Utility of Ground Beetle Species in Field Tests of Potential Nontarget Effects of Bt Crops

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ABSTRACT Characteristics of ground beetles (Coleoptera: Carabidae), including their distribution, diet breadth, and importance as generalist predators, make them candidates for evaluating potential unintended effects of transgenic crops. The abundance and composition of carabids collected from pitfall traps placed in hybrid dent corn were used to determine which species are consistently present and abundant in Iowa carabid communities and to test for population differences in these species caused by transgenic (*Bacillus thuringiensis* [Bt]) or insecticide-based pest management. Power analyses were also used to evaluate the adequacy of the experimental design. Carabid collections indicate *Harpalus pensylvanicus* DeGeer is the best choice to sample based on its apparent ubiquity and abundance in Iowa and other corn-producing states. However, population levels were time-dependent, and composition of carabid communities differed between locations. Considering the numerical dominance of a few species per field, the two to four most abundant species might be used to effectively represent local carabid communities. *H. pensylvanicus* populations were impacted by insecticide use, but no effects of Bt were found. Power analyses indicated that with the experimental design and replication employed, only large effects were detectable; based on the variation between plots, increased replication is needed to make detection of either moderate (30–50%) or small (<30%) effects likely. The recent release of transgenic corn with coleopteran toxicity highlights the importance of these results when evaluating potential unintended effects of these crops on ground beetles. One species not previously recorded in Iowa, *Lebia pulchella* Dejean, also was collected.

KEY WORDS Carabidae, transgenic corn, insecticide, nontarget, *Bacillus thuringiensis*

CORN VARIETIES COMMONLY REFERRED to as "Bt corn" are transgenic plants containing one or more genes from the naturally occurring soil bacterium *Bacillus thuringiensis* (Berliner) (Bt). *B. thuringiensis* produces crystalline (Cry) proteins that disrupt insect digestive systems (Tanada and Kaya 1993). The insertion of *Bt* genes, which are continuously expressed, provides the plant with season-long protection; consumption of plant tissue expressing a specific Cry protein results in death of target pests. Compared with conventional insecticides, Bt crops may benefit growers by reducing the overall costs and complexity of pest management. Bt corn with resistance to the European corn borer, *Ostrinia nubilalis* (Hübner) (Lepidoptera: Crambidae), was released for commercial use in 1996 and has recently accounted for 32% of the corn area planted in the United States (USDA 2004).

Strains of Bt produce different Cry proteins and have selective activity often confined to a single order of insects. This selectivity is based on the specific conditions (i.e., pH levels, enzymes, and receptors in the gut) necessary to solubilize, activate, and bind a given Bt protein. The earliest varieties of Bt corn were modified with the insertion of lepidopteran-specific *Bt* genes for control of *O. nubilalis.* Because Bt must be ingested to have toxic effects, lepidopteran-specific Bt corn should only cause mortality to *O. nubilalis* or other closely related Lepidoptera feeding on corn; the remainder of the insect community, including beneficial arthropods, should not be harmed.

However, because Bt crops are genetically modified, there are still safety concerns. Research has shown that insects not directly feeding on transgenic plants may be impacted by Bt through other pathways. Consumption of Bt-expressing pollen, persistence of Bt-plant material in soil, and secondary poisoning of predators are all potential causes of detrimental effects to nontarget organisms. Consequently, tests of the potential effects of Bt crops have been performed for many nontarget groups, often focusing on moths and butterflies (Losey et al. 1999, Hellmich et al. 2001, Dively et al. 2004), ladybird beetles (Lundgren and Wiedenmann 2002), green lacewings (Dutton et al. 2003), and soil microorganisms (Al-Deeb et al. 2003).

Among nontarget arthropod groups, ground beetles (Coleoptera: Carabidae) have attributes that suggest

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that they may be valuable indicators of the potential effects of growing transgenic corn. Carabids are ubiquitous and abundant inhabitants of corn fields (Esau and Peters 1975, Best et al. 1981, French et al. 2004), and some species seem closely tied to crop fields and their surrounding borders as habitat (Carmona and Landis 1999, Varchola and Dunn 1999, 2001). The diverse feeding habits of ground beetles, which may be carnivorous, herbivorous, or omnivorous, present potential for exposure to Bt from several sources. Depending on the carabid species, possible dietary sources of Bt toxin include direct exposure through consumption of plant material (seeds, pollen, and corn kernels; Larochelle and Larivière 2003) or indirect exposure from feeding on living or dead invertebrates (collembola, lepidopteran larvae, mites, and other ground beetles; Thiele 1977, Lövei and Sunderland 1996).

