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Seasonality and Adult Habitat Use by Four *Diabrotica* Species at Prairie–Corn Interfaces

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ABSTRACT   Although *Diabrotica* pest species have been well studied in field corn, *Zea mays* L., relatively little is known about adult habitat use by pest and nonpest species at prairie– corn interfaces. Therefore, the objective of this work was to compare seasonal patterns of beetles of four *Diabrotica* species and their use of remnant prairie and adjacent field corn habitats in southeastern Nebraska. The study was conducted at five sites in 2001 and continued at three sites in 2002. The *Diabrotica* species included *D. barberi* Smith and Lawrence, *D. cristata* (Harris), *D. virgifera virgifera* LeConte, and *D. undecimpunctata howardi* Barber. Cucurbitacin vial traps were used to monitor *Diabrotica* populations. Available flowers and *Diabrotica* use of these flowers were also documented. The *virgifera* group species (*D. barberi*, *D. cristata*, and *D. v. virgifera*) were closely tied to a primary habitat, corn or prairie, but moved to secondary habitat if relative attractiveness of food sources in the primary habitat decreased; these three species had high initial population densities that decreased over the season. The one *fucata* group species (*D. u. howardi*) was found in various habitats, with low initial densities that increased over the season. Habitat type and contrasts in plant phenology seem to be key factors that influenced habitat choice by *Diabrotica* species; this especially affected the level of noncorn habitat use by pest species *D. v. virgifera* and *D. barberi*. Use of crop and noncrop parts of the agroecosystem by pest species suggests that a more holistic approach rather than a single field view may be appropriate when managing corn rootworms.

KEY WORDS   northern corn rootworm, western corn rootworm, southern corn rootworm, agroecosystem

Diabrotica (Coleoptera: Chrysomelidae) is a large genus of 354 species native to the Americas, with the greatest number of species found in the tropics (Smith 1966, Wilcox 1972, Krysan 1999). All described *Diabrotica* larvae are root feeders (Branson and Krysan 1981). *Diabrotica* has been historically divided into three species groups: *signifera*, *virgifera*, and *fucata* (Wilcox 1972); only species in the latter two groups occur in the United States (Krysan 1986). The *virgifera* and *fucata* groups have been traditionally separated by host range, life history, and ecological traits (Branson and Krysan 1981, Krysan and Smith 1987, Cabrera Walsh 2003), although the genetic validity of these groups is unclear (Clark et al. 2001, Eben and Espinosa de los Monteros 2004). In the United States, species in the *virgifera* group are univoltine and enter diapause as eggs (Branson and Krysan 1981, Krysan 1986, 1999). These species are generally limited to specific grass hosts as larvae (Branson and Krysan 1981, Krysan 1999, Cabrera Walsh 2003). In contrast, members of the *fucata* group in North America are multivoltine and overwinter in reproductive diapause (Branson and Krysan 1981, Krysan 1986). Larvae in the *fucata* group

are polyphagous, with the potential to use many plant families to complete larval development (Branson and Krysan 1981, Krysan 1986). Very few of the total *Diabrotica* species occur in temperate North America, and many of these are considered to be important crop pests (Krysan 1986). Most of the present knowledge of *Diabrotica* has been obtained from the pest species.

Diabrotica adults are very mobile, with various species capable of both trivial and migratory movement (Arant 1929, Coats et al. 1986, Naranjo 1990), which allows exploitation of various habitats for feeding and oviposition. Movement is often highly correlated with the relative attractiveness of food sources. In general, *Diabrotica* beetles prefer plant pollen and reproductive parts of plants as food sources (Sell 1916, Ludwig and Hill 1975, Krysan and Smith 1987). *Diabrotica virgifera virgifera* LeConte and *Diabrotica barberi* Smith and Lawrence, both members of the *virgifera* group, are pests of corn. Corn pollen and silks are highly attractive to beetles of both species (Branson and Krysan 1981, Prystupa et al. 1988, Darnell et al. 2000), but each has been shown to leave corn for other food sources. *D. v. virgifera* has been found feeding on noncrop flowers, and other crops, such as soybean, *Glycine max* (L.) Merr., alfalfa, *Medicago sativa* L.,

Table 1. Site locations, sizes, and vial trap information

Site	County	Size (ha)	Year	Adjacent corn[a]	Management[b]	Total no. traps	No. transects	Trapping period
Ratzlaff	Hamilton	16.2	2001	Continuous corn	Burning	20	4	July 10 to Oct. 2
Brown	Hall	46.5	2001	Continuous corn	Grazing/haying	18	3	July 10 to Aug. 21
Pearl Harbor	Buffalo	129.5	2001	—	Grazing	15	3	July 10 to Sep. 4
Rivers	Lancaster	12.1	2001	Rotated corn	None	15	3	July 13 to Oct. 5
Spring Creek	Lancaster	202.4	2001	Second year corn	Burning/grazing	28	4	July 13 to Sept. 21
Ratzlaff	Hamilton	16.2	2002	Continuous corn	None	35	7	June 25 to Oct.1
Brown	Hall	46.5	2002	Continuous corn	Grazing	20	4	June 25 to Oct.1
Pearl Harbor	Buffalo	129.5	2002	—	Grazing	20	4	June 25 to Oct.1

[a] The Rivers site had corn adjacent in 2001, rotated annually with soybeans. At Pearl Harbor, the cornfield was separated from the prairie by a road and was not directly adjacent.

[b] Ratzlaff prairie was burned in the spring of 2001 and 2003. Brown prairie was grazed by cattle in the spring of 2001 and hayed in Aug. 2001; the eastern half of the prairie (did not include the study area) was grazed by cattle in the late summer of 2002. Pearl Harbor prairie was continuously grazed by four bison; sections were grazed by cattle in June 2001 and 2002. Alternating sections of Spring Creek prairie were burned in the spring of different years; various areas are grazed by cattle throughout the summer. The study area at Spring Creek was grazed in the spring of 2001.

cucurbits, and sunflowers (Shaw et al. 1978, Siegfried and Mullin 1990, Mabry et al. 2004). *D. barberi* beetles, particularly females, will feed on various crop and noncrop flowers after corn pollination (Forbes 1996, Cinereski and Chiang 1968, Naranjo and Sawyer 1988, McKone et al. 2001). While *D. v. virgifera* may leave corn after corn pollination, *D. v. virgifera* exhibits a preference for corn over other crops and habitats (Naranjo 1994). *D. v. virgifera* often leaves one cornfield for a later-planted cornfield as corn phenology changes (Hill and Mayo 1980, Darnell et al. 2000). If present outside of corn, *D. v. virgifera* is often found at relatively low densities compared with populations in cornfields (Hill and Mayo 1980). *Diabrotica undecimpunctata howardi* Barber, a generalist species in the *fucata* group, does not have a strong preference for corn if other high-quality food, such as pollen, is available (Arant 1929, Lawrence and Bach 1989). Although all three species will feed outside of corn, *D. v. virgifera* and *D. barberi* primarily oviposit in corn in most of their geographic range (Hill and Mayo 1980, Naranjo and Sawyer 1988, Siegfried and Mullin 1990, Boetel et al. 1992).

