

Effect of *Bacillus thuringiensis* Transgenic Corn for Lepidopteran Control on Nontarget Arthropods

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ABSTRACT Field populations of nontarget arthropods in transgenic corn with the MON 810 event expressing the Cry1Ab endotoxin from *Bacillus thuringiensis* variety *kurstaki* (*Bt*) were compared with those in conventional, near isogenic corn. The study was conducted at two locations in Georgia in 2001 and 2002 using visual counts, pitfall traps, and corn ear insect evaluations. Results were analyzed by trial using a repeated-measure analysis of variance (ANOVA) and a combined ANOVA of all trials. The only insect whose numbers were strongly affected by the *Bt* corn was the corn earworm, *Helicoverpa zea* (Boddie), a target insect. When averaged over all trials, larvae and adults of sap beetles, *Carpophilus* spp. (mostly *C. lugubris* Murray), and larvae of the otitid fly *Euxesta stigmatis* Loew were less abundant on *Bt* than non-*Bt* corn ears. Kernel damage caused by *H. zea* was less in *Bt* corn, which presumably made *Bt* corn ears less attractive to these insects. There were no consistent significant differences in nontarget phytophagous and predaceous arthropods in the visual counts and pitfall traps between *Bt* and non-*Bt* corn. One exception was *Nabis* spp., which was less abundant in *Bt* than non-*Bt* corn in the combined analysis. Flea beetles, mostly corn flea beetle, *Chaetocnema pulicaria* Melsheimer, were more abundant on *Bt* than non-*Bt* corn in the combined analysis but presumably were not adversely affected by the *Bt* corn. With the exception of nabids, these results indicate that transgenic *Bt* field corn containing the MON 810 event did not have an adverse effect on populations of nontarget phytophagous or predaceous arthropods in the Georgia corn system.

KEY WORDS *Bacillus thuringiensis*, nontarget arthropods, Cry1Ab toxins, soil dwelling arthropods, maize

THE *Bacillus thuringiensis* Berliner (*Bt*) bacterium secretes proteinaceous crystalline Cry endotoxins that are toxic to certain species of pestiferous insects. Susceptibility is determined by specific receptors in the membranous lining of the midgut epithelial cells (Shelton et al. 2002). Specific genes in the *Bt* bacterium coding for the Cry toxins have been inserted into the cells of certain crop plants, like cotton, potatoes, and corn, where the toxins usually are continuously produced throughout the life of the plant (EPA 2001). Corn has been transformed to express the *Bt* endotoxin Cry1Ab, which targets lepidopteran pests.

Transgenic *Bt* corn containing the MON 810 event of the Cry1Ab toxin is marketed as YieldGard *Bt* corn and is available throughout North America (Ostlie et al. 1997, Tenuta et al. 1999). In the corn-growing regions of the midwestern and southern Great Plains of the United States, the main target pests are the European corn borer, *Ostrinia nubilalis* Hübner, and the southwestern corn borer, *Diatraea grandiosella* Dyar. These borers either do not occur or are not econom-

ically important in the coastal plain region of the southeastern United States. In Georgia, the main lepidopteran pests are the corn earworm, *Helicoverpa zea* (Boddie), and the fall armyworm, *Spodoptera frugiperda* (J. E. Smith). *Bt* corn with the MON 810 event suppresses whorl infestations but only partly controls ear infestations of *H. zea* and *S. frugiperda* (Williams et al. 1997, Archer et al. 2001, Buntin et al. 2001, Storer et al. 2001).

The effect of lepidopteran-active *Bt* corn on nontarget arthropods has not been examined in the southeastern United States. Numerous other phytophagous arthropods feed on corn and would be exposed to plant-incorporated Cry toxins. Examples include the common chinch bug, *Blissus leucopterus leucopterus* (Say), the corn flea beetle, *Chaetocnema pulicaria* Melsheimer, corn leaf aphid, *Rhopalosiphum maidis* (Fitch), and several species of leafhoppers, among others (Steffey et al. 1999). A large proportion of the arthropod species present in cornfields is not pestiferous. Many are beneficial natural enemies including numerous species of lady beetles, big eyed bug, *Geocoris* spp., insidious flower bug, *Orius insidiosus* Say, and carabid ground beetles (Steffey et al. 1999). *Bt* corn could potentially have an impact on the popu-

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lations of nontarget arthropod species, both directly and indirectly. Detrimental impacts, if any, could vary widely because of differing levels of sensitivity among the various species present in the corn ecosystem (Wolfenberger and Phifer 2000).

Previous studies have evaluated the potential impact of the transgenic plants expressing the *Bt* Cry toxins on nontarget phytophagous and beneficial species. Most of these studies have shown no significant effect of these toxins on the nontarget arthropods (Sims 1995, Orr and Landis 1997, Pilcher et al. 1997, 2005, Lozzia et al. 1998, Zwahlen et al. 2000, Al-Deeb et al. 2001, Bourguet et al. 2002, Wold et al. 2002). One exception is a series of studies by Hilbeck et al. (1998a, b, 1999), indicating that the Cry1Ab toxin may enhance the mortality of the green lacewing, *Chrysoperla carnea* (Stephens). When *C. carnea* consumed prey that was reared on a diet containing this toxin, *C. carnea* developed at a slower rate, had a lower rate of reproduction, and exhibited a higher mortality rate. However, more recently, Romeis et al. (2004) found no detrimental direct effects of the truncated Cry1Ab toxin on *C. carnea* larvae.

One of the most important components of integrated pest management (IPM) is the preservation of the natural enemies of pestiferous arthropods on the crop system. There is limited understanding of the impact of the Cry toxins expressed in the transgenic plants on the growth, development, and distribution of nontarget arthropods. To ensure that transgenic crops are environmentally sustainable, long-term screenings of these pest management strategies with naturally occurring beneficial arthropods are necessary (Cannon 2000). The objective of this study was to evaluate the effect of *Bt* corn expressing the Cry1Ab toxin on field populations of nontarget phytophagous and beneficial arthropods.

Materials and Methods

Fields studies were conducted at two locations: the Bledsoe Research Farm, Pike County, near Griffin, GA, and the Southwest Georgia Experiment Station, Sumter County, Plains, GA. The experiment evaluated four corn hybrids at each location in 2001 and 2002. Two of the hybrids, 'Pioneer Brand 31B13' and 'Pioneer Brand 32K64' contain the MON 810 event that expresses the Cry1Ab toxin. The other two hybrids, 'Pioneer Brand 3223' and 'Pioneer Brand 32K61', were the conventional and near isogenic lines, respectively, that do not contain the Cry transformation event.

Preparation of the Test Plots. The experimental design was a randomized complete block design with four replicates at both locations. The hybrids were arranged in a factorial design. The plots had the dimensions of 21 by 25 m, with the rows planted 76 cm apart. Plant population was 66,700 plants/ha. The preemergent herbicides atrazine (Aatrex 4L; Syngenta Crop Protection, Greensboro, NC) at 2.24 kg (A.I.)/ha and pendimethalin (Prowl 3.3 EC; BASF, Research Triangle Park, NC) at 1.12 kg (A.I.)/ha were applied to control weeds. The fields were fertilized

preplant at the rate of 112 kg/ha N, 95 kg/ha of P_2O_5 , 95 kg/ha of K_2O , and 168 kg/ha of N as a side dress. Each plot was irrigated as needed during the growing season.

