, are often consumed by *S. invicta* (Buschman et al. 1977, Elvin et al. 1983). The highest densities of velvetbean caterpillar and green cloverworm, *Hyponomeuta scabra* F., were reported in soybean plots where arthropod predators, including *S. invicta*, were eliminated (Brown and Goyer 1982). However, no mention of *S. invicta* was made in a study of velvetbean caterpillar eggs and larvae in Florida (Godfrey et al. 1989). Additionally, no positive or negative association was found between the abundance of *S. invicta* and populations of lepidopteran larvae in Alabama soybeans (Eubanks 2001). The majority of natural enemy taxa in soybean were reported to be negatively associated with *S. invicta* abundance (Eubanks 2001).

Solenopsis invicta has been reported to be a direct and indirect pest of soybeans. Fire ant mounds reduce harvested yield by interfering with harvesting machinery (Adams et al. 1976). Direct loss of yield also has been attributed to *S. invicta* (Adams et al. 1983, Lofgren and Adams 1981, Apperson and Powell 1983). Most researchers believe that seed and seedling destruction by ants leading to stand reduction is the mechanism of yield loss. Adams et al. (1983) concluded that soybeans are often

planted at the time of greatest food stress on *S. invicta* colonies. Radiotracer studies indicate that *S. invicta* feed on growing plants as well as seeds (Adams et al. 1983). Soybeans that emerged in direct association with *S. invicta* had decreased vigor, less root dry matter, and fewer root nodules in comparison with seedlings not associated with *S. invicta* (Shatters and VanderMeer 2000).

The objective of this investigation was to examine the impact of fire ant suppression on soybean foliage-dwelling pests and natural enemies and simultaneously investigate any impact on yield. The experimental methodology compared these variables between soybean with high naturally-occurring densities of *S. invicta* with soybean that had reduced fire ant foraging due to ant baits or broad spectrum pesticides.

Materials and Methods

Soybeans were planted on two research farms (Bradford and Shannon farms) in Tift Co., GA, in 2000 and 2001. These research sites were selected due to their history of conservation tillage and natural occurrence of *S. invicta*. At the Bradford Farm soybeans var. 'Deltapine 6200 RR' were planted with a no-till drill planter with 17.78 cm row spacing on 7 June 2000. At the Shannon Farm soybeans var. 'Northern King S51T1 RR' were planted into rye stubble on 14 June 2000 in rows with 0.91 m spacing. Glyphosate herbicide (Roundup Ultra, Monsanto Corp., St. Louis, MO) was applied approx. 2 wk after planting for early season weed control at both farms. Identical soybean varieties were planted on the same farms in 2001 (Shannon on 4 June and Bradford on 11 June) using the same production practices, except the Shannon Farm was planted into a fallow field after glyphosphate herbicide burndown instead of into rye stubble as the year before. Test sites were partitioned into units that averaged 0.32 ha, and plots were randomly assigned to one of three treatments designed to impact fire ant abundance: (1) untreated, (2) hydramethylnon containing ant bait (Amdro, BASF Corp., Research Triangle Park, NC) broadcast at 1.7 kg/ha, (3) hydramethylnon at 1.7 kg/acre plus an application of chlorpyrifos broadcast at 2.3 L/ha (Lorsban, Dow Agrosiences, Indianapolis, IN). In plots receiving a treatment of hydramethylnon bait, a 4.5 m untreated border was left on all four edges to minimize overlap of treatment effects on foraging fire ant populations. Treatments were arranged in a randomized complete block design with six replications in 2000 and eight replications in 2001.

Fire ant foraging was monitored in each plot every 7 d throughout the season. A 0.6 cm section of hot dog was used as a bait to monitor the presence of actively foraging ants. Two baits were placed in each plot between 0,730 and 0,800 EST and checked between 0,830 and 0,900 EST. Ant counts per bait after a 1 h field exposure were estimated and assigned a value according to the following rating scale: 0: 0 ants, 1: 1-10 ants, 2: 11-50 ants, 3: 51-100 ants, 4: 101-150 ants, 5: >150 ants. Data were analyzed using a repeated measures analysis of variance (SAS Institute 1990) and means were separated using the Ryan-Einot-Gabriel-Welsch test ($P = 0.05$).