Native prairies today only exist as fragmented parts of agroecosystems. The prairie ecosystem has been reduced by an estimated 82–99% (Samson and Knopf 1994). Much of what was once prairie has been replaced by row crops; therefore, a high percentage of many prairies is edge habitat surrounded by crops. In eastern and central Nebraska, four *Diabrotica* species are often found in corn and/or natural systems: *D. v. virgifera*, *D. barberi*, *Diabrotica cristata* (Harris), a nonpest prairie species, and *D. u. howardi*, which is not known to overwinter in this region and migrates from the southern United States each year. Relatively little is known about these four species in prairie systems. *D. cristata* is restricted to prairie and grassland habitats east of the Rocky Mountains (Smith 1966, Yaro and Krysan 1986) and feeds on various prairie forbs as an adult (Wiesenborn and Krysan 1980, Krysan and Smith 1987). *Andropogon gerardii* Vitman, big bluestem, is a larval host for *D. cristata*, but other prairie grasses may also serve as larval hosts (Yaro and Krysan 1986). Both *D. barberi* and *D. v. virgifera* can

often be present in noncorn systems, feeding on various forb or weed flowers and pollen (Ludwig and Hill 1975, McKone et al. 2001, Moeser and Vidal 2005). *D. u. howardi* feeds nonspecifically on floral parts and pollen of >280 plant species (Sell 1916, Lawrence and Bach 1989). All four of these species are present in the Nebraska agroecosystem containing corn and remnant prairie, but the seasonal use of these habitats is not well understood.

Therefore, the overall goal of this study was to increase our understanding of the biology and behavior of four *Diabrotica* species that occur in prairie-corn systems in eastern Nebraska. Objectives included (1) for each species, more clearly determine seasonal patterns and adult use of remnant prairie and adjacent field corn habitats, and (2) qualitatively describe adult association with flowering plants during the season.

Materials and Methods

Study Sites, 2001–2002. Field studies were conducted at five sites in 2001 and continued at three of these sites in 2002. Specific prairies were selected for the study because they were adjacent to cornfields; all five were native, unbroken, tallgrass prairies in east and central Nebraska. Prairie size ranged from 12.1 to 202.4 ha (Table 1). Collectively, the sites were typical of native remnant prairies in eastern Nebraska, but some differences in plant species present and prairie management occurred among sites (Table 1).

Traps and Experimental Design. Cucurbitacin vial traps, a modification of the design of Shaw et al. (1984), were used to monitor *Diabrotica* populations at prairie–corn interfaces. Vial traps were baited with buffalo gourd, *Cucurbita foetidissima* HBK, root powder, which contains cucurbitacins E, I, and glycosides (Metcalf and Metcalf 1992). These compounds are movement arrestants and feeding stimulants for the four *Diabrotica* species included in this study (Metcalf and Metcalf 1992). Vial traps were chosen for monitoring for several reasons: (1) vial traps are relatively passive and it was important to minimally disturb beetle movement or habitat choice in this study and (2) vial traps selectively target *Diabrotica*, which was im-
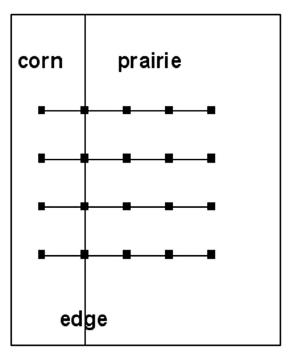
Fig. 1. Transect design used at all prairie–corn sites.

portant in obtaining access to native prairie sites. Acetate sheets were sprayed on both sides with a 1:1 solution (by volume) of Sevin XLR Plus (Bayer CropScience, Research Triangle Park, NC) and water; buffalo gourd root powder was sprinkled onto the sheets. The acetate sheets were later cut into 7.0 by 1.9-cm strips, and one strip was placed in each vial trap. Baited strips were replaced every 7 d at the prairie sites; data from Shaw et al. (1984) showed that similar strips were effective for 12 d in field corn.

Traps were arranged in transects, with each transect starting in corn and ending at an effective center in the prairie (Fig. 1). In each transect, one trap was placed in the cornfield, one on the prairie–corn interface (edge), and a variable number in the prairie (Fig. 1; Table 1). At the Pearl Harbor site, no traps were placed in corn because corn was not directly adjacent to the study location. Traps were located ≈28.3 m apart and were attached to metal fence posts in the prairie and to corn stalks in the cornfield. In corn, traps were placed at ear height; traps were attached at vegetation height in the prairie. In both situations, traps were raised over the season as the corn or vegetation height changed. In 2001, the trapping period was variable per site and was dependent on *Diabrotica* phenology and prairie management constraints (Table 1). In 2002, the study was conducted for 14 wk at all three sites (Table 1). In both years, corn traps were removed 1–2 wk earlier than prairie traps to allow for corn harvest. Temperature and precipitation data were acquired for each site from the nearest weather station (NCDC 2003).

All *Diabrotica* beetles trapped were counted and identified to species on a weekly basis. *Diabrotica*

beetles obtained from the Ratzlaff, Brown, and Rivers sites were also identified to sex (White 1977) on a biweekly basis, starting with beetles collected on 17 July in 2001 and 2 July in 2002. If any species in a given trap sample exceeded 40 individuals, that species' sample was randomly subsampled to 40 individuals, which were identified to sex.

Flowering Plants 2001–2002. In both 2001 and 2002, flowering plants and *Diabrotica* present on flowers were recorded in or near the study area at each site. Flower observations were taken once per week in conjunction with trap monitoring. Flowering plants were identified to genus or species, and *Diabrotica* presence on flowering species was recorded.

Vial Versus Lure Trap: Field Study 2003. Vial trap catches of *D. v. virgifera* were skewed toward males in 2001 and 2002; therefore, a trapping experiment was conducted at the Ratzlaff site in 2003 to determine if the vial trap was providing a clear picture of the sex ratio present in the prairies over time. *D. v. virgifera* was chosen as the target species for this study because of high abundances in both corn and prairie habitats at the Ratzlaff site. Five Trécé Pherocon CRW Kairomone traps (Trécé, Salinas, CA) and five cucurbitacin-baited vial traps were deployed at plant height in alternating fashion through lowland and upland prairie habitats. Each lure and vial trap were separated by ≈45.8 m. Trécé Pherocon lure 8275, which was used in the kairomone traps, was designed to attract *D. v. virgifera;* primary components included 4-methoxycinnamaldehyde and eugenol. 4-methoxycinnamaldehyde has been shown to be highly attractive to *D. v. virgifera* in general (Metcalf and Lampman 1989, Metcalf et al. 1998) and especially to *D. v. virgifera* females (Lance 1993). The lure traps also contained a cucurbitacin-carbaryl stun bait (3.9% carbaryl by content). Cucurbitacin-baited strips used in vial traps were prepared as previously described. Traps were placed in the prairie on 25 July 2003; vial trap baited strips, Pherocon lures, and carbaryl stun baits were changed every 7 d. All *D. v. virgifera* collected were identified to sex on a weekly basis (White 1977).

