2006 Beltwide Cotton Conferences, San Antonio, Texas - January 3 - 6, 2006

CONTROL OF THE TARNISHED PLANT BUG (LYGUS LINEOLARIS) IN COTTON USING THE ENTOMOPATHOGENIC FUNGI, BEAUVERIA BASSIANA AND THE INSECT GROWTH REGULATOR DIAMOND

Donald C. Steinkraus
University Of Arkansas
Fayetteville, AR
Jennifer Lund
Arkansas State University
State University, AR
Tina Gray Teague
University of Arkansas Agricultual Experiment Station - Arkansas State University
State University, AR
Jarrod E. Leland
USDA-ARS-SIMRU
Stoneville, MS

Abstract

Throughout the Delta, insecticides are the sole control method for tarnished plant bug in cotton. Resistance to commonly used pesticides has been noted and the need for alternate control methods is apparent. During the 2005 field season, caged insect experiments were conducted comparing two isolates of *Beauveria bassiana* alone and in combination with the insect growth regulator Novaluron (tradename Diamond®). We observed a greater mortality and more rapid death in Diamond and *Beauveria* treatments compared to untreated controls, but there was no increase in mortality or reduction in days to death when Diamond was combined with *Beauveria.*

Introduction

Tarnished plant bug (*Lygus lineolaris* Palisot de Beauvois) (TPB) is an important pest insect in Midsouth cotton. Current control methods for TPB rely solely on insecticides. Insecticide resistant populations have been noted in the Delta regions from Mississippi up through Arkansas (Hollingsworth et al. 1997, Snodgrass 1996).Effective management alternatives are needed that include efficient, long-lasting and specific biological control agents.

The fungal entomopathogen, *Beauveria bassiana* has been found naturally infecting *Lygus spp*. in Arkansas (Steinkraus and Tugwell 1997), Mississippi (Leland and Snodgrass, 2004), and California (McGuire, 2002) Caged insect trials indicate that the *B. bassiana* can effectively kill 89-100% of adult insects compared to 7-11% in controls (Steinkraus and Tugwell 1997). Previous work has demonstrated that select isolates from *Lygus* spp. are more pathogenic to *Lygus* spp. than GHA (Leland, 2004; Leland et al., 2005; McGuire et al., 2005). Nymph TPB are generally less vulnerable to *B. bassiana*, and therefore the use of a fungal pathogen alone might not provide adequate control of field populations.

Novaluron, (tradename Diamond®) is an insect growth regulator that works by disrupting chitin development and molting. The product shows promise as a new management tool for plant bug nymphs (Barkley and Ellsworth 2004, Smith et al 2004). In this study, a commercially-available *Beauveria* strain (GHA) (Laverlam, SA) along with an experimental strain in production by USDA-ARS were field tested as control agents for suppression of plant bugs alone and in combination with Diamond.

Methods

Diamond and *Beauveria* applications were evaluated in caged insect studies conducted at the University Research Farm on the Judd Hill Plantation near Trumann in NE Arkansas. Cultivar Stoneville 5242 was planted 5 May 2005 in a Dundee silt loam soil. Plots were furrow irrigated. The experiment was arranged in a randomized complete block with 3 replications. Plots for each test were 30 ft long, 3 rows wide and separated by 10 ft alleys; treatment plots were arranged in a RCB with 3 replications.

On the day prior to application, TPB were collected using sweep nets in blooming mustard or wild plant hosts (primarily *Erigeron* spp.). Insects were held overnight at 27°C in cages with water and ears of fresh sweet-corn. For each cage test, 10 organdy sleeve cages, 6 inches diameter by 18 inches long, with 1mmx2 mm openings were secured to randomly-selected individual plants in the center row by tying the lower end of each cage around the plant ca 1 ft from the terminal with twist ties. After sunset, 5 TPB nymphs (3rd to 5th instar) or adults were placed into each cage. There were 5 cages each of TPB nymphs and adults in each plot.

There were six treatments: (a) water control (UTC), (b) the commercially-available *Beauveria* (GHA) at a rate of 1 x 10^{13} conidia per acre, (c) USDA *Beauveria* strain (1 x 10^{13} conidia per acre), (d) Diamond 0.83 EC (12 oz/ac), (e) GHA (1 x 10^{13} conidia per acre) plus Diamond (12 oz/ac) and (f) USDA *Beauveria* (1 x 10^{13} conidia per acre) plus Diamond (12 oz/ac). Applications were made using a 4-row CO_2 charged back pack sprayer calibrated to deliver 11 gpa at 60 psi with TX 10 hollow cone nozzles on 19 inch spacing.