Seedling Stand and Thrips Counts. Plant stand was measured 14 d after planting by counting two adjacent rows per plot. Thrips populations also were examined at 14 and 21 d after planting by collecting 12 plants per plot and dipping them into a cup of 70% ethanol. Thrips were counted, and adults were identified in the laboratory.

Visual Counts. Visual counts of the arthropods on 10 consecutive plants per plot were conducted weekly beginning when the corn plants entered the six-leaf stage. On entering the ear stage of growth, visual counts of arthropods present were conducted weekly until harvest in the ear zone (the primary and secondary ears plus one leaf above and below on the corn stalk) on 20 consecutive corn plants. Samples were randomly selected but were not taken within 4 m of the plot edge. Specimens were periodically collected to verify field identifications.

Soil Arthropod Counts. Pitfall traps were placed in each trial at 1–2 d after planting. Two traps were placed in each plot in 2001, and three traps per plot were used in 2002. Pitfall traps were constructed by placing a 15-cm-diameter cup in a drilled hole in the soil with a smaller plastic 7.6-cm cup containing propylene glycol as a preservative being placed in the bottom of the larger cup (Villa-Castillo and Wagner 2002). A small conical cup had its base removed and placed over the larger cup so the insects would fall into the small plastic cup. The pitfall trap was covered with a 930-cm² wooden cover to exclude rain. Trap contents were emptied weekly during the growing season and taken to the laboratory where contents were placed into vial of 70% ethanol. The contents were examined under a compound microscope, and the arthropods present were identified, counted, and recorded. Pitfall traps were operated from 1 d after planting until 1 wk before harvest.

Corn Ear Insect and Damage Samples. When the corn plants entered the green-silk ear stage, 20 ears per plot in 2001 and 15 ears per plot in 2002 were harvested weekly for 6 wk. The corn ears were returned to the laboratory where they were dissected, and the insects present were identified, counted, and recorded. Insect damage to ears and kernels was evaluated for 20 corn ears harvested per plot on the last sample date. The area of kernels damaged by insect feeding was measured on each ear.

In 2001, all lepidopteran larvae, almost entirely *H. zea*, were collected from the corn ear samples feeding on the *Bt* or non-*Bt* corn. Larval parasitism was assessed by placing larvae individually in plastic diet cups containing meridic diet and rearing them to the adult stage. Parasitism of *H. zea* was not assessed in 2002.

Statistical Analysis. Kernel damage was analyzed by two-way analysis of variance (ANOVA) using PROC MIXED (Littell et al. 1996). Insect count data were transformed by using \log_{10} -transformation before

Table 1. Mean no. thrips on *Bt* and non-*Bt* corn seedlings

| Location and year | Mean \pm SE/plant/ sample date ^a | | F | P |
|-------------------|--|------------------|------|--------------------|
| | Non- <i>Bt</i> | <i>Bt</i> | | |
| Plains 2001 | 11.43 \pm 1.82 | 11.45 \pm 3.00 | 0.01 | 0.993 |
| Griffin 2001 | 2.57 \pm 1.38 | 4.33 \pm 1.86 | 6.70 | 0.029 ^b |
| Plains 2002 | 5.86 \pm 2.93 | 5.46 \pm 2.67 | 0.89 | 0.369 |
| Griffin 2002 | 2.88 \pm 1.06 | 3.55 \pm 1.22 | 0.30 | 0.597 |

^a Sampled at 14 and 21 d after planting.

^b Significant at $P = 0.05$ ($df = 1,9$).

analysis. Results were analyzed by experiment using a repeated-measure ANOVA (RM-ANOVA) for factorial treatment arrangement using PROC MIXED with compound symmetry covariance structure (Littell et al. 1996). Tested fixed effects were replicate, genotype (Ge), *Bt* treatment, sample date, and all two-way interactions. Except for thrips, seasonal mean counts also were analyzed with a combined ANOVA of the four trials using a split-split plot design. Year was the main effect, location was the subplot, and treatments were subsubplots. Year was a fixed effect, and block, block \times year, and block \times year \times location were random effects (Littell et al. 1996). If the covariance estimate for one of the random effects was zero, PROC MIXED used residual error instead of random effect error to test the corresponding main effect.

The *Bt* main effect for all analyses and the *Bt* \times sample date of the RM-ANOVA were used as criteria for assessing effect of *Bt* on each taxon. The *Bt* \times date interaction was considered because if numbers of a taxon tracked differently over time in *Bt* than non-*Bt* plots, thereby causing a significant *Bt* \times date interaction, this may indicate a possible chronic effect of the *Bt* toxin on a particular taxon. The Ge \times *Bt* also was used to indicate whether the *Bt* effect was consistent among genotypes.

Results

Thrips Counts. Thrips collected from seedling corn were mostly corn thrips, *Frankliniella williamsi* Hood. Thrips numbers on *Bt* and non-*Bt* corn were not significantly different in three trials, but thrips were more abundant on *Bt* plants than non-*Bt* plants at Griffin in 2001 (Table 1).

Visual Counts of Nontarget Phytophagous Arthropods. The mean seasonal numbers of arthropod taxa observed in the visual counts on *Bt* and non-*Bt* corn are shown in Table 2. The most abundant phytophagous arthropods observed were chinch bug, corn flea beetle, and leafhoppers. All of these taxa were abundant early in the season. Less abundant taxa were stink bugs, *Euschistus servus* (Say) and *Nezara viridula* L. (Hemiptera: Pentatomidae); sap beetles, mostly *Car-*

Table 2. Mean seasonal no. selected nontarget phytophagous arthropods present on *Bt* and non-*Bt* field corn in visual counts