Statistical Analyses. The PROC MIXED procedure (SAS Institute 2001) was used for all analyses. To compare the overall effect of site and trap location on each species, trap collections at different sites were compared by using the year as a block, and analyzing the factorial of site and general trap location, defined as corn, edge, or prairie. For all analyses, corn represented the commercial cornfield, edge was defined as the interface between prairie and corn, and prairie was defined as the remnant prairie. For specific sites, 2001 and 2002 data were analyzed for *D. barberi, D. v. virgifera,* and *D. u. howardi* as a split-plot in time, with trap location and date in a factorial design and transect as replication. For each species, sites were chosen for analysis if population densities were large enough to allow statistical inferences to be made about habitat choices. *Diabrotica* sex ratio data from 2001 and 2002 were analyzed as the proportion of beetles that were male and as a factorial of site and trap location. The data were not transformed as arcsine square-root

Table 2. Number of species in bloom from each plant family recorded during 2001 and 2002 at each site

Plant family[a]	Sites							
	2001					2002		
	Brown	Pearl Harbor	Ratzlaff	Rivers	Spring Creek	Brown	Pearl Harbor	Ratzlaff
Anacardiaceae			1					1
Apiaceae	1							
Apocynaceae								1
Asclepiadaceae	1		1					1
Asteraceae	8	12	11	13	9	10	11	12
Caesalpiniaceae		1		1			1	
Caprifoliaceae			1		1			1
Convolvulaceae			1		1			1
Euphorbiaceae				1	1			
Fabaceae	5	4	3	1	3	4	4	6
Hypericaceae					1			
Lamiaceae				2				
Lobeliaceae				1				
Malvaceae	1	1	1			1		
Onagraceae			1					1
Poaceae		2	1	2	2	1	1	1
Polygonaceae		1	1	1				1
Rosaceae								1
Solanaceae	1				1			1
Verbenaceae	1	1	1	1	1	1	1	

[a] Plant family names follow classification as presented in the USDA/NCRS PLANTS Database, Version 3.5, National Plant Data Center. (http://plants.usda.gov).

transformation did not improve normality of the data. Data from the 2003 field study at Ratzlaff prairie were analyzed as a split-plot in time, with trap type (lure or vial) and date as a factorial.

Means were separated using a Fisher protected least significant difference (LSD) test. A significance level of $P < 0.10$ was studied for all interactions to avoid using main effects when simple effects were more appropriate. A significance level of $P < 0.05$ was used to separate means in all analyses. Means and SEs for all split-plot designs were obtained from the lsmeans statement in the PROC MIXED procedure (Littell et al. 1996, Steel et al. 1997).

Results

Weather conditions varied greatly over 2001 and 2002. 2000 (before the study) was a dry year; average annual precipitation was 56.3 cm across the five sites, which was 12.5 cm below the 30-yr average (NCDC 2003). Although annual precipitation was normal in 2001 (71.8 cm average across the five sites, 4.1 cm greater than the 30-yr average), the fall of 2001 and winter of 2001–2002 were relatively dry. Precipitation remained low until August 2002; overall, 2002 was considered a drought year in Nebraska. Average precipitation in 2002 at the Ratzlaff, Brown, and Pearl Harbor sites (47.4 cm average) was 19.8 cm below the 30-yr average (NCDC 2003).

Flowering Plants 2001–2002. Fifty-three grass and forb species bloomed at one or more of the five sites during the study periods; approximately one half of the flowering plants were Asteraceae (Table 2; a complete list of plant species, flowering dates, and *Diabrotica* associations with these flowers over time is provided in Campbell 2003). Most flowering plants

were found in various parts of the prairies, although a few exceptions existed. Hoary vervain (*Verbena stricta* Vent.), curlycup gumweed [*Grindelia squarrosa* (Pursh) Dunal], and common sunflower (*Helianthus annuus* L.) were common at multiple sites (Table 2), but were primarily found in edge habitat or disturbed parts of the prairie, such as mowed areas and near dirt roads. At the Ratzlaff site, giant ragweed (*Ambrosia trifida* L.) was mostly confined to the lowland prairie, which was generally moist and occasionally contained irrigation runoff. Giant ragweed was generally less abundant in the Ratzlaff lowland prairie in 2002 than in 2001, possibly because of the invasion of smooth brome (*Bromus inermis* Leyss.) into the lowland area. Corn pollination typically occurred in the third and fourth weeks of July at each site.

***Diabrotica* Presence on Flowering Plants: Observational Data.** All four *Diabrotica* species were observed at least once at each site over 2001 and 2002. Outside of corn, *Diabrotica* were most frequently found on Asteraceae. *D. barberi* and *D. v. virgifera* were present in corn at all sites with corn directly adjacent. Outside of corn, *D. barberi* was observed on flowers of Asteraceae: *Cirsium* sp., *Grindelia squarrosa*, *Helianthus annuus*, *Silphium laciniatum* L., *Vernonia fasciculata* Michx., *Solidago* sp.; and Verbenaceae: *Verbena stricta. D. v. virgifera* was present on flowers of Poaceae: *Setaria glauca* L. Beauv. within cornfields, and flowers of Asteraceae: *Ambrosia trifida*, *Helianthus annuus*, *Helianthus maximilianii* Schrad., *Solidago* sp., and *Vernonia fasciculata* outside of cornfields. *D. u. howardi* was infrequently observed in corn at four sites in 2001 and one site in 2002. Outside of corn, *D. u. howardi* was present on flowers of Asteraceae: *Ambrosia trifida*, *Cirsium* sp., *Conyza canadensis* (L.) Cronquist, *Grindelia squarrosa*, *Helianthus ann-*

Table 3. Mean *Diabrotica* per vial trap (±SE) collected at each site during 2001–2002

Site	Corn	Edge	Prairie
D. barberi			
Ratzlaff	1.7 ± 3.0 C a	0.8 ± 3.0 B a	0.2 ± 3.0 A a
Brown	26.1 ± 3.0 B a	3.5 ± 3.0 B b	0.1 ± 3.0 A b
Pearl Harbor	n/a	1.2 ± 4.6 B a	0.0 ± 4.6 A a
Rivers	52.3 ± 4.6 A a	22.1 ± 4.6 A b	2.5 ± 4.6 A c
Spring Creek	9.5 ± 4.6 C a	0.2 ± 4.6 B a	0.0 ± 4.6 A a
D. v. virgifera			
Ratzlaff	216.5 ± 21.1 A a	17.1 ± 21.1 A b	9.5 ± 21.1 A b
Brown	120.0 ± 21.1 B a	0.7 ± 21.1 A b	0.1 ± 21.1 A b
Pearl Harbor	n/a	1.0 ± 21.1 A a	0.2 ± 21.1 A a
Rivers	9.4 ± 29.9 C a	0.7 ± 29.9 A a	0.1 ± 29.9 A a
Spring Creek	50.9 ± 29.9 BC a	0.4 ± 29.9 A a	0.0 ± 29.9 A a
D. u. howardi			
Ratzlaff	2.1 ± 2.0 A a	4.6 ± 2.0 B a	5.6 ± 2.0 B a
Brown	1.0 ± 2.0 A a	5.5 ± 2.0 B b	1.6 ± 2.0 C a
Pearl Harbor	n/a	3.2 ± 2.0 B a	1.6 ± 2.0 C a
Rivers	3.0 ± 3.0 A a	11.0 ± 2.3 A b	19.3 ± 2.3 A c
Spring Creek	3.0 ± 2.3 A a	3.6 ± 2.3 A a	6.4 ± 2.3 B a

Means within species and column followed by the same capital letter are not significantly different in a one-way analysis of variance ($P >$ 0.05). Means within species and row followed by the same lowercase letter are not significantly different in a one-way analysis of variance ($P >$ 0.05). SEs reported were obtained from PROC MIXED lsmeans statement. Edge is defined as the interface between corn and prairie. Prairie site by location (corn, edge, prairie) interaction was significant for *D. barberi* ($F = 8.33$; df, 7,5; $P = 0.0165$), *D. v. virgifera* ($F = 3.31$; df, 7,5.54; $P = 0.0917$), and *D. u. howardi* ($F = 6.86$; df, 7,7; $P = 0.0105$). Corn was not sampled at the Pearl Harbor site, because the corn was not directly adjacent.