After 48 hrs plants were cut below the cage and taken to the laboratory where TPB were removed and sorted. Dead insects were placed in moist filter paper lined chambers, and living insects were placed individually in 2 oz cups with a 1/2 inch cube of wet florist water foam (Water Foam from Styrofab, Waxahatchie, TX) and a kernel of canned corn. Living insects were held for ten days at 23°C and checked daily for death. Dead insects were checked for molting problems (unable to shed exoskeleton, deformed wings, etc) and outward signs of fungal infection. Results from each of the two experiments were pooled together and ANOVA statistics were used to test the effects of lifestage and treatment on days to death (DTD). Differences in mean DTD were analyzed using Bonferroni adjusted comparisons.

Results

Percent Recovery. We recovered between 85 and 94% of released insects from the cages. We had a lower success rate in recovering nymphs (between 73 and 90% recovered) than in adults (between 89 and 100% recovered) (Figure 1).

Average DTD. Overall there was a significant effect of treatment, stage and the interaction of the two on mean DTD (all $p < .0004$). When looking at each treatment separately, adults had a significantly higher mean DTD than nymphs for all treatments (Bonferroni Adjusted all $p < .0010$). Both UTC nymphs and adults had significantly higher DTDs than all other treatments (all $p < 0.0234$) (Figure 2). There was no difference in the different treatments when looking at nymphs only. For adults, the USDA strain treatments were significantly different than the Diamond (both $p < 0.0012$), while the GHA treatments were not.

Percent Mortality. Initial mortality (to day 2) ranged from 2.9 to 10.3 % for adults and 17.2 to 25.8% for nymphs. Mortality from day 3 to 5 ranged from 4.7 to 34.0 % for adults and 25.5 to 44.6 % for nymphs. Mortality from day 6 to 10 ranged from 45.9 to 64.8 % for adult insects and 23.9 to 40.1 % for nymphs (see Table 1 for cumulative percent mortalities). When nymphs and adults are combined, it appears that initial mortality (to day 2) is consistent between all treatments (ranging between 10.3 to 17.8%), but by day 5 fewer untreated TPB died than in other treatments (25.8 % for UTC, 37.4 to 57.0% for treated). By day 10, mortality was 69.0 % in the UTC compared to 84.8 to 94.5% from Diamond and/or *Beauveria* treatments (see Figure 3).

Percent Sporulation. There was higher percentage of sporulation in insects treated with *Beauveria* over UTC and Diamond only treated insects. Fungal treated insects averaged 41.5 to 55.2 % of insects sporulating, while UTC and Diamond treated insects averaged 2.2 ± 0.9 % of insects and 14.5 ± 5.3 % of insects exhibiting fungal outgrowth. There did appear to be contamination with 4.4% of UTC adults, 13.2% of Diamond only treated adults, and 16.3% of Diamond only treated nymphs exhibiting fungal outgrowth (see Figure 4).

Percent Molting Difficulties. No molting problems were observed in adults. For GHA+ treatments, 3.0 % of nymphs were observed to exhibit molting difficulties compared to 6.5% for USDA+, and 10.1% for

Diamond alone. There was 1 nymph of the 129 insects recovered that showed molting problems in USDA without Diamond treatment. No molting problems were observed in the UTC or GHA without Diamond treatment.

Discussion

Greater mortality and more rapid death were observed in Diamond and *Beauveria* treatments compared to the untreated control.

No increase in mortality or reduction in days to death was measured when Diamond was added to *Beauveria.* There was a slight increase effect on the USDA+ treated adults, which was not significantly different than the USDA alone but was different than the both GHA treated insects. It is unknown why the insect growth regulator affected pathogenicity to adults and not nymphs.

The fungal contamination exhibited in the UTC and Diamond treated insects could be from several different sources. First we used field collected insects in our cage study. *B. bassiana* has been shown to exist in low levels in natural populations (Steinkraus and Tugwell 1997, Leland and Snodgrass 2005) Some of the "contamination" could be simply a result of us using these wild collected individuals. The second explanation could be drift from our spraying. Thirdly, the contamination could have occurred when we were transferring insects from cages to cups.

Future work

We will repeat caged insect studies in 2006. We will expand tests to include season long plant bug sampling and crop monitoring. We will also start studies that will include the use of early season trap crops.