Although ground beetles have often been used to show the unintended impacts of insecticide use (Gholson et al. 1978, Kunkel et al. 2001), the recent release of Bt corn with coleopteran-specific toxins (U. S. EPA 2003) suggests increased potential for non-target effects on carabid species and a corresponding need to ensure that experimental methods used are appropriate. In addition, because of variability in composition of carabid communities and in species abundance over time, considering all ground beetles collectively may obscure any real treatment effects for some species. Accordingly, the objectives of this study were to (1) determine which ground beetle species are consistently present and abundant in Iowa corn, (2) test such species for differences in abundance caused by transgenic corn or traditional pest management practices using a standard experimental design (randomized complete block), and (3) determine the scale of sampling programs needed to attain adequate statistical power to detect differences in ground beetle populations.

Materials and Methods

To examine the composition of ground beetle communities in Iowa corn fields, season-long collections were made at two distinct areas representative of relatively diverse (Treynor) and simple (Sorenson) agricultural systems with regard to landscape composition and agronomic practices. In one area (Sorenson), plots were also subjected to different pest management regimes to test for impacts of conventional insecticides or Bt corn production on the abundance of carabid beetle species.

Treynor Area. The Deep Loess Research Station (DLRS) near Treynor, IA, has been maintained by the USDA-ARS as a research farm to study soils, erosion, and hydrology under agricultural production regimes since the 1960s. The topography at DLRS is rolling, and many of the fields are long and narrow (<30 m) in shape to allow for contouring and terracing to prevent erosion. Four fields at DLRS were selected based on their differences with regard to surrounding habitat and management practices and consequent effects on ground beetle populations. All fields were planted to nontransgenic corn hybrids but differed in terms of management and surrounding habitats. Management included fields in traditional rotations of corn and soybeans (fields 1 and 3), corn after 3 yr of alfalfa (field 2), and an ongoing conversion from a corn–soybean rotation to an organic system (field 4). Surrounding vegetation types were soybeans, corn, and alfalfa. Fields near Treynor were located within 2 km of each other and were only sampled in 2001. One or two transects (depending on field width) containing 10 pitfall traps extended into the field beginning with a trap in the first row of corn and continuing every 3 m. Pitfall traps were placed in the field for 7 d on a biweekly basis from June to September. While pitfall traps are generally ineffective for obtaining absolute measures of population density, they are effective for the intended purpose in this study (to make relative comparisons between similar habitats sampled concurrently; see Spence and Niemelä 1994).

Sorenson Area. Plots at the Sorenson farm (located 13 km west of Ames, IA) were located within a single 198 by 190-m field, which had previously been planted to a rotation of corn and soybeans. Surrounding vegetation included grass, trees, and soybeans (2001) or corn (2002). A randomized complete block experimental design with three replications was used. Blocks (61 by 198 m) were separated by 3-m alleyways and divided into three treatment plots that were 80 rows (61 m) wide. Plots were surrounded by five border rows (4 m) of isoline corn. Treatments included Bt corn (Pioneer 34M95; event MON810), a non-Bt isoline (Pioneer 34M94), and the non-Bt isoline treated with permethrin (Pounce 1.5G). Insecticide applications were made on 2 August 2001 and 27 June and 21 August 2002 at the recommended rate of 170 g (A.I.)/ha. Corn was planted on 16 and 9 May 2001 and 2002, respectively. Herbicide applications were made to all plots before planting and at vegetative stages 3–4 (Ritchie et al. 1997). In 2002, treatments were planted in the same locations within the field. Within each plot (80 rows), there were six rows (24, 29, 34, 39, 44, and 49) designated for pitfall trapping. During each sampling period, two rows of every treatment were trapped. The rows were trapped in a rotation of the following pairs: 24 and 39, 29 and 44, and 34 and 49. Each row contained five trapping stations that were 4.5 m apart, for a total of 10 traps per treatment and 30 per block. Pitfall traps were in use for 7 d on a biweekly basis from June through October 2001 or September 2002.