uus, Helianthus maximilianii, Liatris pycnostachya Michx., *Silphium laciniatum, Solidago* sp., *Vernonia fasciculate;* Fabaceae: *Amorpha canescens* Pursh.; Convoluvlaceae: *Convolvulus arvensis* L.; Anacardiaceae: *Rhus glabra* L.; Rosaceae: *Rosa* sp.; and Euphorbiaceae: *Euphorbia marginata* Pursh. Very low densities of *D. cristata* were observed in corn at two sites in 2001 and one site in 2002, only during corn pollination when green silks were present. Outside of corn, *D. cristata* was present on flowers of Asteraceae: *Cirsium* sp., *Echinacea pallida* (Nutt.) Nutt., *Helianthus annuus, Helianthus maximilianii, Ratibida pinnata* (Vent.) Barnhart, *Rudbeckia hirta* L., *Solidago* sp., *Vernonia fasciculata;* Fabaceae: *Amorpha canescens, Dalea purpurea* Vent., *Dalea candida* Michx. ex Willd., *Trifolium pratense* L.; Convolvulaceae: *Convolvulus arvensis;* Rosaceae: *Rosa* sp.; and Caprifoliaceae: *Sambucus canadensis* L. *Amorpha canescens* and the *Dalea* prairie clover species often bloomed before some *Diabrotica* species were present, but *D. cristata,* which was present slightly earlier than *D. barberi* and *D. v. virgifera,* was frequently found on these species.

Diabrotica present on flowers were often observed feeding on pollen and floral plant parts. Also, flowers with *Diabrotica* present frequently exhibited typical signs of *Diabrotica* feeding (e.g., characteristic injury patterns and *Diabrotica* frass). This provides evidence that *Diabrotica* were selecting flowers as feeding sites and that *Diabrotica* presence on a specific flower usually meant that the plant was an acceptable food host.

Vial Trap Data: Overall Species Comparisons 2001–2002. Total beetle densities collected during the study from the vial traps varied greatly by species and site. *D. v. virgifera* was most prevalent at the five sites throughout the study (range: 57–26,308 beetles, mean: 5,481.5 ± 3,188.2), followed by *D. u. howardi* (range: 63–3,266 beetles, mean: 1,162.0 ± 386.9), *D. barberi*

(range: 4–2,769, mean: 631.9 ± 330.0), and *D. cristata* (range: 0–35, mean: 6.4 ± 4.2). Variability of *D. barberi, D. v. virgifera,* and *D. u. howardi* abundances across sites was indicated by significant site by trap location interactions (Table 3). Within corn, abundances of *D. barberi* and *D. v. virgifera* varied significantly among sites (Table 3).

D. barberi was predominantly found in corn (Table 3). In sites directly adjacent to corn, 11.0–50.5% of *D. barberi* were trapped outside of corn (data not shown). *D. barberi* was most abundant at the Rivers site, and the number of beetles that were trapped declined significantly with increasing distance from corn (Table 3). The other three sites with corn directly adjacent to prairie (Brown, Ratzlaff, and Spring Creek sites) exhibited a similar numerical trend, but this trend was not statistically significant (Table 3). The mean number of *D. barberi* trapped in corn and edge habitats at the Rivers site was significantly greater than the mean number trapped in similar habitats at other sites (Table 3). *D. barberi* was rarely trapped at the Pearl Harbor site, the westernmost of the five sites (Table 3).

In both 2001 and 2002, the greatest densities of *D. v. virgifera* were found at the Ratzlaff site (Table 3). The Ratzlaff site also had the highest mean trap collection of *D. v. virgifera* outside of corn, but this trend was not significantly different from other sites (Table 3). At the Ratzlaff and Brown sites, significantly greater densities of *D. v. virgifera* were trapped in corn versus the edge and prairie locations (Table 3); both of these sites were adjacent to continuous corn. Similar numerical trends were evident at the Spring Creek and Rivers sites, but there were no statistical differences among locations at these sites (Table 3).

Generally, the highest proportion of *D. u. howardi* was trapped outside of cornfields (Table 3). At the

Table 4. Overall analysis of variance for each *Diabrotica* species by site and year

Site	Year	Effects	df	*D. barberi*		*D. v. virgifera*		*D. u. howardi*	
				F	P	F	P	F	P
Brown	2001	Week	5,60	4.23	0.0023	8.96	<0.0001	5.60	<0.0001
		trap location	5,10	10.79	0.0009	13.62	0.0003	2.81	0.0773
		Week × trap location	25,60	5.75	<0.0001	8.71	<0.0001	2.20	0.0067
Brown	2002	Week	13,186	7.33	<0.0001	7.16	<0.0001	16.85	<0.0001
		Trap location	4,12	14.81	0.0001	20.41	<0.0001	5.32	0.0106
		Week × trap location	49,186	7.65	<0.0001	7.53	<0.0001	7.80	<0.0001
Pearl Harbor	2001	Week	7,68	2.30	0.0366	4.91	0.0002	9.48	<0.0001
		Trap location	4,8	0.87	0.5201	22.93	0.0002	4.07	0.0435
		Week × trap location	28,68	0.97	0.5187	5.06	<0.0001	1.93	0.0145
Pearl Harbor	2002	Week	13,195	—	—	18.76	<0.0001	12.54	<0.0001
		Trap location	4,12	—	—	1.04	0.4267	0.96	0.4651
		Week × trap location	52,195	—	—	1.19	0.2031	0.97	0.5332
Ratzlaff	2001	Week	11,152	7.15	<0.0001	17.10	<0.0001	5.01	<0.0001
		Trap location	4,12	24.68	<0.0001	85.12	<0.0001	2.55	0.0934
		Week × trap location	42,152	2.73	<0.0001	12.65	<0.0001	1.92	0.0023
Ratzlaff	2002	Week	13,387	5.40	<0.0001	53.61	<0.0001	19.00	<0.0001
		Trap location	4,21	18.22	<0.0001	658.44	<0.0001	10.32	<0.0001
		Week × trap location	51,387	5.17	<0.0001	43.73	<0.0001	10.77	<0.0001
Rivers	2001	Week	11,104	4.44	<0.0001	26.09	<0.0001	22.17	<0.0001
		Trap location	4,8	65.41	<0.0001	105.66	<0.0001	7.61	0.0078
		Week × trap location	43,104	4.46	<0.0001	25.69	<0.0001	2.61	<0.0001
Spring Creek	2001	Week	9,173	23.71	<0.0001	36.50	<0.0001	24.01	<0.0001
		Trap location	6,18	21.97	<0.0001	157.83	<0.0001	1.78	0.1594
		Week × trap location	49,173	22.53	<0.0001	40.04	<0.0001	1.56	0.0198

For this analysis, trap location included corn, edge, and increasing distances into the prairie. Corn is the adjacent commercial cornfield, edge is the interface between corn and prairie, and prairie is the remnant prairie.

Rivers site, the mean number of *D. u. howardi* per trap increased significantly with distance away from corn (Table 3); this trend is the reverse of that seen for *D. barberi* and *D. v. virgifera.* Similar numerical trends were observed at the Ratzlaff and Spring Creek sites, but there were no significant differences in mean densities among locations (Table 3). A different pattern was evident at the Brown site, where the mean trap collection at the edge location was significantly greater than the mean densities in the corn and prairie locations (Table 3). The mean density of *D. u. howardi* captured in edge and prairie habitats at the Rivers site was significantly greater than mean densities trapped in similar habitats at other sites (Table 3).