Treatments were applied within 5 d after planting in both 2000 and 2001. Treatments were reapplied when the weekly fire ant monitoring revealed fire ant foraging in the treated plots. Reapplication of treatments took place in early July of 2000 and late July in 2001. In 2000 the Shannon Farm site was treated with a broadcast foliar application of diflubenzuron (Dimilin, Uniroyal Chemical, now Crompton Crop Protec-

tion, Middlebury, CT) in mid-September for control of velvetbean caterpillars. Late-season applications of I-cyhalothrin (Karate, Syngenta Crop Protection, Greensboro, NC) at the Shannon Farm site and tralomethrin (Scout Xtra, Aventis Crop Science, now Bayer Crop Science, Research Triangle Park, NC) at the Bradford Farm site were made as curative pest controls in 2001.

In 2000, soybean looper, *Pseudoplusia includens* Walker, moths from an existing laboratory colony were given butcher paper as oviposition substrate. The paper was removed 24 h after being exposed to egg-laying moths so that all eggs were <24 h old. Sections of paper, approx. 2.5 cm², each containing 10 eggs were attached to soybean plants by clipping them to the underside of a fully opened leaf with a paper clip. Ten egg stations were placed in each experimental plot and examined 24 h later for presence/absence and signs of predation. Each egg station was flagged to assist in relocation after the field exposure. Sentinel eggs were placed in the experimental plots on four different dates beginning in late June and ending in late July. Data were arcsine transformed and analyzed using a repeated measures analysis of variance. In 2001, sentinel *Helicoverpa zea* Boddie eggs from an existing laboratory colony were singly placed on soybean foliage using bovine serum albumin (BSA) solution as an adhesive. Fifty individual eggs were placed in the experimental plots on four different dates beginning in early July 2001 and ending in early August. Plants containing eggs were flagged to facilitate relocating the sentinel eggs. Eggs were examined 24 h after placement for absence/presence and signs of predation. Data from all dates were grouped and analyzed using PROC GLM of SAS (SAS Institute 1990).

Soybean looper pupae, collected from a laboratory colony maintained on the Tifton Campus, were exposed in the experimental plots using two different techniques. Circular white cardboard lids, approx. 11 cm², containing an attached pupa were secured to the underside of a soybean leaf. Pupae, either on a cup lid or removed from its webbing, also were placed directly onto the soil surface beneath the soybean canopy. Numbers of pupae placed in each plot varied between 5-20 depending upon number available on a particular date. Pupae were examined 24 h after placement for absence/presence, signs of predation and emergence. Data were combined from all dates and analyzed using PROC GLM of SAS (SAS Institute 1990).

The soybean foliage was sampled weekly beginning in the V1 stage and continued to maturity. Two randomly selected 25-sweep samples were taken down a single row each of all plots with a 38-cm diam net (Kogan and Pitre 1980). Samples were placed into a plastic bag, labeled, returned to the laboratory and frozen. At a later date, samples were processed and the commonly collected arthropods were recorded. Count data for each species or group were analyzed using a repeated measures analysis of variance (SAS Institute 1990). Only sampling dates when selected arthropods were present were used in the analyses. Beginning and ending sampling dates that were included in the analysis for each organism or group in 2000 were as follows: *Geocoris* sp., *Nabis* sp., and spiders (primarily Oxyopidae) 5 July to 12 October, soybean looper and velvetbean caterpillar 16 August to 3 October, green cloverworm 5 July to 3 October, stink bugs 16 August to 12 October, and threecornered alfalfa hopper, *Spissistilus festinus* (Say), 10 August to 12 October. In 2001, sampling dates included in the analyses were very similar to the previous year. From each plot a randomly selected 15.2 × 1.8 m section was harvested with a plot combine each year. The seeds were cleaned and the percent moisture and weight (converted to 13% moisture) recorded. Yield was compared between treatments using an ANOVA (SAS Institute 1990).

Results

In both years the plots with fire ant suppressing treatments had significantly less fire ant foraging than the untreated controls. The control plots had significantly more ants on the hot dog lure than both the chemically-treated plots, with there being slightly more activity in the plots with only hydramethylnon fire ant bait in comparison with plots with bait and chlorpyriphos in 2000 ($F = 328.43$; $df = 2, 12$; $P < 0.0001$) (Fig. 1). In 2001, the untreated control plots also had more foraging activity than either of the ant suppression treatments ($F = 333.13$; $2, 18$ df ; $P < 0.0001$) (Fig. 1). Based on the numbers of fire ants observed on the hot dog lures 1 h after field exposure, it appears that this technique is a viable method of monitoring fire ant activity in the soybean field.

More sentinel soybean looper eggs were recovered from the plots that had been treated with either the hydramethylnon bait plus chlorpyriphos or the hydramethylnon bait only in 2000 ($F = 32.13$; $df = 2, 12$; $P < 0.0001$) (Fig. 2A). The same strong treatment effect also was observed in 2001 with sentinel corn earworm eggs with lower survival after 24 h in the untreated control plots compared with those placed in plots that had been treated with fire ant suppression materials ($F = 6.42$; $2, df = 6$; $P = 0.03$) (Fig. 2B).