| Taxa | Location and year | Mean \pm SE/plot/ sample date | | <i>Bt</i> main effect ^a | | <i>Bt</i> \times date interaction ^b | |
|-------------------------------|-------------------|------------------------------------|-----------------|------------------------------------|--------------------|---|--------------------|
| | | Non- <i>Bt</i> | <i>Bt</i> | F | P | F | P |
| Chinch bugs | Plains 2001 | 1.63 \pm 0.42 | 3.15 \pm 0.84 | 7.84 | 0.021 ^c | 2.10 | 0.024 ^c |
| | Griffin 2001 | 0.59 \pm 0.16 | 1.17 \pm 0.26 | 4.36 | 0.066 | 1.30 | 0.232 |
| | Plains 2002 | 0.77 \pm 0.19 | 0.57 \pm 0.15 | 1.90 | 0.201 | 1.02 | 0.435 |
| | Griffin 2002 | 0.44 \pm 0.08 | 0.47 \pm 0.17 | 2.35 | 0.159 | 1.14 | 0.329 |
| | Combined | 0.85 \pm 0.12 | 1.29 \pm 0.22 | 3.01 | 0.090 | — | — |
| Flea beetles | Plains 2001 | 1.69 \pm 0.23 | 1.94 \pm 0.25 | 1.73 | 0.221 | 1.25 | 0.257 |
| | Griffin 2001 | 1.47 \pm 0.17 | 1.86 \pm 0.19 | 3.93 | 0.079 | 0.33 | 0.978 |
| | Plains 2002 | 0.95 \pm 0.14 | 1.08 \pm 0.16 | 0.74 | 0.411 | 0.53 | 0.906 |
| | Griffin 2002 | 3.00 \pm 0.46 | 3.36 \pm 0.54 | 0.01 | 0.961 | 1.66 | 0.080 |
| | Combined | 1.84 \pm 0.15 | 2.11 \pm 0.17 | 4.30 | 0.044 ^c | — | — |
| Leafhoppers | Plains 2001 | 0.56 \pm 0.08 | 0.71 \pm 0.11 | 1.07 | 0.328 | 0.54 | 0.875 |
| | Griffin 2001 | 0.46 \pm 0.08 | 0.40 \pm 0.07 | 2.21 | 0.172 | 0.76 | 0.675 |
| | Plains 2002 | 0.70 \pm 0.13 | 0.80 \pm 0.11 | 1.96 | 0.195 | 0.57 | 0.878 |
| | Griffin 2002 | 3.00 \pm 0.46 | 3.36 \pm 0.54 | 0.01 | 0.961 | 1.66 | 0.080 |
| | Combined | 0.62 \pm 0.05 | 0.64 \pm 0.05 | 0.26 | 0.620 | — | — |
| Stink bugs | Plains 2001 | 0.06 \pm 0.02 | 0.05 \pm 0.22 | 1.64 | 0.233 | 1.19 | 0.300 |
| | Griffin 2001 | 0 | 0 | — | — | — | — |
| | Plains 2002 | 0.22 \pm 0.05 | 0.22 \pm 0.06 | 0.31 | 0.592 | 0.77 | 0.694 |
| | Griffin 2002 | 0.01 \pm 0.01 | 0.04 \pm 0.02 | 2.45 | 0.152 | 0.95 | 0.501 |
| | Combined | 0.08 \pm 0.02 | 0.04 \pm 0.01 | 0.15 | 0.697 | — | — |
| Sap beetles | Plains 2001 | 0.57 \pm 0.21 | 1.09 \pm 0.40 | 1.64 | 0.232 | 1.19 | 0.299 |
| | Griffin 2001 | 0.03 \pm 0.02 | 0.02 \pm 0.04 | 0.06 | 0.814 | 0.71 | 0.731 |
| | Plains 2002 | 0.46 \pm 0.13 | 0.49 \pm 0.11 | 0.42 | 0.534 | 0.77 | 0.687 |
| | Griffin 2002 | 0.13 \pm 0.05 | 0.13 \pm 0.04 | 0.13 | 0.725 | 0.55 | 0.879 |
| | Combined | 0.30 \pm 0.06 | 0.43 \pm 0.10 | 1.41 | 0.242 | — | — |
| <i>Diabrotica</i> spp. adults | Plains 2001 | 0 | 0.02 \pm 0.01 | 1.80 | 0.213 | 2.17 | 0.019 ^c |
| | Griffin 2001 | 0.03 \pm 0.02 | 0.01 \pm 0.01 | 2.25 | 0.168 | 1.73 | 0.073 |
| | Plains 2002 | 0.35 \pm 0.12 | 0.23 \pm 0.06 | 0.78 | 0.399 | 0.57 | 0.874 |
| | Griffin 2002 | 0.25 \pm 0.08 | 0.13 \pm 0.05 | 0.91 | 0.366 | 1.36 | 0.190 |
| | Combined | 0.17 \pm 0.04 | 0.10 \pm 0.02 | 1.82 | 0.184 | — | — |

^a Trial $df = 1,9$; combined $df = 1,41$.

^b Trial: 2001: $df = 12,144$; 2002: $df = 15,180$.

^c Significance at $P < 0.05$.

pophilus lugubris Murray (Coleoptera: Nitidulidae); and adult corn rootworms, *Diabrotica undecimpuncta howardi* Barber and *Diabrotica virgifera virgifera* LeConte (Coleoptera: Chrysomelidae). *Diabrotica v. howardi* was present at both locations but *D. v. virgifera* only occurred at Griffin. Corn leaf aphid was present but was not abundant enough for meaningful analysis in any trial.

The *Bt* main effect and *Bt* × date interactions in each trial and the combined *Bt* main effect were not significant for leafhoppers, stink bugs, sap beetles, and *Diabrotica* spp. adults except for the *Bt* × date interaction of *Diabrotica* spp. adults at Plains in 2001. Chinch bugs were more abundant in *Bt* than non-*Bt* corn, with a significant *Bt* × date interaction at Plains in 2001. However, the *Bt* main effect and *Bt* × date interactions in the other trials and the *Bt* main effect in the combined analysis for chinch bugs were not significant. The *Bt* main effect and the *Bt* × date interactions for flea beetles were not significant in any trial, but the combined *Bt* main effect was significant with flea beetles being more abundant in *Bt* than non-*Bt* corn.

Ge × *Bt* interactions were not significant for stink bugs, sap beetles, and *Diabrotica* adults in any trial ($P > 0.05$) but were significant in one of the four trials for chinch bugs ($F = 6.12$, $df = 12,144$; $P = 0.0354$), flea beetles ($F = 8.83$, $df = 15,180$; $P = 0.0156$), and leafhoppers ($F = 8.39$, $df = 15,144$; $P = 0.0177$). The Ge × *Bt* interaction in the combined analysis was significant for flea beetles ($F = 8.55$, $df = 1,41$; $P = 0.0056$) and leafhoppers ($F = 7.87$, $df = 1,41$; $P = 0.0077$) but was not significant for the other phytophagous taxa.

Visual Counts of Predaceous Arthropods. The mean seasonal numbers of predaceous arthropods in *Bt* and non-*Bt* corn are shown in Table 3. The main predaceous heteropteran taxa observed in the visual counts were insidious flower bug, *Orius insidiosus* (Say), damsel bugs, *Nabis* spp., and big eyed bugs, *Geocoris* spp. *Geocoris* spp. were almost entirely *G. punctipes* (Say), but a few *G. uliginosus* (Say) were observed late in each season. Coccinellid species included *Coleomegilla maculata* DeGeer; convergent lady beetle, *Hippodamia convergens* Guerin-Meneville; seven-spotted lady beetle, *Coccinella septempunctata* L.; and *Scymnus* lady beetle, *Scymnus* spp. Several adults of the multicolored Asian lady beetle, *Harmonia axyridis* (Pallas), also were observed in most trials, but total counts of this species did not exceed four beetles in any trial. The hooded beetle, *Notoxus monodon* (F.) (Coleoptera: Anthicidae), and spiders (Araneae, many species) also were observed.

The mean seasonal numbers of *C. maculata* were not different between *Bt* and non-*Bt* corn in three trials but were more abundant on the *Bt* than non-*Bt* corn plants at Plains in 2002. Numbers of *Geocoris* spp. also were not significantly different between *Bt* and non-*Bt* corn in three trials, but were more abundant on the *Bt* than non-*Bt* corn at Griffin in 2002. The *Bt* main effect of the combined analysis was not significant for these two taxa. Conversely, *O. insidiosus* abundance was not different between *Bt* and non-*Bt* corn in any single

trial, but the combined *Bt* main effect was near significance ($P = 0.0544$), with mean numbers being greater on *Bt* than non-*Bt* corn. Mean seasonal abundance of *Nabis* spp. was small and not significantly different between *Bt* and non-*Bt* corn in any single trial, but when combined over all trials, *Nabis* spp. were significantly more abundant on the non-*Bt* than *Bt* corn. Mean numbers of the other predaceous arthropods did not differ significantly on the *Bt* and non-*Bt* corn in any single trial or in the combined analyses (Table 3).