D. cristata, while locally abundant at some sites, was rarely collected in vial traps. Over 2001 and 2002, only 51 *D. cristata* were trapped (35 at the Rivers site). All *D. cristata* were trapped in edge and prairie habitats except for one individual trapped in corn at the Ratzlaff site in 2001. *D. cristata* was not trapped at the Brown site in either year.

Vial Trap Data: Temporal Variation in *Diabrotica* Abundance. Where *D. barberi*, *D. v. virgifera*, and *D. u. howardi* densities were great enough to allow statistical inferences to be made about temporal variation, the interaction of date by trap location was significant for most sites each year (Table 4). This indicates that beetle abundance was dynamic throughout the season. *D. cristata* densities were too low to be included in date by location analyses. Examples of temporal variation within sites that exhibited general trends observed for each species are presented below.

At the Rivers site in 2001, corn was the predominant habitat choice of *D. barberi*, with the highest mean density trapped from 20 to 27 July when corn was pollinating (Fig. 2). The mean number of *D. barberi* trapped in corn was significantly greater than mean densities collected in edge or prairie habitats, except for the 21–28 September and 29 September to 5 October sampling periods (Fig. 2). Mean beetle densities in edge and prairie habitats were not significantly different from mid-July to mid-August, but significantly more beetles were trapped in edge habitat than in the prairie habitat from 25 August to 21 September (Fig. 2).

Similar seasonal *D. barberi* habitat use patterns were observed at the Brown site in 2002 (Fig. 3). Corn was the primary habitat choice, with a low mean density of beetles trapped in the edge habitat for most of the season and the prairie relatively unused as habitat (Fig. 3). The mean density trapped in corn was significantly greater than the mean densities trapped in edge or prairie habitats between 9 July and 20 August 2002 (Fig. 3). Although the mean number of beetles trapped in the edge habitat was generally greater than mean densities trapped in the prairie, significant differences only occurred between 21 and 27 August 2002 (Fig. 3).

At the Brown site in 2002, *D. v. virgifera* was trapped almost entirely in corn, with few beetles captured in edge or prairie habitats (Fig. 4). *D. v. virgifera* densities rapidly increased to maximum levels and declined (Fig. 4); mean trap collections were significantly greater in corn than in other habitats between mid-July and early August (Fig. 4). After 27 August 2002, *D. v. virgifera* densities declined to very low levels (Fig. 4).

The Ratzlaff site was atypical compared with the other sites in both 2001 and 2002. Unlike the Brown
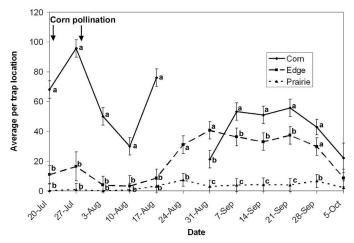
Fig. 2. Mean *D. barberi* per vial trap (±SE) at the Rivers site in 2001 by date and trap location. Trap location means within date with the same letter are not significantly different ($P > 0.05$). The lack of letters indicates no significant differences within date. Edge is the interface between corn and prairie; prairie is the remnant prairie; corn is the first-year commercial cornfield directly adjacent to the prairie. Each point represents mean (±SE) of beetles collected in vial traps during 1 wk. SEs reported were obtained from PROC MIXED lsmeans statement. Corn and edge means are based on three traps per location per date; prairie means are based on nine traps per date. Corn habitat was not sampled from 17 to 28 August. Date by location interaction was significant ($F = 10.85$; df, 21,132; $P < 0.0001$).

site, corn pollination generally occurred before the peak trap collections were recorded (Fig. 5; Table 5). In 2001, *D. v. virgifera* mean densities captured in corn were significantly greater than mean densities trapped in the other habitats only from 24 July through 7 August (Fig. 5). During the 7–14 August period, the mean density trapped in corn was not significantly different from the mean density trapped in the lowland prairie (Fig. 5). From 14 to 28 August 2001, mean trap collections in corn were again significantly greater than mean densities in the other habitats, and

mean trap collection in the lowland prairie was significantly greater than mean densities captured at other locations outside of corn (Fig. 5). After 4 September 2001 until the end of the trapping period, no trap location means were significantly different (Fig. 5).

In 2002 at the Ratzlaff site, mean trap collections in corn were significantly greater than mean trap collections in any other habitat, except in the weeks of 25 June to 2 July, 2–9 July, and 17–24 September (Table 5). Mean trap collections in the other habitats were
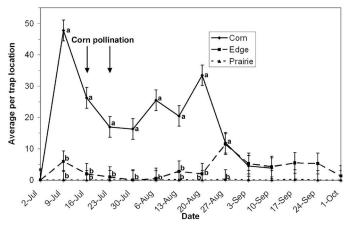

Fig. 3. Mean *D. barberi* per vial trap (±SE) at the Brown site in 2002 by date and trap location. Means followed by the same letter within date are not significantly different ($P > 0.05$). The lack of letters indicates no significant differences within date. Edge is the interface between corn and prairie; prairie is the remnant prairie; corn is the continuous commercial cornfield directly adjacent to the prairie. Each point represents the mean (±SE) of beetles collected in vial traps during 1 wk. SEs reported were obtained from PROC MIXED lsmeans statement. Corn and edge means are based on 4 traps per location per date; prairie means are based on 12 traps per date. Date by location interaction was significant ($F = 19.28$; df, 23,220; $P < 0.0001$)
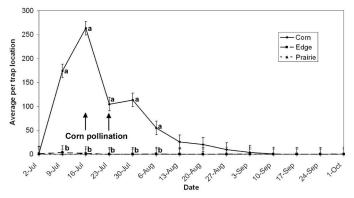
Fig. 4. Mean *D. v. virgifera* per vial trap (±SE) at the Brown site in 2002 by date and trap location. Means followed by the same letter within date are not significantly different (*P* > 0.05). Lack of letters indicates no significant differences within date. Edge is the interface between corn and prairie; prairie is the remnant prairie; corn is the continuous commercial cornfield directly adjacent to the prairie. Each point represents the mean (±SE) of beetles collected in vial traps during 1 wk. SEs reported were obtained from PROC MIXED lsmeans statement. Corn and edge means are based on 4 traps per location per date; prairie means are based on 12 traps per date. Date by location interaction was significant (*F* = 19.01; df, 23,220; *P* < 0.0001). Trapping in the corn was discontinued after 10 September.

highly variable over time (Table 5). In the 2 wk after corn pollination, mean trap collections were significantly greater in the edge habitat than in the prairie or lowland prairie habitats (Table 5). The mean trap collection in the lowland prairie was only significantly greater than mean densities trapped in edge and prairie locations in the week of 27 August to 3 September 2002 (Table 5). The mean trap collection in the edge habitat generally declined from 6 August to 3 September 2002, whereas mean densities increased in the lowland prairie habitat (Table 5). Mean trap collections in the upland prairie habitat remained relatively low throughout all trapping periods (Table 5).

In both years, at all study sites, *D. u. howardi* mean trap collections were initially low and then increased until the end of the study period (Fig. 6). At the Rivers site in 2001, mean trap collections in corn were generally low, and mean trap collections in the prairie were generally highest (Fig. 6). However, mean trap collections in the prairie habitat were only significantly greater than mean densities trapped in edge or corn habitats for 3 wk in September (Fig. 6). Early in the season, *D. u. howardi* was not trapped consistently in any specific location (Fig. 6).