Trap Construction and Establishment. Pitfall traps were made from two 355-ml clear plastic cups (TP12; Solo, Solo Cup Company, Highland Park, IL), and a cover was made from two 25.4-cm-diameter plastic plates (PS15W; Solo). The first plastic cup had holes drilled in the bottom for drainage and was placed into the ground so that the lip was flush with the soil surface. When a trapping station was not being used, a cup filled with soil was placed in the inner cup to keep the hole open and in an attempt to maintain soil conditions around the trap as similar to field conditions as possible. Pitfall trap covers were held together with three bolts so that the long end of the bolts acted as legs supporting the plates. The pitfall traps were placed in the row, and one or two corn plants were removed as needed to accommodate the trap and cover. Pitfall traps were prepared for use by filling a second cup with 50–100 ml of antifreeze and placing it into the buried cup. The soil was leveled out around the trap so that the lip of the inner cup was flush with the ground. The legs of the cover were pushed into the ground over the trap, leaving room (2–3 cm) for insects to move freely under the cover. At collection time, a snap lid was placed over each cup, and the traps were returned to the laboratory for processing. The pitfall traps were kept at 4°C until processed.

Sample Processing. To remove soil and antifreeze, the trap contents were first sifted over a 2.4-mm mesh screen. The large (>3 mm) ground beetle species were removed, rinsed with tap water, and placed in 70% ETOH. The remaining contents were rinsed with tap water over a 45-μm mesh screen to remove fine soil particles. The remaining arthropods and soil were placed in a 125-ml flat-topped modified separatory funnel filled with a saturated (45%) sugar solution. When added to the saturated sugar solution, the arthropods floated, while the soil settled out and was removed. The arthropods remaining in the separatory funnel were poured through a 45-μm screen, rinsed with 70% ETOH, and placed into 70% ETOH along with the large ground beetles removed previously. Samples were held at 4°C until ground beetles were identified using a dissecting microscope. Species were sorted with the assistance of a reference collection identified by Dr. Charles Staines (Department of Systematic Biology-Entomology, Smithsonian Institution) from ground beetles collected during these studies.

Data Analysis. The abundance of carabid species collected from each field was not analyzed statistically but calculated as proportions of the total number of ground beetles collected at each of the fields at Treynor (2001) or within each year at Sorenson (2001 and 2002). Those species that were most common across fields and years were considered as potential species for statistical analysis in Iowa corn.

Repeated-measures analyses of variance (RM-ANOVAs) were conducted to test whether the types of pest management (transgenic or insecticide-based) affected abundance of the most common carabid species collected at the Sorenson plots. For each of the most consistently abundant species from 2001 to 2002 [*Microlestes linearis* LeConte, *Poecilus lucublandus* (Say), and *Harpalus pensylvanicus* DeGeer], separate repeated-measures analyses were conducted for 2001 and 2002, with the mean number of an individual species per trap (within a plot) used as the dependent variable. Because collections in 2001 began several weeks before insecticide treatment, only the last sample before insecticide treatment and all postinsecticide treatment samples were included. The model (Proc Mixed; SAS Institute 1999) included effects of pest management (between-subject effect), time (within-subject effect), and an interaction of treatment and time. Degrees of freedom for *F* tests of model effects were determined using the Kenward-Roger method, which reduces bias for experiments with modest sample sizes (Kenward and Roger 1997). Repeated samples of a plot were related using a heterogeneous first-order autoregressive (ARH[1]) covariance structure, which incorporates both the correlation coefficient, ρ, and the variability within each level of the repeated measure, σ_k, to create a covariance matrix (see Kowalchuk et al. 2004). Dunnett's test was used as a post-ANOVA procedure to compare the abundance of carabids species in untreated isoline corn to either transgenic or insecticidal corn pest management.