The *Bt* × date interaction was not significant for any predaceous arthropod in any trial with the exception of *Nabis* spp. at Plains in 2001 (Table 3). The Ge × *Bt* interaction was not significant for any predaceous taxa in the single analyses (2001: $F = 0.01$ – 2.52 , $df = 12,144$; $P = 0.9952$ – 0.1471 ; 2002: $F = 0.00$ – 2.66 , $df = 15,180$; $P = 1.0000$ – 0.1372) or for any of the combined analyses ($F = <0.01$ – 2.00 , $df = 1,41$ – 53 ; $P = 0.9996$ – 0.1644).

Pitfall Trap Samples. The mean seasonal numbers of arthropod taxa recovered in pitfall traps are shown in Table 4. The main taxa recovered were spiders (many species, Araneae); rove beetles, *Lathrobium* spp. and *Sepidophilus* spp. (Coleoptera: Staphylinidae); anthicid beetles, *Notoxus* spp. and *Athicus* spp. (Coleoptera: Anthicidae); the tiger beetle, *Megacephala carolina* L. (Coleoptera: Cicindellidae); carabid ground beetles, primarily *Harpalus pennsylvanicus* DeGeer and *Calasoma sayi* (Say); scarab beetles, *Altaneus* spp., *Cyclocephala immaculata* (Olivier), and *Balbobocerasoma farctus* F. (Coleoptera: Scarabaeidae); click beetles, *Conoderus* spp. (Coleoptera: Elateridae); cucujids, mostly *Ahasverus advena* (Waltl) (Coleoptera: Cucujidae); ants, mostly *Solenopsis invicta* Buren (Hymenoptera: Formicidae); and crickets, *Gryllus pennsylvanicus* Burmeister and *Allenemobius fasciatus* (DeGeer) (Orthoptera: Gryllidae).

The mean seasonal number of most soil-dwelling arthropod taxa did not differ significantly between plots of *Bt* and non-*Bt* corn hybrids (Table 4). At Griffin, staphylinids, anthicids, and ants were more abundant in the non-*Bt* than *Bt* corn in 2001, but the *Bt* main effect was not significant in the other three trials. Conversely, cicindellids were present in higher numbers in the *Bt* corn plots than non-*Bt* corn plots at Griffin in 2002. Abundance of all other soil-dwelling arthropods was not significantly different between the *Bt* and non-*Bt* corn plots in any single trial. The *Bt* main effect was not significant in the combined analysis of any soil-dwelling arthropod taxa (Table 4).

The *Bt* × date interaction was not significant for any taxa in any trial with the exception of cucujids at Griffin in 2002 (Table 4). The Ge × *Bt* interactions also were not significant ($P > 0.05$) for any soil-dwelling taxa in any single trial with the exception of cicindellids at Griffin in 2002 ($F = 5.68$, $df = 15,180$; $P = 0.0441$). Furthermore, Ge × *Bt* interactions in the combined analyses were not significant for any soil-dwelling arthropod taxa ($F = <0.01$ – 3.55 , $df = 1,41$ – 53 ; $P = 0.9664$ – 0.0667).

Table 3. Mean seasonal no. predaceous arthropods in *Bt* and non-*Bt* corn in four trials

| Taxa | Location and Year | Mean \pm SE/plot/sample date | | Bt main effect ^a | | Bt \times date interaction ^b | |
|--------------------------------|-------------------|--------------------------------|-----------------|-----------------------------|--------------------|---|--------------------|
| | | Non- <i>Bt</i> | <i>Bt</i> | <i>F</i> | <i>P</i> | <i>F</i> | <i>P</i> |
| <i>Orius insidiosus</i> | Plains 2001 | 1.29 \pm 0.08 | 1.64 \pm 0.31 | 2.19 | 0.173 | 0.56 | 0.855 |
| | Griffin 2001 | 2.49 \pm 0.42 | 3.27 \pm 0.56 | 4.20 | 0.070 | 0.91 | 0.534 |
| | Plains 2002 | 2.93 \pm 0.38 | 3.17 \pm 0.42 | 0.64 | 0.444 | 0.25 | 0.997 |
| | Griffin 2002 | 1.97 \pm 0.36 | 1.84 \pm 0.29 | 0.05 | 0.822 | 0.86 | 0.586 |
| | Combined | 2.20 \pm 0.18 | 2.49 \pm 0.20 | 3.92 | 0.054 | — | — |
| <i>Geocoris</i> spp. | Plains 2001 | 0.48 \pm 0.09 | 0.59 \pm 0.09 | 1.37 | 0.272 | 0.43 | 0.939 |
| | Griffin 2001 | 0.90 \pm 0.12 | 1.25 \pm 0.14 | 6.57 | 0.031 ^c | 1.66 | 0.087 |
| | Plains 2002 | 1.79 \pm 0.22 | 1.88 \pm 0.24 | 0.01 | 0.936 | 0.79 | 0.673 |
| | Griffin 2002 | 1.07 \pm 0.14 | 0.96 \pm 0.12 | 0.01 | 0.961 | 0.81 | 0.641 |
| | Combined | 1.08 \pm 0.08 | 1.19 \pm 0.08 | 2.22 | 0.144 | — | — |
| <i>Nabis</i> spp. | Plains 2001 | 0.07 \pm 0.03 | 0.02 \pm 0.01 | 2.76 | 0.131 | 3.73 | 0.001 ^d |
| | Griffin 2001 | 0.06 \pm 0.02 | 0.02 \pm 0.01 | 4.50 | 0.063 | 0.98 | 0.466 |
| | Plains 2002 | 0.17 \pm 0.05 | 0.10 \pm 0.03 | 1.47 | 0.257 | 1.41 | 0.159 |
| | Griffin 2002 | 0.02 \pm 0.01 | 0 | 3.00 | 0.117 | 0.91 | 0.538 |
| | Combined | 0.08 \pm 0.02 | 0.04 \pm 0.01 | 8.88 | 0.004 ^d | — | — |
| <i>Hippodamia convergens</i> | Plains 2001 | 0.07 \pm 0.03 | 0.04 \pm 0.03 | 0.71 | 0.421 | 0.71 | 0.725 |
| | Griffin 2001 | 0.01 \pm 0.01 | 0.03 \pm 0.02 | 0.62 | 0.450 | 1.04 | 0.415 |
| | Plains 2002 | 0.18 \pm 0.06 | 0.17 \pm 0.04 | 0.09 | 0.773 | 0.61 | 0.845 |
| | Griffin 2002 | 0.07 \pm 0.03 | 0.09 \pm 0.04 | 0.03 | 0.865 | 0.35 | 0.979 |
| | Combined | 0.09 \pm 0.02 | 0.09 \pm 0.02 | 0.02 | 0.882 | — | — |
| <i>Coleomagilla maculata</i> | Plains 2001 | 0.64 \pm 0.09 | 0.57 \pm 0.09 | 0.19 | 0.673 | 1.07 | 0.387 |
| | Griffin 2001 | 0.78 \pm 0.10 | 0.81 \pm 0.12 | 0.01 | 0.955 | 1.56 | 0.119 |
| | Plains 2002 | 0.44 \pm 0.08 | 0.71 \pm 0.10 | 5.09 | 0.051 ^c | 0.66 | 0.800 |
| | Griffin 2002 | 0.73 \pm 0.11 | 0.75 \pm 0.11 | 0.01 | 0.976 | 1.60 | 0.097 |
| | Combined | 0.64 \pm 0.05 | 0.71 \pm 0.05 | 0.51 | 0.478 | — | — |
| <i>Coccinella septempuncta</i> | Plains 2001 | 0.05 \pm 0.03 | 0 | 1.44 | 0.262 | 1.59 | 0.109 |
| | Griffin 2001 | 0.01 \pm 0.01 | 0 | 1.00 | 0.343 | 1.00 | 0.445 |
| | Plains 2002 | 0.11 \pm 0.05 | 0.08 \pm 0.04 | 0.35 | 0.566 | 1.34 | 0.196 |
| | Griffin 2002 | 0.04 \pm 0.04 | 0.02 \pm 0.01 | 0.01 | 0.915 | 1.08 | 0.380 |
| | Combined | 0.05 \pm 0.01 | 0.03 \pm 0.01 | 1.90 | 0.176 | — | — |
| <i>Scymnus</i> sp. | Plains 2001 | 0.17 \pm 0.07 | 0.16 \pm 0.04 | 0.36 | 0.563 | 0.56 | 0.587 |
| | Griffin 2001 | 0.11 \pm 0.04 | 0.11 \pm 0.06 | 0.13 | 0.723 | 0.89 | 0.547 |
| | Plains 2002 | 0.75 \pm 0.11 | 0.73 \pm 0.10 | 0.01 | 0.992 | 0.70 | 0.759 |
| | Griffin 2002 | 0.27 \pm 0.07 | 0.29 \pm 0.06 | 0.32 | 0.585 | 1.10 | 0.368 |
| | Combined | 0.34 \pm 0.04 | 0.34 \pm 0.04 | 0.17 | 0.680 | — | — |
| Total Coccinellids | Plains 2001 | 0.93 \pm 0.13 | 0.77 \pm 0.10 | 0.55 | 0.479 | 1.29 | 0.239 |
| | Griffin 2001 | 0.92 \pm 0.11 | 0.96 \pm 0.13 | 0.02 | 0.905 | 1.43 | 0.169 |
| | Plains 2002 | 1.48 \pm 0.17 | 1.71 \pm 0.19 | 1.57 | 0.242 | 1.05 | 0.410 |
| | Griffin 2002 | 1.12 \pm 0.13 | 1.16 \pm 0.13 | 0.03 | 0.859 | 1.15 | 0.324 |
| | Combined | 1.13 \pm 0.07 | 0.17 \pm 0.07 | 0.11 | 0.742 | — | — |
| Spiders | Plains 2001 | 0.59 \pm 0.08 | 0.63 \pm 0.09 | 0.02 | 0.889 | 1.12 | 0.352 |
| | Griffin 2001 | 0.85 \pm 0.10 | 0.86 \pm 0.10 | 0.01 | 0.969 | 0.63 | 0.801 |
| | Plains 2002 | 0.77 \pm 0.09 | 0.83 \pm 0.10 | 0.01 | 0.959 | 0.67 | 0.791 |
| | Griffin 2002 | 0.50 \pm 0.07 | 0.55 \pm 0.08 | 0.20 | 0.662 | 0.64 | 0.805 |
| | Combined | 0.68 \pm 0.04 | 0.72 \pm 0.05 | 0.14 | 0.706 | — | — |
| Anthicids | Plains 2001 | 0.05 \pm 0.03 | 0.04 \pm 0.02 | 0.05 | 0.827 | 0.76 | 0.683 |
| | Griffin 2001 | 0.16 \pm 0.05 | 0.20 \pm 0.07 | 0.17 | 0.691 | 0.58 | 0.840 |
| | Plains 2002 | 0.42 \pm 0.10 | 0.54 \pm 0.11 | 2.27 | 0.167 | 0.81 | 0.647 |
| | Griffin 2002 | 0.03 \pm 0.02 | 0.05 \pm 0.02 | 0.56 | 0.536 | 0.49 | 0.916 |
| | Combined | 0.17 \pm 0.03 | 0.22 \pm 0.04 | 1.68 | 0.203 | — | — |