Like *D. v. virgifera*, *D. u. howardi* mean trap collections at the Ratzlaff site in 2002 were strongly af-
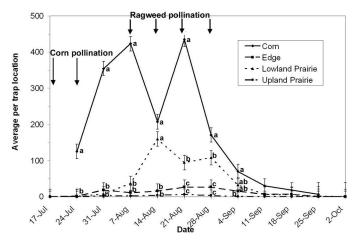

Fig. 5. Mean *D. v. virgifera* per vial trap (±SE) at the Ratzlaff site in 2001 by date and trap location. Means within date followed by the same letter are not significantly different (*P* > 0.05). Lack of letters indicates no significant differences within date. Edge is the interface between corn and prairie; prairie is the remnant prairie; corn is the continuous commercial cornfield directly adjacent to the prairie. Each point represents the mean (±SE) of beetles collected in vial traps during 1 wk. SEs reported were obtained from PROC MIXED lsmeans statement. Corn and edge means are based on 4 traps per location per date; prairie means are based on 12 traps per date. Date by location interaction was significant (*F* = 18.74; df, 31,167; *P* < 0.0001).

Table 5.   Mean *D. v. virgifera* per vial trap (±SE) at the Ratzlaff site in 2002 by date and trap location

Week	Corn	Edge	Upland Prairie	Lowland Prairie
25 June to 2 July	0.0 ± 12.9 a	0.0 ± 12.9 a	0.0 ± 9.0 a	0.0 ± 13.9 a
2–9 July	27.0 ± 12.9 a	0.0 ± 12.9 a	0.0 ± 9.0 a	0.0 ± 13.9 a
9–16 July[a]	223.4 ± 12.9 a	7.6 ± 12.9 b	1.2 ± 9.0 b	0.7 ± 13.9 b
16–23 July[a]	245.1 ± 12.9 a	15.4 ± 12.9 b	0.9 ± 9.0 b	0.3 ± 13.9 b
23–30 July	527.9 ± 12.9 a	63.1 ± 12.9 b	4.7 ± 9.0 c	3.7 ± 13.9 c
30 July to 6 Aug.	525.7 ± 12.9 a	62.1 ± 12.9 b	1.8 ± 9.0 c	3.5 ± 13.9 c
6–13 Aug.[b]	558.4 ± 12.9 a	51.3 ± 12.9 b	2.5 ± 9.0 c	18.5 ± 13.9 bc
13–20 Aug.[b]	332.0 ± 12.9 a	24.4 ± 12.9 b	2.7 ± 9.0 b	20.5 ± 13.9 b
20–27 Aug.[b]	368.9 ± 12.9 a	29.6 ± 12.9 b	1.4 ± 9.0 b	34.0 ± 13.9 b
27 Aug. to 3 Sept.[b]	214.6 ± 12.9 a	9.3 ± 12.9 c	2.8 ± 9.0 c	71.5 ± 13.9 b
3–10 Sept.	95.6 ± 12.9 a	4.6 ± 12.9 b	0.5 ± 9.0 b	15.0 ± 13.9 b
10–17 Sept.	92.7 ± 12.9 a	37.1 ± 12.9 b	5.2 ± 9.0 c	23.0 ± 13.9 bc
17–24 Sept.	24.6 ± 12.9 a	1.3 ± 12.9 a	0.6 ± 9.0 a	1.5 ± 13.9 a
24 Sept. to 1 Oct.[c]	NA	0.1 ± 12.9 a	0.0 ± 9.0 a	0.3 ± 13.9 a

Date by location interaction was significant ($F = 61.19$; df, 38,405; $P < 0.0001$). Means within rows (dates) followed by the same letter are not significantly different in a one-way analysis of variance ($P > 0.05$). SEs reported were obtained from PROC MIXED lsmeans statement. Edge is defined as the interface between corn and prairie; prairie is defined as remnant prairie. Corn and edge means are based on seven traps per location per date; upland prairie means are based on 15 traps per date; and lowland prairie means are based on six traps per date.

[a] Corn was pollinating.
[b] Giant ragweed (*A. trifida*) was pollinating in lowland prairie.
[c] Corn was not sampled over this week to allow for corn harvest.
NA, not applicable.

fected by the presence of the lowland prairie. Mean trap collections in corn were generally low and were either not significantly different or were significantly lower than mean densities trapped in the edge and prairie habitats (Fig. 7). Between 25 June and 13 August, there were no significant differences in mean densities at any trap location (Fig. 7). Between 20 August and 24 September, mean trap collections in the lowland prairie were significantly greater than mean trap collections in other habitats (Fig. 7). After 27 August, mean trap collections in the edge habitat were often significantly greater than mean densities in any habitat but the lowland prairie (Fig. 7).

Vial Trap Sex Ratios. Within sites, sample sizes of males and/or females were very uneven in one or more habitat locations seasonally or over some dates; therefore, it was not possible to statistically analyze male and female habitat preferences at each site. Total vial trap collections of *D. barberi*, *D. u. howardi*, and *D. v. virgifera* were predominantly male at all sites in both years (Table 6). In general, the proportion of *D. u. howardi* beetles captured in vial traps was less
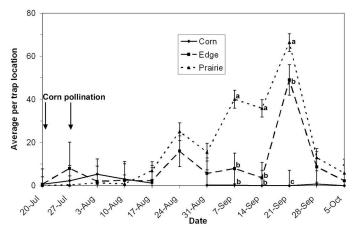

Fig. 6.   Mean *D. u. howardi* per vial trap (±SE) at the Rivers site in 2001 by date and trap location. Means within date followed by the same letter are not significantly different ($P > 0.05$). Lack of letters indicates no significant differences within date. Edge is the interface between corn and prairie; prairie is the remnant prairie; corn is the continuous commercial cornfield directly adjacent to the prairie. Each point represents the mean (±SE) of beetles collected in vial traps during 1 wk. SEs reported were obtained from PROC MIXED lsmeans statement. Corn and edge means are based on three traps per location per date; prairie means are based on nine traps per date. Corn habitat was not sampled from 17 to 28 August. Date by location interaction was significant ($F = 4.16$; df, 21,132; $P < 0.0001$).
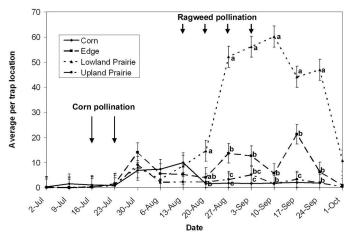
Fig. 7. Mean *D. u. howardi* per vial trap ($\pm$SE) at the Ratzlaff site in 2002 by date and trap location. Means within date followed by the same letter are not significantly different ($P > 0.05$). Lack of letters indicates no significant differences within date. Edge is the interface between corn and prairie; prairie is the remnant prairie; corn is the continuous commercial cornfield directly adjacent to the prairie. Each point represents the mean ($\pm$SE) of beetles collected in vial traps during 1 wk. SEs reported were obtained from PROC MIXED lsmeans statement. Corn and edge means are based on 7 traps per location per date; upland prairie means are based on 15 traps per date; and lowland prairie means are based on 6 traps per date. Date by location interaction was significant ($F = 15.05$; df, 38,405; $P < 0.0001$).

skewed toward males than *D. barberi* and *D. v. virgifera* collections. Although variable, data do indicate that both male and female *D. barberi*, *D. v. virgifera*, and *D. u. howardi* were trapped at all habitat locations at most sites during the 2-yr study.