Statistical power represents the likelihood that a false null hypothesis (that treatments are similar) will be correctly rejected by a particular test. Specifically, power is defined as $1 - \beta$, where β is the type II error rate for testing a null hypothesis. Both retrospective and prospective power analyses were used on carabid abundance data. Retrospective analysis was used to determine if the experimental design employed in the Sorenson plots was adequate to test for possible effects of pest management on *M. linearis*, *P. lucublandus*, and *H. pensylvanicus.* Because a greater number of samples and insecticide treatments were included in 2002 than in 2001, only these data were used to estimate power. Power analysis software (NCSS 2002) used estimates of the between-subject variability, within-subject variability, and least-squares estimated means to determine the power of treatment effect tests including (1) only transgenic and untreated isoline corn and (2) insecticide-treated and untreated isoline corn. A prospective power analysis estimated the power of similar experiments that might be conducted in the future. This analysis assumed use of a repeated-measures test to detect treatment differences and estimated the likelihood of detecting treatment effects with an increasing number of replicates per treatment. For this procedure, the control was assumed to differ (be greater than) the experimental treatment by either 50 or 30%. The within-subject error and between-subject error terms and the pattern of beetle abundance over time were estimated using *H. pensylvanicus* data from all three treatments.

Results

Abundance of Carabid Species. The relative abundance and total number of carabids collected at each field and year are shown in Table 1. A total of 23 carabid species was found among the four fields at Treynor (2001), with the fewest species (12) collected in field 1, a rotation of corn and soybeans, and the most (20) in field 4, the organic production system. At Sorenson, 32 and 23 carabid species were identified from collections in 2001 and 2002, respectively. Overall, the species captured in the highest numbers were *Elaphropus granarius* (Dejean), *H. pensylvanicus, M. linearis, P. lucublandus,* and *Scarites quadriceps* (Chaudior). However, many species were rare (<1%)

Table 1. Relative species abundance of ground beetles (Coleoptera: Carabidae) collected in pitfall traps in fields near Sorenson and Treynor, IA

Species	Sorenson 2001 $N = 4{,}421$	Sorenson 2002 $N = 3{,}251$	Treynor field 1 $N = 146$	Treynor field 2 $N = 488$	Treynor field 3 $N = 698$	Treynor field 4 $N = 1{,}303$
Agonum decorum Say	<1%	<1%	—	—	—	—
Agonum extensicolle Say	2%	<1%	—	—	—	—
Agonum gratiosum (Mannerheim)	<1%	—	—	—	—	—
Agonum punctipennis Casey	<1%	—	—	—	—	—
Amara angustata Say	<1%	—	—	—	—	—
Amphasia interstitialis Say	—	—	—	—	—	<1%
Anisodactylus carbonarius (Say)	<1%	<1%	—	—	—	—
Anisodactylus harrisi (Say)	—	—	—	<1%	—	<1%
Anisodactylus sanctaerucis (Fabricius)	<1%	<1%	—	—	—	—
Bembidion quadrimaculatum (L.)	<1%	<1%	—	—	—	—
Bembidion rapidum (LeConte)	<1%	2%	—	<1%	<1%	<1%
Calosoma obsoletum Say	—	—	—	<1%	—	—
Chlaenius erythropus Germar	<1%	—	10%	6%	<1%	<1%
Chlaenius lithophilus (Say)	<1%	<1%	<1%	—	—	<1%
Chlaenius nemoralis (Say)	<1%	<1%	—	—	—	—
Chlaenius pensylvanicus Say	<1%	—	3%	<1%	—	<1%
Chlaenius platyderus Chaudoir	<1%	—	—	—	<1%	—
Clivina bipustulata (Fabricius)	1%	3%	1%	—	—	<1%
Colliuris pensylvanicus (L.)	<1%	<1%	—	—	—	—
Dicaelus elongatus Bonelli	<1%	—	—	—	—	—
Dicaelus politus Dejean	<1%	<1%	1%	1%	<1%	<1%
Dyschirius globulosus Say	<1%	—	—	—	—	—
Elaphropus granarius (Dejean)	34%	7%	—	—	—	<1%
Harpalus caliginosus (Fabricius)	<1%	<1%	2%	<1%	2%	3%
Harpalus faunus Say	—	<1%	—	—	1%	<1%
Harpalus herbivagus Say	<1%	—	—	<1%	—	—
Harpalus pensylvanicus DeGeer	11%	25%	49%	43%	80%	89%
Galerita janus Fabricius	<1%	<1%	10%	3%	12%	<1%
Lebia pulchella Dejean	<1%	—	—	—	—	—
Microlestes linearis LeConte	14%	30%	6%	<1%	<1%	<1%
Poecilus lucublandus (Say)	28%	27%	—	<1%	<1%	<1%
Poecilus chalcites (Say)	<1%	<1%	—	—	<1%	<1%
Pterostichus coracinus Newman	<1%	<1%	2%	36%	<1%	<1%
Pterostichus stygicus (Say)	<1%	<1%	<1%	<1%	<1%	<1%
Scarites quadriceps Chaudior	4%	3%	14%	8%	<1%	2%
Stenolophus conjunctus (Say)	<1%	—	—	—	1%	<1%
Stenolophus ochropezus (Say)	<1%	<1%	—	—	—	—