^a df = 1,9.^b 2001: df = 12,144; 2002: df = 15,180.^c Significance at $P < 0.05$.^d Significance at $P < 0.01$.

Corn Ear Insect and Damage Samples. The main taxa observed in corn ears were corn earworm larvae, larvae and adults of sap beetles, *Carpophilus* spp. (mostly *C. lugubris*), and larvae of the otitid fly *Euxesta stigmatis* Loew. Corn earworm numbers peaked during early ear development in the brown-silk stage, whereas sap beetle and *E. stigmatis* numbers increased to a peak when the corn ears reached physiological maturity. Chinch bugs and *O. insidiosus* also were observed in low numbers in corn ears but were not

significantly different between *Bt* and non-*Bt* corn ($F = 0.01$ – 1.71 ; df = 1,9; $P = 0.9797$ – 0.2239).

The seasonal mean numbers of corn earworm larvae were significantly higher in the non-*Bt* than in the *Bt* corn ears in both trials at Plains but were not significantly different in either Griffin trial (Table 5). The mean abundance of corn earworms also was greater in the non-*Bt* than the *Bt* corn when combined over all trials. Furthermore, the *Bt* \times date interaction was significant in all trials. The *Ge* \times *Bt* interaction was not

Table 4. Mean seasonal no. nontarget arthropods collected in pitfall traps in *Bt* and non-*Bt* field corn fields