Vial Versus Lure Trap Experiment 2003. Significantly more *D. v. virgifera* were collected per trap in the lure traps (mean = 34.3 ± 5.4) than in the vial traps (mean = 13.7 ± 5.3; $F = 7.32$; df, 1,47; $P = 0.0095$). The vial trap consistently collected a significantly higher percentage of *D. v. virgifera* males than did the lure trap (Table 7). The mean percentage of beetles that were male in the vial traps did not change significantly over the course of the study (Table 7), whereas the

mean percentage of males collected in the lure traps gradually decreased from late July to late August (Table 7). This trend contributed to the significant interaction between trap type and date (Table 7).

Discussion

Flowering Plants. Flowering plant abundances may have differed both between years and among sites because of differences in prairie management and/or environmental conditions. The type of prairie management (i.e., grazing, burning, haying) can impact the overall composition of plants present and the number of blooming forbs, which can then affect the insect

Table 6. Percentage of *Diabrotica* subsamples collected in vial traps that were male, by species, site, year, and trap location

Site	Year	Total sample	Subsample size by location			Percentage male		
			Corn	Edge	Prairie	Corn	Edge	Prairie
D. barberi								
Brown	2001	378	267	18	1	98.1	55.6	100.0
Brown	2002	422	315	102	5	99.4	56.9	0.0
Ratzlaff	2001	112	51	35	26	98.0	85.7	100.0
Ratzlaff	2002	69	55	8	6	94.6	50.0	83.3
Rivers	2001	1,486	661	330	157	99.1	90.0	93.0
D. undecimpunctata howardi								
Brown	2001	21	4	8	9	25.0	75.0	77.8
Brown	2002	556	31	212	313	83.9	84.4	85.6
Ratzlaff	2001	338	13	82	236	30.8	81.7	80.9
Ratzlaff	2002	1,834	168	318	952	54.2	80.5	86.9
Rivers	2001	750	26	69	616	73.1	88.4	79.7
D. v. virgifera								
Brown	2001	1,476	330	17	5	81.8	94.1	80.0
Brown	2002	1,670	472	11	1	91.3	100.0	100.0
Ratzlaff	2001	4,556	626	253	388	86.4	70.8	94.1
Ratzlaff	2002	16,318	1,852	771	713	86.5	91.7	98.3
Rivers	2001	211	182	18	5	96.7	55.6	80.0

Edge is defined as the interface between corn and prairie; prairie is defined as remnant prairie. Subsample size is the total no. of *Diabrotica* identified to sex from vial trap-collected beetles.

Table 7. Percentage of *D. v. virgifera* collected in vial and lure traps that were male (±SE) by week at the Ratzlaff site, 2003

	Vial trap		Lure trap	
	Total	Mean percentage male	Total	Mean percentage male
25 July to 1 Aug.	20	100.0 ± 10.0 A a	21	71.9 ± 7.0 A b
1–8 Aug.	71	100.0 ± 8.1 A a	77	38.9 ± 7.0 B b
8–15 Aug.	78	100.0 ± 8.1 A a	219	31.2 ± 6.3 BC b
15–22 Aug.	83	94.7 ± 6.3 A a	286	14.7 ± 6.3 C b
22–29 Aug.	102	90.0 ± 6.3 A a	148	12.5 ± 8.1 C b
29 Aug. to 5 Sept.	57	99.0 ± 6.3 A a	222	28.2 ± 7.0 BC b

Means within the same column followed by the same capital letter are not significantly different in a one-way analysis of variance ($P > 0.05$). Means within the same row followed by the same lowercase letter are not significantly different in a one-way analysis of variance ($P > 0.05$). SEs reported were obtained from PROC MIXED lsmeans statement. Total indicates no. of *D. v. virgifera* collected per trap type on each date. Trap type by date interaction was significant ($F = 2.77$; df, 5,36; $P = 0.0324$). Vial trap bait was powdered buffalo gourd root, which contained cucurbitacins E, I, and glycosides. Lure trap bait consisted predominantly of 4-methoxycinnamaldehyde and eugenol.

community (Stoner and Joern 2004). The Rivers and Ratzlaff prairies, which were never grazed or hayed, typically had the greatest abundances of blooming plant species and also had higher *Diabrotica* densities outside of corn than other sites (Tables 2 and 3). The substantial reduction in precipitation from fall 2001 through July 2002 (NCDC 2003) led to much drier conditions at most sites in 2002 than 2001, which also affected flowering plant abundances. Because *Diabrotica* beetles are typically pollen feeders, the abundances of flowering plants may have directly impacted the extent to which *Diabrotica* species used prairie habitat.

Diabrotica Use of Flowering Plants. Of the four species present in the corn–prairie system, *D. v. virgifera* and *D. barberi* beetles seemed to be the most selective when locating food hosts; both species were almost entirely found on plants from Poaceae and Asteraceae. *D. cristata* and *D. u. howardi* beetles exploited a broader range of plants, as they were found on Poaceae, Asteraceae, Fabaceae, and five other plant families. Despite these differences, all species seemed to favor Asteraceae outside of corn, but this may have been partially because plants from Asteraceae were the most abundant and provided the most available pollen sources. In the corn–prairie system studied, *Diabrotica* species were not found on all available blooming plants (Table 2). Therefore, it seems that there were definite host choices made, some of which were consistent at the genus level and some that were more restrictive at the species level.

D. v. virgifera and *D. barberi* were primarily found on corn plants, which is consistent with their use of corn as a primary larval and adult host (Krysan 1986, Krysan and Smith 1987). Over 2 yr, *D. v. virgifera* was found on the fewest number of noncorn plant species. Adults were only documented to feed on corn, *Ambrosia, Helianthus, Setaria, Solidago,* and *Vernonia; Setaria* was mostly present in cornfields. These observations are consistent with those of Ludwig and Hill (1975), who found large amounts of corn and small amounts of *Ambrosia* and *Amaranthus* pollen in *D. v. virgifera* guts. Naranjo (1994) also found in habitat preference experiments that *D. v. virgifera* would typically choose corn over flowering weeds. Moeser and

Vidal (2005) reported that *D. v. virgifera* in European cornfields most commonly fed on *Amaranthus/Chenopodium,* corn, *Ambrosia,* and *Helianthus* pollen. Observations from this study agree with the conclusions of Moeser and Vidal (2005) that *D. v. virgifera* adult host preferences are wider than previously described (Ludwig and Hill 1975, Naranjo 1994). In this study, *D. barberi* was generally less selective than *D. v. virgifera* in terms of adult host preference, but, like *D. v. virgifera,* primarily used plants from Asteraceae and Poaceae as food hosts. This is consistent with Cinereski and Chiang (1968), who found that *D. barberi* collected in corn fed on corn pollen but also frequently fed on pollen from Asteraceae and other Poaceae.

Overall Species Abundance. Because the prairie sites were chosen for their proximity to cornfields, *D. barberi* and *D. v. virgifera* had high abundances at the study sites. *D. barberi* abundance was highest at the Rivers site, which was the only site adjacent to first-year corn (Table 3). *D. v. virgifera* densities were greatest at sites with continuous corn and were lowest at the Rivers site (Table 3). Previous studies have established that *D. barberi* abundance is enhanced over time by crop rotation (Hill and Mayo 1980). In areas where *D. v. virgifera* and *D. barberi* coexist, *D. v. virgifera* apparently has an increased competitive advantage the longer fields are in continuous corn, which often leads to increases in *D. v. virgifera* densities and a reduction in *D. barberi* densities over time (Hill and Mayo 1980).