at one or more fields, and the presence and abundance (relative and absolute) of the most common species varied widely both between areas (Sorenson versus Treynor; Table 1) and between fields within an area (Treynor; Table 1; Fig. 1). *Lebia pulchella* Dejean was the only species not previously recorded from Iowa (Bousquet and Larochelle 1993).

Effects of Insecticidal and Transgenic Pest Management. RM-ANOVA of the three most common species at Sorenson showed few differences between treatments when transgenic or insecticidal corn pest management were compared with production of unmanaged (untreated isoline) corn in 2001 and 2002 (Table 2). However, in 2002, significantly more *H. pensylvanicus* were collected in insecticide-treated plots. For each species–year combination tested, beetle abundance was influenced by collection date, as indicated by significant time effects in the analyses (Table 2) and the graphs of abundance over time for *M. linearis, P. lucublandus,* and *H. pensylvanicus* (Figs. 2 and 3).

Statistical Power of Sampling Programs. Retrospective power analyses for the RM-ANOVA conducted on 2002 data indicate that the likelihood of detecting any existing treatment differences was always <0.70. The test for an insecticide effect on *H. pensylvanicus* was the most powerful (probability of detection = 0.60), whereas all other tests of treatment effects in 2002 (insecticide and Bt-based pest management effects for *M. linearis, P. lucublandus,* and Bt effect for *H. pensylvanicus*) showed statistical power of <0.20. Prospective power analyses using data for *H. pensylvanicus* in 2002 indicated that, for a 50% treatment effect, three replicates would only provide a 0.27 likelihood of detection. However, increasing to six or nine replicates would increase the chances to 0.61 or 0.81, respectively (Fig. 4). In general, the 30% effect size was estimated to require twice the replication (to have an equal chance of detection) as the 50% effect.

Discussion

Abundance of Carabid Species. Data on carabid species abundance in Iowa were intended to help determine which species from this family could be sampled adequately for statistical analyses on relative abundance. Detection of any satisfactory species would be useful by allowing an improved focus in studies on nontarget effects. Six collections from different years, areas, and fields (Table 1) indicate that

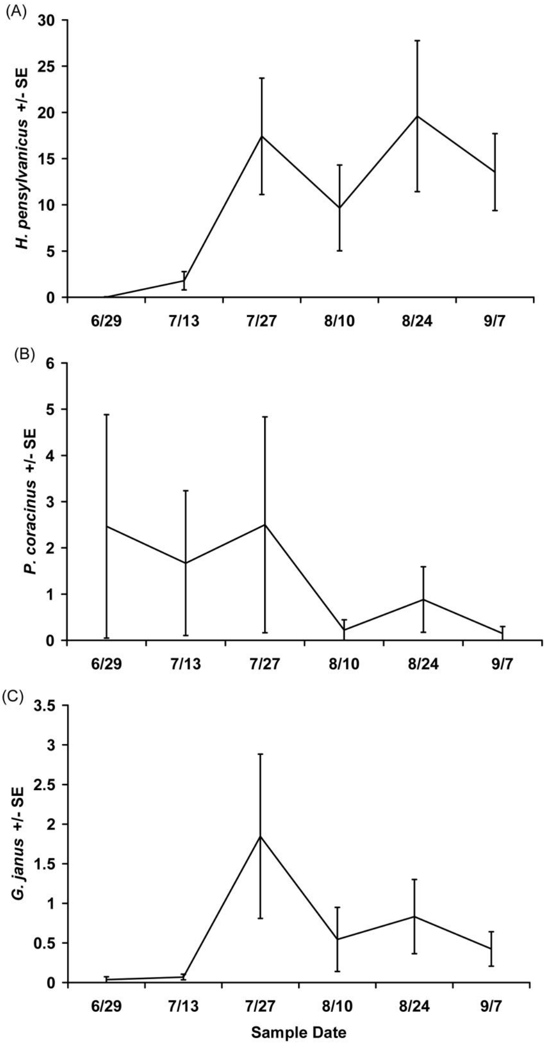

Fig. 1. Mean number of (A) *H. pensylvanicus,* (B) *P. coracinus,* and (C) *G. janus* captured per trap (±SE) from fields at Deep Loess Research Station near Treynor, IA, in 2001.