| Taxa | Location and year | Mean ± SE/plot/sample date | | <i>Bt</i> main effect ^a | | <i>Bt</i> × date interaction ^b | |
|--------------|-------------------|----------------------------|-------------|------------------------------------|--------------------|---|--------------------|
| | | Non- <i>Bt</i> | <i>Bt</i> | <i>F</i> | <i>P</i> | <i>F</i> | <i>P</i> |
| Carabids | Plains 2001 | 0.81 ± 0.11 | 0.79 ± 0.13 | 0.29 | 0.606 | 1.13 | 0.343 |
| | Griffin 2001 | 2.56 ± 0.08 | 2.79 ± 0.36 | 0.01 | 0.960 | 0.54 | 0.898 |
| | Plains 2002 | 0.65 ± 0.08 | 0.98 ± 0.12 | 3.24 | 0.106 | 1.25 | 0.237 |
| | Griffin 2002 | 0.57 ± 0.12 | 0.67 ± 0.14 | 0.29 | 0.604 | 1.37 | 0.164 |
| | Combined | 1.11 ± 0.10 | 1.27 ± 0.11 | 0.34 | 0.564 | — | — |
| Cicindellids | Plains 2001 | 0.64 ± 0.14 | 0.55 ± 0.13 | 0.89 | 0.371 | 0.41 | 0.960 |
| | Griffin 2001 | 0.42 ± 0.08 | 0.52 ± 0.12 | 0.12 | 0.733 | 1.15 | 0.319 |
| | Plains 2002 | 1.83 ± 0.25 | 1.59 ± 0.25 | 2.42 | 0.155 | 0.99 | 0.465 |
| | Griffin 2002 | 0.85 ± 0.20 | 1.65 ± 0.36 | 19.10 | 0.002 ^d | 0.83 | 0.639 |
| | Combined | 0.97 ± 0.10 | 1.12 ± 0.13 | 0.05 | 0.817 | — | — |
| Staphylinids | Plains 2001 | 0.56 ± 0.13 | 0.54 ± 0.10 | 0.12 | 0.742 | 0.66 | 0.790 |
| | Griffin 2001 | 0.23 ± 0.06 | 0.09 ± 0.03 | 6.70 | 0.029 ^c | 0.80 | 0.661 |
| | Plains 2002 | 1.15 ± 0.40 | 1.06 ± 0.14 | 0.72 | 0.419 | 0.84 | 0.633 |
| | Griffin 2002 | 0.48 ± 0.07 | 0.54 ± 0.09 | 0.28 | 0.610 | 0.93 | 0.531 |
| | Combined | 0.62 ± 0.05 | 0.93 ± 0.06 | 0.94 | 0.337 | — | — |
| Anthicids | Plains 2001 | 0.07 ± 0.03 | 0.15 ± 0.09 | 0.44 | 0.525 | 1.39 | 0.174 |
| | Griffin 2001 | 0.03 ± 0.02 | 0 | 24.00 | 0.001 ^d | 1.40 | 0.165 |
| | Plains 2002 | 1.34 ± 0.26 | 1.41 ± 0.15 | 0.41 | 0.537 | 1.04 | 0.177 |
| | Griffin 2002 | 0.23 ± 0.05 | 0.32 ± 0.08 | 0.44 | 0.523 | 0.72 | 0.758 |
| | Combined | 0.42 ± 0.06 | 0.47 ± 0.07 | 0.42 | 0.524 | — | — |
| Scarabs | Plains 2001 | 0.21 ± 0.07 | 0.28 ± 0.08 | 0.63 | 0.448 | 1.81 | 0.052 |
| | Griffin 2001 | 0.15 ± 0.07 | 0.13 ± 0.04 | 0.01 | 0.990 | 0.63 | 0.826 |
| | Plains 2002 | 0.44 ± 0.10 | 0.58 ± 0.12 | 1.44 | 0.261 | 0.71 | 0.772 |
| | Griffin 2002 | 0.16 ± 0.07 | 0.12 ± 0.03 | 0.01 | 0.972 | 0.99 | 0.470 |
| | Combined | 0.24 ± 0.04 | 0.28 ± 0.04 | 0.92 | 0.342 | — | — |
| Cucujids | Plains 2001 | 0.02 ± 0.08 | 0.03 ± 0.09 | 1.05 | 0.333 | 1.37 | 0.215 |
| | Griffin 2001 | 0.01 ± 0.04 | 0.83 ± 1.69 | 0.72 | 0.419 | 0.72 | 0.419 |
| | Plains 2002 | 0.58 ± 0.10 | 0.77 ± 0.13 | 1.29 | 0.285 | 0.92 | 0.544 |
| | Griffin 2002 | 0.28 ± 0.06 | 0.21 ± 0.05 | 1.40 | 0.267 | 2.87 | 0.001 ^d |
| | Combined | 0.22 ± 0.03 | 0.46 ± 0.04 | 3.10 | 0.086 | — | — |
| Elaterids | Plains 2001 | 0.31 ± 0.08 | 0.13 ± 0.04 | 4.00 | 0.077 | 1.50 | 0.129 |
| | Griffin 2001 | 0.05 ± 0.02 | 0.05 ± 0.02 | 0.01 | 0.991 | 1.73 | 0.058 |
| | Plains 2002 | 0.81 ± 0.17 | 0.79 ± 0.15 | 0.17 | 0.693 | 1.22 | 0.262 |
| | Griffin 2002 | 0.15 ± 0.04 | 0.19 ± 0.05 | 0.42 | 0.533 | 0.71 | 0.775 |
| | Combined | 0.34 ± 0.05 | 0.31 ± 0.05 | 0.12 | 0.732 | — | — |
| Crickets | Plains 2001 | 1.16 ± 0.17 | 1.68 ± 0.22 | 4.12 | 0.073 | 0.77 | 0.677 |
| | Griffin 2001 | 3.33 ± 0.45 | 2.39 ± 0.37 | 5.02 | 0.052 | 0.58 | 0.722 |
| | Plains 2002 | 6.65 ± 0.53 | 4.92 ± 0.43 | 2.57 | 0.144 | 0.66 | 0.822 |
| | Griffin 2002 | 5.73 ± 0.63 | 5.93 ± 0.68 | 2.27 | 0.169 | 0.81 | 0.669 |
| | Combined | 4.39 ± 0.27 | 3.87 ± 0.25 | 0.93 | 0.339 | — | — |
| Ants | Plains 2001 | 1.81 ± 0.51 | 1.60 ± 0.40 | 0.01 | 0.908 | 0.65 | 0.797 |
| | Griffin 2001 | 0.15 ± 0.04 | 0.05 ± 0.02 | 5.98 | 0.037 ^c | 1.11 | 0.352 |
| | Plains 2002 | 3.78 ± 0.47 | 3.57 ± 0.48 | 0.36 | 0.562 | 1.51 | 0.103 |
| | Griffin 2002 | 1.90 ± 0.20 | 2.21 ± 0.34 | 0.02 | 0.891 | 0.54 | 0.914 |
| | Combined | 1.97 ± 0.19 | 1.93 ± 0.19 | 0.37 | 0.547 | — | — |
| Spiders | Plains 2001 | 0.97 ± 0.14 | 0.74 ± 0.09 | 1.57 | 0.242 | 0.87 | 0.581 |
| | Griffin 2001 | 0.79 ± 0.12 | 0.60 ± 0.11 | 1.48 | 0.255 | 0.47 | 0.939 |
| | Plains 2002 | 1.48 ± 0.14 | 1.51 ± 0.15 | 0.02 | 0.904 | 0.71 | 0.775 |
| | Griffin 2002 | 1.01 ± 0.13 | 0.80 ± 0.11 | 0.75 | 0.412 | 0.90 | 0.564 |
| | Combined | 1.08 ± 0.07 | 0.93 ± 0.06 | 2.76 | 0.104 | — | — |

^a Trial df = 1,9; combined df = 1,41.

^b 2001: df = 12,144; 2002: df = 15,180.

^c Significance at *P* < 0.05.

^d Significance at *P* < 0.01.

significant in combined analysis (*F* = 0.82, df = 1,41; *P* = 0.3707) or in any single trial except at Plains in 2001 (*F* = 7.62, df = 1,41; *P* = 0.0222). Additionally, no parasitoids were recovered from a total of 3968 *H. zea* larvae that were collected and reared from ear samples in 2001.

The mean number of adult sap beetles was higher in non-*Bt* corn ears than *Bt* corn ears at Plains in 2002. Sap beetles in the other trials and the sap beetle larvae in all trials were not significantly different between *Bt* and non-*Bt* corn. Nevertheless, when averaged over all

trials, sap beetle adults and larvae were significantly more abundant in non-*Bt* than *Bt* corn ears. *Ge* × *Bt* interactions were not significant for sap beetle larvae and adults in the single trials but were significant for adults in the combined analysis (*F* = 12.60; df = 1,41; *P* = 0.0010).