References

Barkley and Ellsworth. 2004. Transitioning Lygus chemical controls to more selective options for Arizona cotton. Arizona Cotton Report. P-138, May: 187-205

Hollingsworth et al. 1997. Responses of Arkansas populations of tarnished plant bugs (Heteroptera: Miridae) to insecticides, and tolerance differences between nymphs and adults. Journal of Economic Entomology. 90 (1): 21-26

Smith et al. 2004. Performance of Diamond (Novaluron) for control of Heliothines and plant bugs, 2003. Proceeding of the 2004 Beltwide Cotton Conferences. 1854-1659

Leland. (in press) Characteristics of *Beauveria bassiana* isolates from *Lygus lineolaris* populations of Mississippi**.** Journal of Agricultural and Urban Entomology.

Leland et al. 2005. Strain selection of a fungal entomopathogen, *Beauveria bassiana*, for control of plant bugs (*Lygus* spp.) (Heteroptera : Miridae). Biological Control. 35, 104-114

Leland and Snodgrass. 2005. Prevalence of naturally occurring *Beauveria bassiana* in *Lygus lineolaris* (Heteroptera: Miridae) populations from wild host plants of Mississippi. Journal of Agriculture and Urban Entomology. 23, 157-163

McGuire. 2002. Prevalence and distribution of naturally occurring *Beauveria bassiana* in San Joaquin Valley populations of *Lygus hesperus* (Heteroptera: Miridae). Journal of Agriculture and Urban Entomology. 19, 237-246.

McGuire et al. 2005. Biological and molecular characteristics of *Beauveria bassiana* isolates from California *Lygus hesperus* (Hemiptera: Miridae) populations. Biological Control. 33,307-314

Snodgrass. 1996. Insecticide resistance in field populations of the tarnished plant bug (Heteroptera: Miridae) in cotton in the Mississippi Delta. Journal of Economic Entomology. 89: 783-790

Steinkraus and Tugwell. 1997. *Beauveria bassiana* (Deuteromycotina: Moniliales) effects on *Lygus lineolaris* (Hemiptera: Miridae). Journal of Entomological Science. 32(1): 79-90

Acknowledgements

Special Thanks to Larry Fowler and staff at the University Of Arkansas Division of Agriculture at Judd Hill for their assistance. The University of Arkansas and Arkansas State University also contributed to this research effort and are acknowledged for their encouragement of innovative cotton management systems. We would also like to thank Dr. Stephen Coghlan for statistical advice and guidance. Finally, we would like to thank the USDA-CSREES for funding this project.

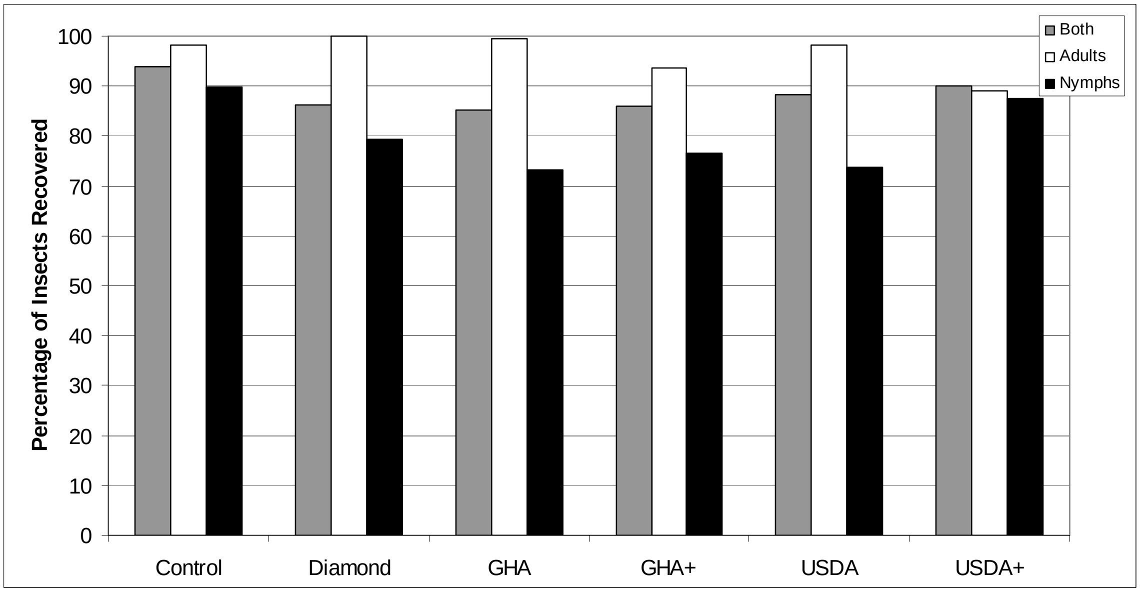

Figure 1: Percent Recovery. Percentage of total insects released into cages that were recovered 2 days post treatment. The percentage includes both dead and living insects recovered.