a small number (1–4) of species probably comprise the majority (80% or more) of beetles present in single site studies. However, different species were also common at each of two geographically distinct locations. Only *H. pensylvanicus* overlapped between lists of the four most abundant species in southwestern (Treynor) and central (Sorenson) Iowa. Previous research in field corn supports the generalization that, while the most abundant species may vary among areas, *H. pensylvanicus* is a common species across Iowa (Esau and Peters 1975, Gholson et al. 1978, Varchola and Dunn 2001). Other studies on carabid communities suggest *H. pensylvanicus* is common in corn and crops rotated with corn in other states (Wiedenmann et al. 1992, Ellsbury et al. 1998, Crist and Ahern 1999, Ward and Ward 2001, Witmer et al. 2003, Hough-Goldstein et al. 2004). Because *H. pensylvanicus* is a

Table 2. Results from RM-ANOVA on common carabid species, 2001–2002

Year	Species	Effect	F	df[a]	P
2001	*H. pensylvanicus*	Treatment	0.68	2, 6.7	0.536
		Time	13.14	5, 8.9	<0.001
		Treatment × time	0.46	10, 9.6	0.878
	M. linearis	Treatment	2.05	2, 9.6	0.182
		Time	6.59	5, 10.0	0.005
		Treatment × time	2.62	10, 10.9	0.065
	P. lucublandus	Treatment	2.10	2, 9.5	0.175
		Time	3.80	5, 9.6	0.037
		Treatment × time	0.49	10, 10.3	0.860
2002	*H. pensylvanicus*	Treatment	4.26	2, 12.5	0.039
		Time	11.50	7, 12.5	<0.001
		Treatment × time	0.96	14, 13.8	0.528
	M. linearis	Treatment	0.77	2, 9.5	0.489
		Time	4.29	7, 10.4	0.018
		Treatment × time	0.25	14, 11.6	0.991
	P. lucublandus	Treatment	0.00	2, 8.8	0.997
		Time	7.15	7, 12.6	0.001
		Treatment × time	0.70	14, 13.9	0.742

[a] Degrees of freedom (numerator, denominator) for indicated tests computed using Kenward-Roger method.

generalist species with an omnivorous feeding habit (Larochelle and Larivière 2003), it should be useful for studying the nontarget effects of transgenic corn by providing several routes of potential exposure to plant-incorporated toxins, including consumption of corn tissues and kernels (Kirk 1973).

Population trends of carabid species over the course of the growing season present a challenge in selecting appropriate species to sample. Both statistical analysis (time effect; Table 2) and visual examination of species graphs (Figs. 1–3) indicate large differences in mean abundance for common beetle species depending on when samples are collected. Although *H. pensylvanicus* populations were consistently abundant late in the season (August), sampling during the early season (June and July) captured mostly *M. linearis, P. lucublandus,* and *E. granarius* (Sorenson) and *S. quadriceps* and *Pterostichus coracinus* Newman (Treynor).

Effects of Insecticidal and Transgenic Pest Management. No significant negative effects of transgenic or insecticidal pest management were found for the three common species tested in either year (Table 2). The only significant treatment effect was an increase in *H. pensylvanicus* collected in insecticide-treated plots during 2002. It is unclear why more *H. pensylvanicus* were collected in the insecticide treatment, but because pitfall traps measure a combination of population density and activity (Thiele 1977), permethrin applications could have simply increased beetle movement without impacting population levels in insecticide-treated plots. Previous research indicates sublethal insecticide exposures may increase predator or parasitoid walking and dispersal behaviors (Wiles and Jepson 1994, Longley and Jepson 1996, Umoru et al. 1996, Singh et al. 2001). An alternate explanation is that *H. pensylvanicus* adults moved into treated plots to scavenge on other arthropod species killed by pesticide treatments. Feeding experiments conducted by