A strong treatment effect was observed for the survival of soybean looper pupae in the experimental plots when placed on the plant foliage ($F = 14.67$; $df = 2, 27$; $P < 0.0001$) and on the ground ($F = 11.21$; $df = 2, 18$; $P = 0.0007$) (Table 1). In both placement procedures there was significantly lower recovery of soybean looper pupae in the control plots where fire ant foraging was highest, compared with both of the fire ant suppression treatments.

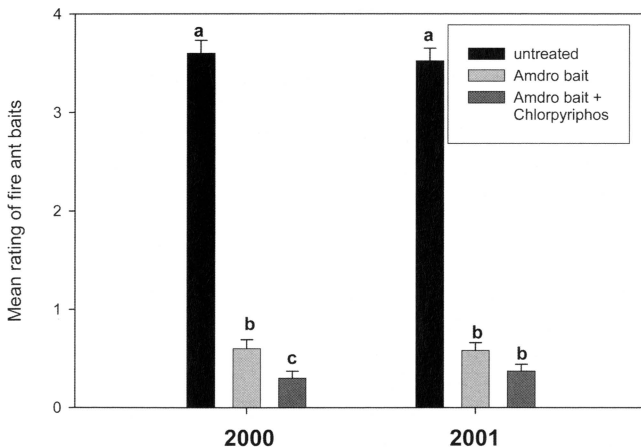


Fig. 1. Mean rating of *S. invicta* foraging in soybean under three different fire ant suppression treatments. Rating scale was as follows: 0: 0 ants, 1: 1-10 ants, 2: 11-50 ants, 3: 51-100 ants, 4: 101-150 ants, 5: >150 ants. Treatments within the same year with identical letters are not significantly different (Ryan-Einot-Gabriel-Welsch multiple range test, $P > 0.05$).

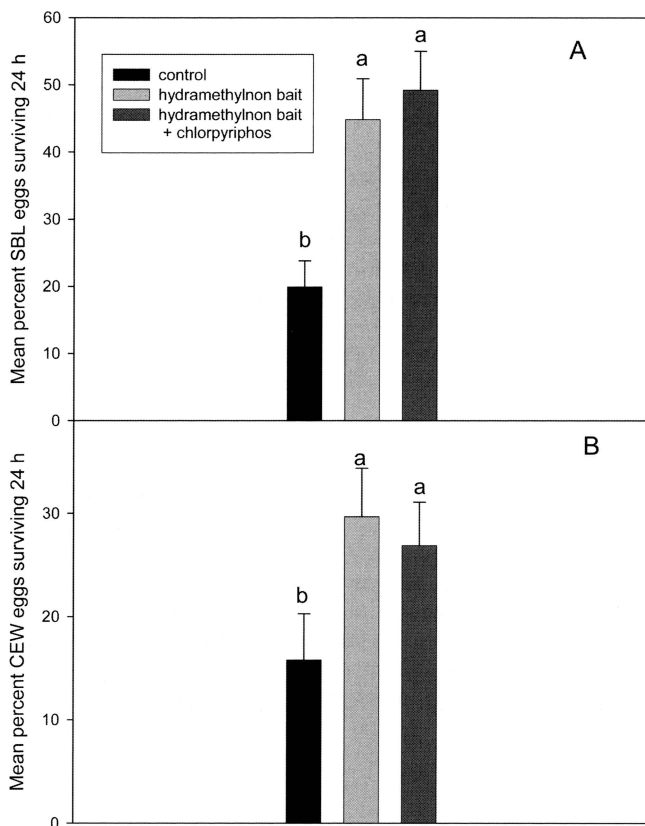


Fig. 2. Survival of lepidopteran eggs after a 24 h field exposure in soybean under three different fire ant suppression treatments. A. Mean survival of soybean looper (SBL) eggs, 2000. B. Mean survival of corn earworm (CEW) eggs, 2001. Identical letters within year denote no difference between treatments (Ryan-Einot-Gabriel-Welsch multiple range test, $P > 0.05$).

The seasonal abundance for the majority of herbivore and natural enemy taxa examined in this study were not affected by the fire ant suppression treatments (Table 2). However, treatment effects were observed in 2001, with fewer green cloverworm and spiders in the experimental plots treated with hydramethylnon bait plus chlorpyrifos than the control plots and the hydramethylnon bait only.