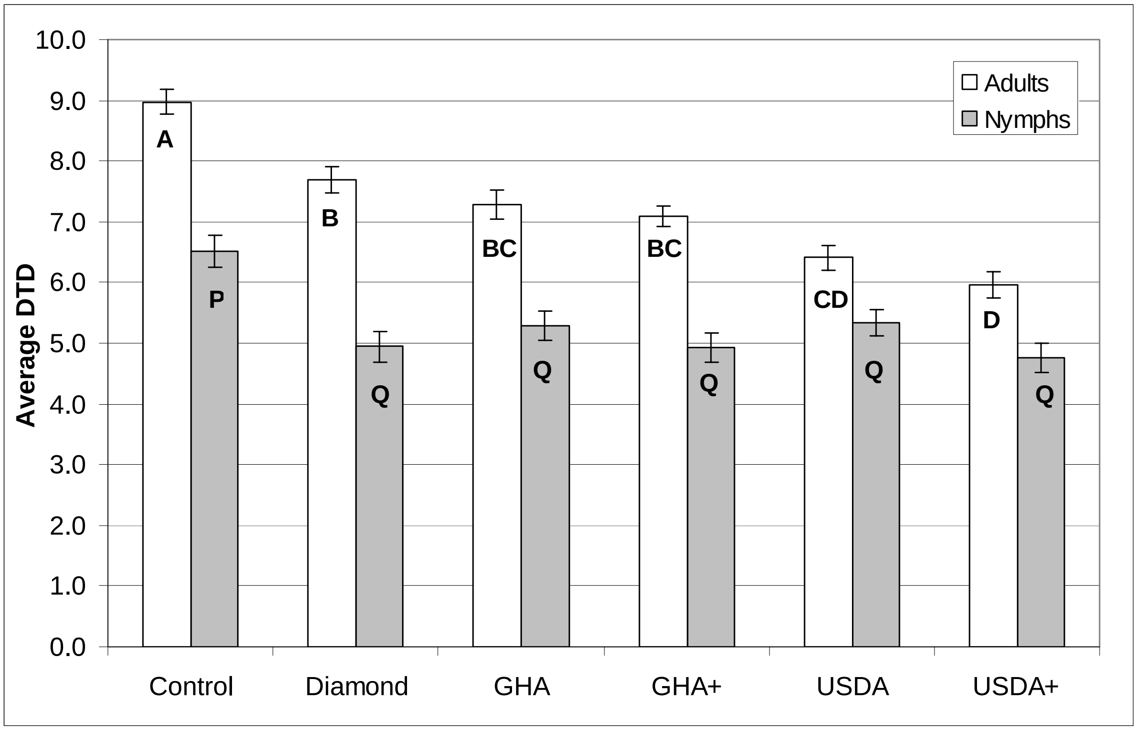

Figure 2: Average Days to Death (DTD). Adults with the same letter (A, B, C, or D) signify no significant difference between treatments. Nymph average DTDs with the same letter (P or Q) represent no significant difference between treatments.