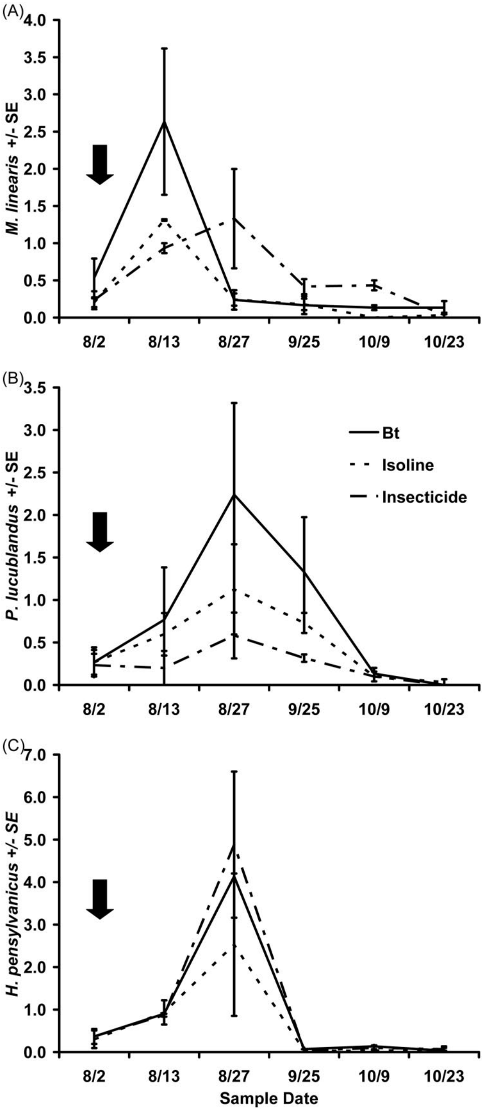

Fig. 2. Mean number of (A) *M. linearis,* (B) *P. lucublandus,* and (C) *H. pensylvanicus* captured per trap (±SE) from transgenic (Bt), insecticide-treated, and untreated isoline plots at Sorenson in 2001. Timing of insecticide application noted by downward arrow.

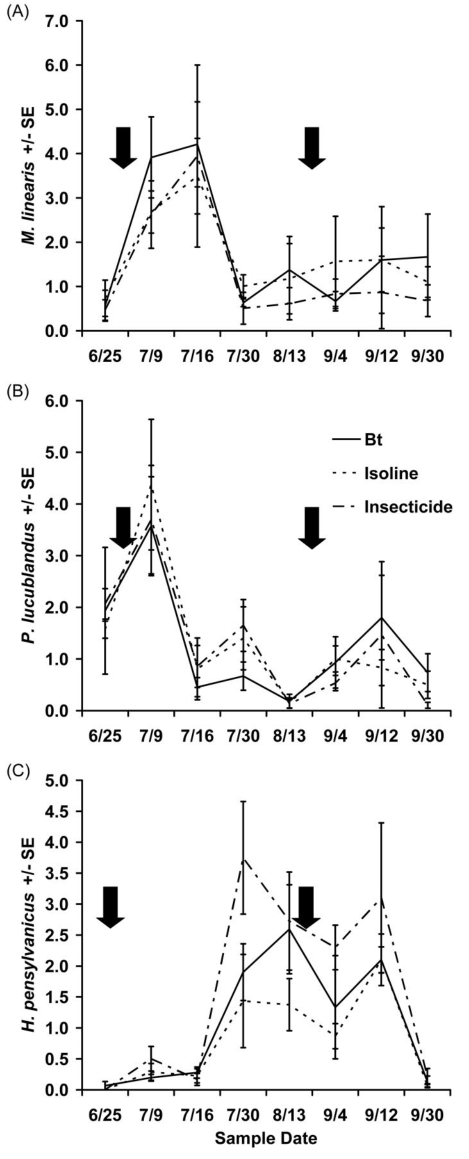

Fig. 3. Mean number of (A) *M. linearis,* (B) *P. lucublandus,* and (C) *H. pensylvanicus* captured per trap (±SE) from transgenic (Bt), insecticide-treated, and untreated isoline plots at Sorenson in 2002. Timing of insecticide applications noted by downward arrows.