Soybean yield was low in 2000 due to very dry conditions, 856-1300 kg/ha, and no differences were detected between treatments at either farm (Bradford: $F = 0.11$; $df = 2, 4$; $P = 0.9027$; Shannon: $F = 0.52$; $df = 2, 4$; $P = 0.6319$). In 2001, yields were higher, 1367-2795 kg/ha, but like the previous season no differences between yields were detected between treatments (Bradford: $F = 0.27$; $df = 2, 6$; $P = 0.7713$; Shannon: $F = 1.12$; $df = 2, 6$; $P = 0.3866$). Thus, it appears that fire ant suppression has little or no impact on soybean production under Georgia growing conditions.

Table 1. Mean percent survival (\pm standard error) of soybean looper pupae after 24 h exposure in soybean under three different fire ant suppression treatments, Tift Co. GA, 2000 and 2001

Pupal location	Untreated	Hydramethylnon bait	Hydramethylnon bait plus chlorpyrifos	<i>F</i>	<i>P</i>
Foliage	57.8 \pm 10.5b	83.2 \pm 12.9a	91.9 \pm 9.9a	$F_{2,27} = 14.67$	<0.0001
Ground	4.6 \pm 4.6b	30.2 \pm 12.9a	49.3 \pm 18.0a	$F_{2,18} = 11.21$	0.0007

Treatment means for each location followed by the same letter are not significantly different ($P < 0.05$, Tukey's studentized range test).

Discussion

The chemical exclusion techniques used to decrease fire ant foraging and abundance were successful in creating soybean with very low *S. invicta* numbers to be compared with soybean with a naturally high infestation of *S. invicta* (Fig. 1). This corresponds with pitfall captures that were reported from a concurrent study with the very same treatments (Seagraves et al. 2004). Despite this high level of fire ant foraging in the untreated controls, very few fire ants were sampled in the foliage via sweep net. It is unclear if this is because *S. invicta* rarely forages high on soybean plants as reported by Kidd and Apperson (1984), or if there is a diel component to fire ant foraging and time of sampling. The sweep net samples were obtained in late morning each week and possibly temporally avoided capturing fire ants on the soybean plant because their plant foraging activity had ceased (or declined) during that time of day. Soil surface temperatures of 25-30°C are reported as optimum for fire and foraging (Vogt et al. 2003), whereas infrequent foraging was observed below 20°C or above 32°C. It is common in south Georgia to have temperatures above 30°C by late morning, thus the ant foraging could have declined prior to our sweep net sampling.

This study demonstrates that in soybean where *S. invicta* foraging is high there is increased predation on the eggs of pest caterpillars placed in the soybean canopy and soybean looper pupae placed both on the ground and on the soybean foliage. It appears that the increased survival of lepidopteran sessile stages in the chemically treated plots is due to the decreased abundance and activity of *S. invicta*. This is supported by a limited number of direct observations and the fact that there were little or no differences between the treatments in the abundance of other natural enemy taxa that are often cited as predators of lepidopteran eggs (i.e., predaceous bugs and spiders). This agrees with past studies that have cited *S. invicta* as an important component of predation on lepidopteran eggs and pupae in soybean (Buschman et al. 1977, Brown and Goyer 1982, Lee et al. 1990).

Even though there was a large difference in predation on the lepidopteran eggs, there was no difference in the abundance of naturally-occurring caterpillars between the plots with high numbers of foraging *S. invicta* and those that had reduced *S. invicta* numbers. It may be that egg mortality is not a reliable predictor of future larval populations. These results are unclear given that *S. invicta* has been documented to be an important predator of small to medium-sized velvetbean caterpillar larvae (Elvin et al. 1983), which occurred in very high numbers across all treatments in our 2001

Table 2. Seasonal abundance (means per 25 sweeps ± SE) of select arthropods in soybeans under three different fire ant suppressing treatments, Tift County GA 2000 and 2001