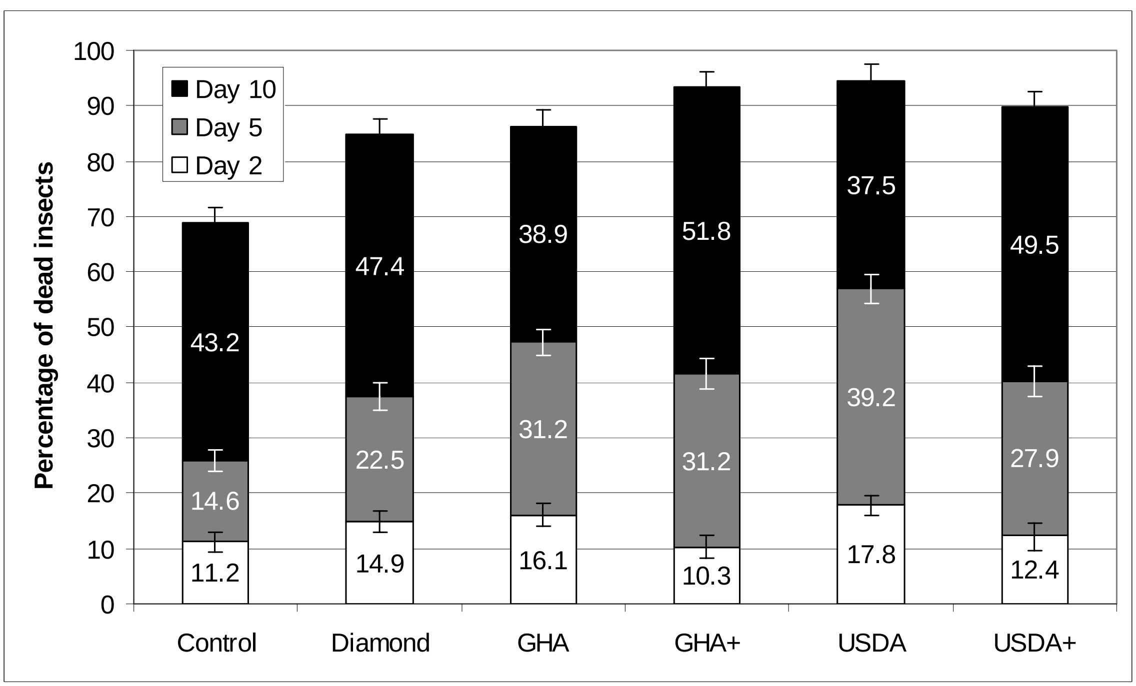

Figure 3: Percentage of Dead Insects. This graph shows the percentage of insects (both nymphs and adults) dying between DAT 0 and 2, and DAT 3 and 5 and DAT 6 to 10.

	Nymphs					Adults				
	N	% Died 2	% Died 5	% Died 10	% Survival	N	% Died 2	% Died 5	% Died 10	% Survival
UTC	157	17.2	42.7	82.8	17.2	172	5.8	10.5	56.4	43.6
Diamond	139	22.3	66.9	97.1	2.9	174	6.9	24.7	79.3	20.7
GHA	128	25.8	54.7	92.2	7.8	164	9.1	29.9	81.1	18.9
GHA+	134	24.6	68.7	92.5	7.5	172	2.9	28.5	91.9	8.1
USDA	129	20.2	58.9	95.3	4.7	156	10.3	44.2	93.6	6.4
USDA+	153	25.5	69.9	95.4	4.6	176	4.5	19.3	84.1	15.9

Table 1: Cumulative Percentage of Dead Insects. This table shows the cumulative percentage of dead adults and nymphs at days 2, 5, and 10. It also includes the % survival of insects for each treatment.

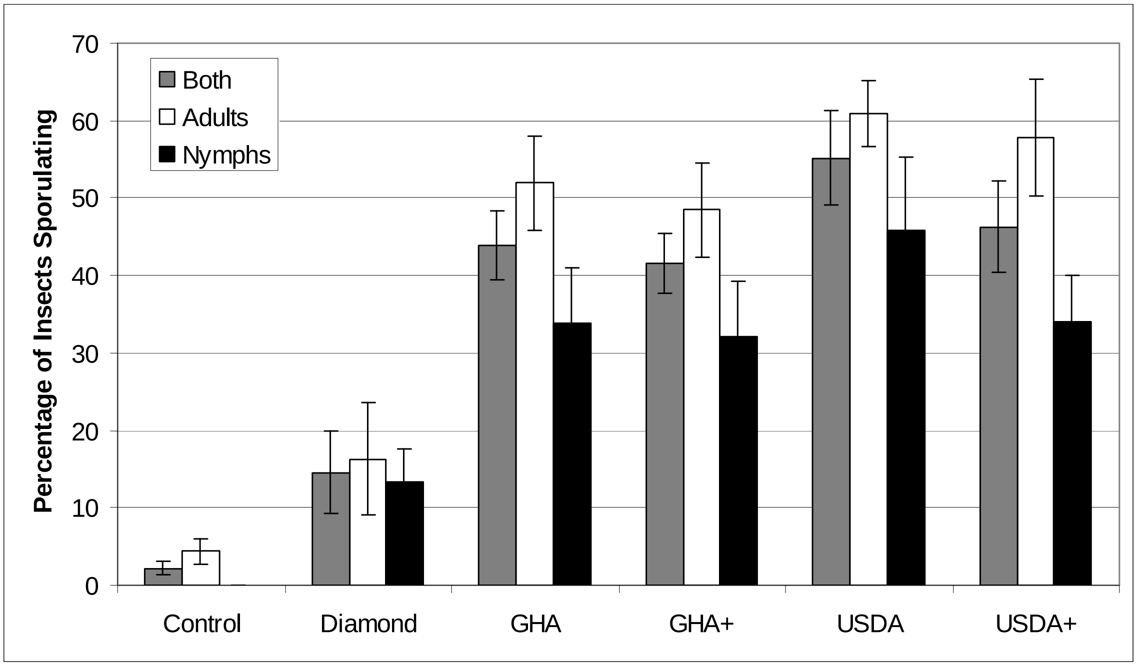

Figure 4: Percent Sporulation. Percentage of nymphs, adults and both exhibiting fungal outgrowth (sporulating).