Best and Beegle (1977) concluded that *H. pensylvanicus* readily fed on dead insects and often preferred them over live prey. Whatever the cause, increased collection of carabid species in insecticide-treated field plots is sometimes observed (Dixon and McKinlay 1992, Bel'skaya et al. 2002).

The apparent lack of negative treatment effects is surprising considering the published data on the adverse effects of insecticides on ground beetles (Los and Allen 1983, Cárcamo et al. 1995, Lee et al. 2001) but underscores the fact that failure to find significant differences is not equivalent to an absence of effects. The results of the retrospective power analysis confirm that, in cases where negative impact of insecticide applications might be suspected (e.g., *M. linearis* in 2002; Fig. 3A), lack of significance may merely reflect insufficient statistical power. Because the variety of Bt corn planted has lepidopteran-specific toxicity, the lack of significant transgenic treatment effects is not surprising; only indirect effects of Bt (through reduced prey quality) seem possible, and such effects would likely be much smaller in magnitude than impacts from direct toxicity.

Statistical Power of Sampling Programs. Although low power estimates from retrospective analyses may indicate problems in an experimental design, this is not

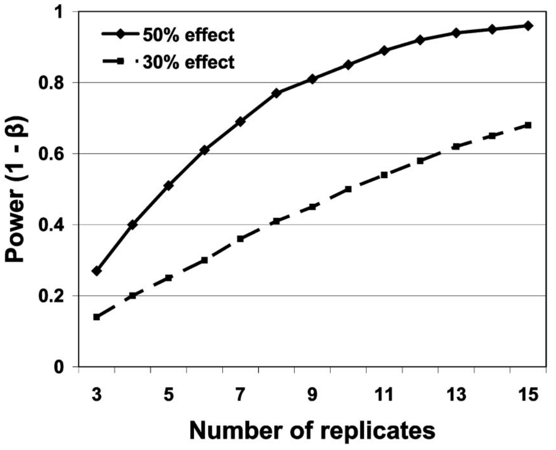

Fig. 4. Results of prospective power analysis indicating the estimated number of replicates needed to detect treatment differences of 50 and 30% relative to a control with a probability of $1 - \beta$.

always the case, because power analysis estimates the likelihood of detecting effects assumed to be present. In some cases, extremely low (<0.20) power may only reflect the similarity of two or more treatments (e.g., the isoline and chemical treatments for *P. lucublandus* in 2002; Fig. 3B). Interpretation of both retrospective and prospective power analyses also is made more difficult by the lack of consensus on what constitutes acceptable power. For nontarget studies, statistical power of 0.70 or higher is often suggested (Marvier 2002, Perry et al. 2003), but this largely depends on what effect size is considered probable or biologically significant (which is also generally unclear; Perry et al. 2003). Regardless, both the retrospective and prospective power analyses using the 2002 ground beetle sampling data lead to two important points. First, when apparent treatment differences are not detected, insufficient replication is a likely cause. Additionally, even field studies with large-to-moderate treatment effects may require much more replication than is commonly used to have a better than 0.50 likelihood of detection.

Conclusions. The release of transgenic crops with coleopteran activity highlights the need to improve both basic knowledge of carabid communities and the methods used to assess possible treatment differences for potentially impacted species. Given (1) the variation in species composition between areas and fields within an area and (2) fluctuations in species abundance during a season, treatment of all carabids collectively (as is sometimes done) may cause problems that outweigh the convenience it affords. For studies in Iowa corn fields, the omnivore *H. pensylvanicus* would be the best species to sample based on its apparent ubiquity and abundance. However, even with *H. pensylvanicus,* spatial and temporal variability suggest that the sensible course of action is to identify and test the most abundant two to four species collected at a site during periods of interest. This can be accomplished using existing reference collections or with the aid of a specialist. With regard to transgenic or insecticide-based pest management, only *H. pensylvanicus* populations were impacted by insecticide use, but no effects of Bt were found. However, with regard to the scale of sampling programs, only large treatment differences (e.g., 100% effect) are likely to be detectable using commonly employed experimental designs and analyses. Based on the variation between replicates (between-subject variability) found in this study, greatly increased replication is needed to make detection of either modest (30–50%) or small (<30%) effects likely.

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