Arthropod sampled	Untreated	Hydramethylnon bait		F	P
		Hydramethylnon	Hydramethylnon bait + chlorpyrifos		
2000					
Soybean looper	1.20 ± 0.14a	1.20 ± 0.17a	1.37 ± 0.17a	$F_{2,12} = 0.31$	0.7359
Velvetbean caterpillar	2.66 ± 0.49a	1.96 ± 0.45a	2.54 ± 0.50a	$F_{2,12} = 0.29$	0.7516
Green cloverworm	1.24 ± 0.18a	0.94 ± 0.16a	0.65 ± 0.07a	$F_{2,12} = 2.32$	0.1409
Stink bugs ¹	0.72 ± 0.13a	0.62 ± 0.11a	0.62 ± 0.11a	$F_{2,12} = 0.15$	0.8633
Threecornered alfalfa hopper	2.36 ± 0.23a	2.78 ± 0.32a	3.34 ± 0.27a	$F_{2,12} = 1.41$	0.2821
<i>Geocoris</i> sp.	0.81 ± 0.11a	0.74 ± 0.12a	0.79 ± 0.12a	$F_{2,12} = 0.11$	0.8955
<i>Nabis</i> sp.	0.88 ± 0.10a	0.78 ± 0.09a	0.94 ± 0.13a	$F_{2,12} = 0.56$	0.5842
Spiders ²	0.81 ± 0.07a	0.84 ± 0.09a	0.68 ± 0.07a	$F_{2,12} = 1.25$	0.3225
2001					
Soybean looper	2.49 ± 0.29a	2.17 ± 0.35a	2.27 ± 0.31a	$F_{2,18} = 0.24$	0.7898
Velvetbean caterpillar	11.81 ± 3.64a	11.20 ± 3.41a	11.85 ± 3.74a	$F_{2,18} = 0.09$	0.9146
Green cloverworm	1.47 ± 0.17a	1.69 ± 0.25a	0.97 ± 0.19b	$F_{2,18} = 6.35$	0.0082
Stink bugs ¹	1.38 ± 0.21a	1.28 ± 0.22a	1.44 ± 0.30a	$F_{2,18} = 0.10$	0.9068
Threecornered alfalfa hopper	7.03 ± 1.36a	9.21 ± 2.69a	8.89 ± 1.60a	$F_{2,18} = 0.28$	0.7554
<i>Geocoris</i> sp.	0.15 ± 0.03a	0.16 ± 0.03a	0.16 ± 0.03a	$F_{2,18} = 0.09$	0.9176
<i>Nabis</i> sp.	0.54 ± 0.09a	0.61 ± 0.13a	0.50 ± 0.07a	$F_{2,18} = 0.53$	0.5995
Spiders ²	1.11 ± 0.09a	1.11 ± 0.09a	0.79 ± 0.07b	$F_{2,18} = 4.93$	0.0197

Means in same row followed by same letter are not significantly different ($P > 0.05$, Ryan-Einot-Gabriel-Weisch multiple range test).

¹ Includes adults and nymphs of *Nezara viridula*, *Acrosternum hilare*, and *Euschistus servus*.

² Primarily *Pauceitia viridins* (Araneae: Oxyptidae), but includes other spiders.

sampling. Some insight may be provided by Eubanks (2001) who reported no negative association between caterpillars and *S. invicta* abundance in soybean. However, the Eubanks (2001) study combined all lepidopteran species, and it was concluded that some caterpillar species may be more susceptible to fire ant predation than others.

Stink bugs as a group (southern green, brown, and green stink bugs combined) were also unaffected by the reduction of fire ant foraging. Previous studies have suggested that *S. invicta* is an important natural enemy of stink bugs in soybean, specifically *Nezara viridula* (Ragsdale et al. 1981, Krispyn and Todd 1982, Stam et al. 1987, Eubanks 2001). However, in our season-long reduction of fire ants we saw no increase in the abundance of stink bugs in our monitored experimental plots in either year. Along with the caterpillars and stink bugs, we were unable to demonstrate any change in the abundance of other foliage dwelling insects and spiders. It follows that *S. invicta* spends little time foraging in the soybean canopy or is unable to detect and capture those arthropods we counted in any significant amount.

There was no difference in yield between soybeans with naturally high *S. invicta* abundance or those that had low abundance of *S. invicta* from shortly after planting to maturity. One of the causal mechanisms for yield loss has been ant mounds interfering with harvest machinery (Adams et al. 1983). In our experimental plots, the fire ant mounds were not of sufficient height, less than 0.3 m, plus the soil was too sandy to obstruct the combine.

In conclusion, this study lends further credence to *S. invicta* being a frequent predator of lepidopteran eggs and pupae in southern soybean cropping systems. The study was unable to demonstrate any measurable positive or negative consequence of ambient *S. invicta* populations on soybean yield or biological control of key soybean pests. Since there is no likely benefit to controlling *S. invicta*, when viewed as a potential crop pest, in soybean it should be viewed as a component of the natural enemy complex that should be conserved in cropping systems if possible. Increasing the vegetational diversity and complexity of cropping systems have been ideas tested for increasing the abundance and performance of *S. invicta* as a predator in other cropping systems (Woolwine and Reagan 2001, Harvey and Eubanks 2004). It remains a question if actively enhancing *S. invicta* could have a positive effect on the biological control of key pests in soybean, but is a question worthy of future consideration.

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