Similarly, the *Bt* main effect and *Bt* × date interactions were not significant for *E. stigmatis* larvae in any trial except at Plains in 2001, where larvae were more abundant on ears of non-*Bt* than *Bt* plants (Table 5). However, in the combined analysis, *E. stig-*

Table 5. Mean seasonal no. target and nontarget arthropods observed in ears of *Bt* and non-*Bt* corn

| Taxa | Location and year | Mean \pm SE/plot/sample date | | <i>Bt</i> main effect ^a | | <i>Bt</i> \times date interaction ^b | |
|--------------------------|-------------------|--------------------------------|------------------|------------------------------------|--------------------|--|--------------------|
| | | Non- <i>Bt</i> | <i>Bt</i> | <i>F</i> | <i>P</i> | <i>F</i> | <i>P</i> |
| Corn earworms | Plains 2001 | 23.8 \pm 1.59 | 18.9 \pm 1.44 | 27.34 | 0.001 ^d | 2.48 | 0.022 ^c |
| | Griffin 2001 | 15.4 \pm 0.79 | 15.9 \pm 0.97 | 0.26 | 0.622 | 2.94 | 0.019 ^c |
| | Plains 2002 | 18.5 \pm 2.05 | 12.6 \pm 2.07 | 47.20 | 0.001 ^d | 7.62 | 0.001 ^d |
| | Griffin 2002 | 16.8 \pm 1.48 | 17.4 \pm 2.32 | 3.43 | 0.095 | 4.35 | 0.001 ^d |
| | Combined | 18.9 \pm 0.80 | 16.5 \pm 0.93 | 45.42 | 0.001 ^d | — | — |
| Sap beetles | Plains 2001 | 84.5 \pm 12.02 | 79.8 \pm 10.94 | 0.19 | 0.674 | 0.58 | 0.770 |
| | Griffin 2001 | 9.2 \pm 2.02 | 7.8 \pm 2.07 | 0.75 | 0.408 | 0.66 | 0.655 |
| | Plains 2002 | 113.8 \pm 15.68 | 96.8 \pm 15.78 | 6.92 | 0.027 ^c | 0.78 | 0.570 |
| | Griffin 2002 | 30.9 \pm 4.13 | 30.3 \pm 4.88 | 1.14 | 0.313 | 0.56 | 0.782 |
| | Combined | 60.7 \pm 5.59 | 54.9 \pm 5.31 | 6.71 | 0.013 ^c | — | — |
| Sap beetle larvae | Plains 2001 | 0.50 \pm 0.36 | 0.08 \pm 0.08 | 1.02 | 0.339 | 0.72 | 0.654 |
| | Griffin 2001 | 5.58 \pm 1.92 | 4.83 \pm 1.58 | 0.91 | 0.366 | 0.45 | 0.810 |
| | Plains 2002 | 9.27 \pm 2.79 | 6.02 \pm 1.55 | 1.92 | 0.199 | 0.84 | 0.554 |
| | Griffin 2002 | 0.69 \pm 0.17 | 1.05 \pm 0.36 | 0.04 | 0.840 | 0.69 | 0.632 |
| | Combined | 4.02 \pm 0.89 | 3.00 \pm 0.71 | 4.78 | 0.035 ^c | — | — |
| <i>Euxesta stigmatis</i> | Plains 2001 | 12.84 \pm 2.73 | 10.91 \pm 2.74 | 5.41 | 0.046 ^c | 0.88 | 0.523 |
| | Griffin 2001 | 3.13 \pm 1.51 | 1.19 \pm 0.43 | 0.42 | 0.535 | 0.69 | 0.630 |
| | Plains 2002 | 9.27 \pm 3.37 | 6.02 \pm 0.20 | 1.92 | 0.199 | 0.41 | 0.842 |
| | Griffin 2002 | 0.70 \pm 0.21 | 1.03 \pm 0.45 | 0.01 | 0.906 | 0.42 | 0.891 |
| | Combined | 6.55 \pm 1.16 | 4.96 \pm 0.96 | 4.76 | 0.035 ^c | — | — |

^a Trial df = 1,9; combined df = 1,41.

^b df = 5,60.

^c Significance at $P < 0.05$.

^d Significance at $P < 0.01$.

matis larvae were significantly more abundant in the non-*Bt* than the *Bt* corn ears. The $Ge \times Bt$ interaction was not significant in any single trial or the combined analysis ($F = 0.88$; $df = 1,41$; $P = 0.3547$).

Corn kernel damage in 2001 was caused mostly by *H. zea*, with minor kernel damage by sap beetles in both trials. Non-*Bt* corn ears had significantly more kernel damage than *Bt* corn ears at Plains (6.67 ± 0.34 and 4.01 ± 0.26 cm², respectively; $F = 16.83$, $df = 1,9$; $P = 0.0027$) and at Griffin (5.17 ± 0.18 and 3.37 ± 0.11 cm², respectively; $F = 11.89$; $df = 1,9$; $P = 0.0073$).

Discussion

In this study, we used the seasonal mean numbers, *Bt* main effects, and *Bt* \times date interactions of each arthropod taxon to compare total population density and trends in populations in *Bt* and non-*Bt* corn over the season. A consistent effect of plant-incorporated *Bt* toxin in corn on a particular taxon could be inferred if the *Bt* main effect of the combined analysis was significant or if *Bt* effect of mean seasonal number in the single trials were significant in more than one of the four trials. Significant *Bt* \times date interactions in more than one trial also may indicate a possible chronic effect of the *Bt* toxin on a particular taxon. By these three criteria, the only arthropod that had significant *Bt* main effects in the combined analysis and more than one single trial and significant *Bt* \times date interactions in several trials was *H. zea*, which is susceptible to the Cry1A toxin and is a target pest. No other arthropod taxa had a similar level of consistently significant differences between *Bt* and non-*Bt* corn.

Populations of other nontarget phytophagous insects that feed on corn plant tissues would have a high

probability ingesting the Cry toxins while feeding on *Bt* corn. While there is no known specific mechanism to expect that plant-incorporated *Bt* toxins would affect any nontarget phytophagous arthropods without specific *Bt* protein binding sites, there is little published data comparing nontarget phytophagous insects in *Bt* and non-*Bt* crops. Flea beetles were more abundant in *Bt* than non-*Bt* corn when averaged over all trials but were not significantly different in any single trial. It is unclear whether the *Bt* effect on flea beetle is real or a statistical or sampling artifact. However with this exception, we found no consistent effect on the phytophagous thysanopteran, heteropteran, and coleopteran taxa examined in this study. Two previous studies (Bourguet et al. 2002, Hassell and Shepard 2002) also found no consistent differences in the numbers of thrips and aphids in the lepidopteran-active *Bt* transgenic and nontransgenic corn. Likewise, Lozzia et al. (2000) found no measurable effect of lepidopteran-active *Bt* toxins in transgenic corn on the twospotted spider mite *Tetranychus urticae* Koch.

In general, no consistent significant differences in populations of foliage-inhabiting predaceous arthropods were found between the *Bt* and non-*Bt* corn. *Orius insidiosus* is a prevalent predator in cornfields in the eastern United States and was the most abundant foliage-inhabiting predatory arthropod in our study. Al-Deeb et al. (2001) found no significant differences in the developmental time, body weight, or length of mature *O. insidiosus* when fed larvae of *O. nubilalis* that had been reared on a mericid diet containing the *Bt* Cry1Ab toxins or on a diet without the toxin. Moreover, they did not observe significant differences of the number of *O. insidiosus* adults and nymphs on *Bt* and non-*Bt* corn in the field. Field studies conducted

by Orr and Landis (1997), Pilcher et al. (1997, 2005), Wold et al. (2002), Hassell and Shepard (2002), and Bourguet et al. (2002) also did not find any significant differences in the field populations of *O. insidiosus* in lepidopteran-active *Bt* transgenic and isogenic susceptible corn. Zwahlen et al. (2000) found no significant differences in the total mortality and developmental time from hatch to adult eclosion of a related species, *Orius majusculus* (Reuter), when fed on the thrips, *Anoprotrothrips obscurus* (Muller), that had consumed *Bt* corn or non-*Bt* corn. The data from our study showed no significant differences in *O. insidiosus* populations in *Bt* and non-*Bt* corn in any single trial but a tendency for greater abundance in *Bt* corn when averaged over all trials. This result and results of previous studies indicate that lepidopteran-active Cry1A toxins in *Bt* corn do not adversely affect *O. insidiosus* populations. The reason for greater abundance of *O. insidiosus* in *Bt* corn in our study is not clear and warrants further study.

Coccinellids also were prevalent in all trials. The most abundant species was *C. maculata* followed by *Scymnus* spp. No consistent differences in populations of coccinellids were observed between *Bt* and non-*Bt* corn. Pilcher et al. (1997) observed no detrimental effects of the lepidopteran-active *Bt* corn toxins on *C. maculata* when fed corn pollen and *O. nubilalis* larvae reared on both *Bt* and non-*Bt* corn. Pilcher et al. (2005) also found no effect of *Bt* corn on field populations of *C. maculata* and *Cycloneda munda* Say. Additionally, Wold et al. (2002) showed no consistent significant differences in field populations of several coccinellids, including *C. maculata*, *H. convergens*, *C. septempunctata*, and *H. axyridis* in *Bt* and non-*Bt* sweet corn. The data from our studies are consistent with these other studies indicating that transgenic corn expressing the lepidopteran-active Cry1Ab toxin did not adversely affect on predatory coccinellid populations.

Nabis spp. is not a prevalent predator in corn (Wiedenmann 1999), and populations were low in all trials of this study. While populations did not differ between *Bt* and non-*Bt* corn in any single trial, nabids were more abundant on non-*Bt* than *Bt* corn when averaged over all trials. Nabids are opportunistic, generalist predators that feed on many prey including lepidopteran eggs and larvae (Wiedenmann 1999). It is possible that nabid numbers were lower in *Bt* corn because of reduced availability of lepidopteran prey. Naranjo (2005) and Whitehouse et al. (2005) also found densities of *Nabis* spp. were lower in *Bt* cotton, most likely because of reduced availability of lepidopteran prey. However, the very low population levels in our study do not permit a definitive comparison of the effect of *Bt* corn on nabids.

The big eyed bug, *G. punctipes*, is a prevalent predatory insect in cornfields in the southeastern United States and was the second most abundant predatory arthropod in our study. The effect of *Bt* corn on *G. punctipes* has not been previously reported. We found no consistently significant differences in the number of *G. punctipes* in visual counts of *Bt* and

non-*Bt* corn. The populations of the hooded beetle and spiders inhabiting foliage also were not significantly different between *Bt* and non-*Bt* corn.

The mean seasonal numbers of arthropods collected in pitfall traps did not differ consistently between *Bt* and non-*Bt* corn for any taxa. Where significant differences occurred they were mostly in the 2001 Griffin trial and significant differences did not occur in the combined analysis of any taxa. The random arrangement of plots at Griffin in 2001 resulted in most *Bt* corn plots being adjacent to a grassy field margin and wooded area, which may have influenced soil-dwelling arthropods in this trial. Lozzia (1999) examined populations of carabid beetles in lepidopteran-active *Bt* and non-*Bt* corn by pitfall traps and found no consistent, significant differences in their abundance. Likewise, Duan et al. (2004) used pitfall traps to compare populations of ground-dwelling arthropods in conventional and coleopteran-active *Bt* transgenic potatoes and found no consistent differences in populations of carabids, staphylinids, and other nonpredatory coleopterans.

The Cry1Ab toxin expressed in the *Bt* corn usually suppresses *H. zea* growth and development but only partially prevents kernel damage by *H. zea* in the corn ears (Buntin et al. 2001, EPA 2001). Storer et al. (2001) examined the life history traits of *H. zea* on *Bt* and non-*Bt* corn hybrids containing either MON 810 or Bt11 events. They found that the mortality of *H. zea* increased to $\approx 75\%$ in *Bt* hybrids as *H. zea* larvae progressed through pupation and adult emergence. Surviving larvae also developed at a slower rate (Storer et al. 2001). We also found fewer *H. zea* larvae and less kernel damage in ears of *Bt* corn than non-*Bt* corn.

Sap beetle larvae and adults feed on kernels near the ear tip. They are considered a major pest of sweet corn but a minor pest of field corn (Kaster 1999). Sap beetles are attracted to corn ears that have been damaged by other insects, especially the activity of *H. zea* (Dowd 2000). Control of nonlepidopteran pests such as sap beetles by the Cry1Ab toxin is not expected. Nevertheless, *Bt* corn may indirectly affect sap beetle numbers, because reduction in kernel damage by lepidopterans may reduce the attractiveness of *Bt* corn to sap beetles. Our data support this hypothesis with small but significant reductions in mean number of sap beetle larvae and adults in *Bt* corn ears when averaged over all trials. Dowd (2000) examined the distribution of sap beetles in *Bt* and non-*Bt* sweet corn in Illinois where damage by corn earworm was reduced $\approx 80\%$ in the *Bt* corn and found that sap beetle numbers were not consistently significantly different in the *Bt* versus non-*Bt* corn. Al-Deeb et al. (2001) also did not find significant differences in the number of sap beetles in ears of *Bt* and non-*Bt* corn. These studies indicate that there is no overt effect of currently deployed Cry1Ab events on *Carpophilus* spp. populations in corn ears. We also did not find a significant difference in sap beetle numbers between *Bt* and non-*Bt* corn in any single trial. However, consistent trends for greater numbers in non-*Bt* corn became significant when averaged over all trials. We found similar results with

E. stigmatis larvae in corn ears. *Euxesta stigmatis* has not been previously studied on *Bt* corn.

The $Ge \times Bt$ interaction provided a measure of whether the *Bt* effect was consistent among corn genotypes. $Ge \times Bt$ interactions were not significant for most taxa including the target pest, indicating that the *Bt* had a similar effect in both corn hybrids. The $Ge \times Bt$ interaction was significant in three cases: flea beetles and leafhoppers on foliage and sap beetle adults in corn ears. This suggests that the *Bt* corn affected these taxa differently in each corn genotype. It also possible that the *Bt* effect occurred in only one of the two corn genotypes, which was related to differences in growth and developmental traits of the corn hybrids used in this study.

In summary, no consistent effects of the transgenic *Bt* corn containing the MON 810 event were observed on almost all nontarget phytophagous and beneficial arthropods evaluated in this study. These results are consistent with findings of most previous studies. Nabids were less abundant in *Bt* than non-*Bt* corn, but low numbers made a robust assessment difficult. Flea beetles were more abundant and *O. insidiosus* also tended to be more abundant on *Bt* than non-*Bt* corn. Although the reason for increased abundance on *Bt* corn is not clear, these taxa presumably were not adversely affect by the *Bt* corn containing the MON 810 event. Understanding of interactions between targeted pest, transgenic plants, and beneficial and other nontarget arthropods, is necessary for successful long-term use transgenic insect-resistant crops.

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