Diabrotica cristata, although closely tied to prairie habitat, was not equally abundant at all prairies. *D. cristata* was commonly observed at the Rivers site in 2001 and the Ratzlaff and Pearl Harbor sites in both years. However, *D. cristata* beetle densities were relatively low in the corn–prairie system compared with the other three species because only 51 beetles were trapped over 2 yr. *D. cristata* populations have been reported as patchy in distribution not only among prairies but also within prairies (Wiesenborn and Krysan 1980). This trend was also observed in this study because *D. cristata* were often locally abundant in one area of a prairie rather than evenly distributed throughout the prairie.

Diabrotica u. howardi abundance was greatest in the prairies and at the prairie–corn interfaces. As observed in this study, initial densities are typically low in Nebraska because this species apparently migrates from overwintering sites into Nebraska each season (Arant 1929, Hill and Mayo 1980, Krysan 1986). Population size probably peaked later in the season than the other species (Figs. 6 and 7), because *D. u. howardi* is multivoltine with an adult reproductive diapause (Krysan 1999).

Diabrotica **Seasonality.** For each *Diabrotica* species, the interaction of beetle density per habitat location by date was significant at most sites (Figs. 2–7; Tables 4 and 5). This suggests that habitats used by *Diabrotica* species varied over time. Habitat and food host preferences during the season seemed to be strongly dependent on the interaction of the relative phenologies of corn and other plants with the seasonality of each beetle species. *D. cristata* beetles were observed in the field before the other *Diabrotica* species in each year of the study (usually mid-late June). Because of this, *D. cristata* was able to use lead plant (*Amorpha canescens*), as a resource without much competition from other *Diabrotica* species. *D. barberi* and *D. v. virgifera* usually were present by early July, which often synchronized peak emergence and/or densities with corn pollination. Because *D. u. howardi* apparently recolonized the area each season, the temporal relationship with blooming plants was variable each year. Species in the *virgifera* group were generally more closely tied to their primary habitat, whether corn (*D. barberi* and *D. v. virgifera*) or prairie (*D. cristata*), whereas the only *fucata* species, *D. u. howardi*, was found in various habitats. The three species in the *virgifera* group seemed to use blooming plants in the preferred habitat first, but would move into secondary habitat to feed on blooming plants if the relative quality of feeding sites declined in the preferred habitat. *D. cristata* was rarely observed in corn but, when present, it usually coincided with corn pollination. Similarly, most of the *D. barberi* and *D. v. virgifera* were trapped in noncorn habitats after corn pollinated (Figs. 2–5; Table 5). During both years at the Ratzlaff site, giant ragweed, *Ambrosia trifida*, growing in the lowland prairie seemed to function as a large pollen sink that was very apparent and attractive to *D. v. virgifera* as corn became less attractive as a food host. *D. v. virgifera* densities increased in ragweed and declined in corn each year during the period this contrast in plant phenology occurred (Fig. 5; Table 5). It is interesting to note that when giant ragweed bloomed later in 2002 than in 2001, the response by *D. v. virgifera* to the ragweed pollen sink also occurred later in 2002 (Table 5; Fig. 5). While the Ratzlaff prairie was atypical in that it had a disturbed area bisecting the prairie, which supported growth of *A. trifida*, it does show that some weedy species that occur outside of cornfields (e.g., in roadsides, ditches, drainage areas) may serve as important feeding sites for *Diabrotica* pest species late in the season when quality food sources in cornfields are scarce.

Sampling. The sex ratio of beetles captured in vial traps was skewed toward males in all habitats throughout the season. Previous studies have shown that vial traps may be somewhat biased toward males for *D. barberi* (<1–34% female; Shaw et al. 1984) and *D. v. virgifera* (11–68% female; Shaw et al. 1984; 32.9% female in continuous cornfields; Levine and Gray 1994). In both studies, the bait source of cucurbitacin used in the traps was powdered squash fruit from a *Cucurbita andreana* L. × *C. maxima* L. cross, rather than the *C. foetidissima* root powder used in this study (Shaw et al. 1984, Levine and Gray 1994). It is not known if the source of cucurbitacin in this study or unique prairie habitat interaction with the traps led to the more extreme male sex bias.

The male sex bias associated with use of the vial trap in this study and previous studies (Shaw et al. 1984, Levine and Gray 1994), coupled with the significant difference in *D. v. virgifera* sex ratio between the vial trap and Trécé lure trap (Table 7), suggests that the vial trap does have an inherent bias toward males and probably underrepresents the proportion of females that are present. Because a major kairomone component of the lure trap (4-methoxycinnamaldehyde) is especially attractive to female *D. v. virgifera* (Metcalf and Lampman 1989, Lance 1993), it is possible that the lure trap was biased toward females and that neither trap provided a true picture of the actual sex ratio that was present outside of corn. However, it is important to note that data from both traps collectively indicate that both sexes were active in the Ratzlaff prairie habitat, especially after corn pollination, when the giant ragweed was in bloom.

It has been commonly reported in the literature that *D. v. virgifera* and *D. barberi* that move outside of corn are predominantly female (Naranjo and Sawyer 1988, Lance et al. 1989). Additionally, most research has focused on female behavior because of the importance of egg development and oviposition to population dynamics and pest management. This study strongly suggests that male *Diabrotica* are also moving out of corn into noncorn habitats during the season. This agrees with recent work by Moeser and Vidal (2005), which suggested that *D. v. virgifera* males are highly motile. Males may be present in noncorn habitats to increase fitness; like females, males may increase longevity and reproductive fitness by feeding in noncorn habitats when corn is no longer an attractive food source. Males may also be searching for unmated females outside of corn. More research is needed to understand the basis and extent of male movement outside of corn.

Remnant Prairies as a Resource for *Diabrotica*. Prairie fragmentation represents a major loss of habitat for *D. cristata* and other nonpest *Diabrotica* prairie species. Although *D. cristata* can complete larval development on corn in the laboratory (Yaro and Krysan 1986, Krysan and Smith 1987), it has only rarely been observed in corn when corn is pollinating or has green silks, suggesting that *D. cristata* behaviorally is limited to prairie habitat. Therefore, *D. cristata* could be much more susceptible than the other three *Diabrotica* spe-

cies to local extinctions from prairie management (Stoner and Joern 2004).

Despite the number of pest *Diabrotica* trapped outside of corn, prairies most likely only serve as a sink habitat, one where annual mortality exceeds annual recruitment, for *D. barberi* and *D. v. virgifera* (Pulliam and Danielson 1991). Although *D. barberi* beetles have been shown to move in and out of corn (Lance et al. 1989, Naranjo 1991) and feed on pollen from various plants (Cinereski and Chiang 1968), oviposition occurs primarily in corn (Boetel et al. 1992). *D. v. virgifera* in most of its geographical range has also been shown to oviposit primarily in corn (Hill and Mayo 1980, Siegfried and Mullin 1990). Preliminary data from soil samples collected in corn and prairie locations at the Ratzlaff site indicate that all *D. v. virgifera* and *D. barberi* eggs were recovered from corn (L.A.C., unpublished data). This information collectively suggests that *D. v. virgifera* and *D. barberi* oviposition in prairie habitat may be an uncommon event.

If *D. barberi* and *D. v. virgifera* oviposit in prairie habitats, a major remaining question is whether larvae can survive to adulthood on prairie grasses. Dominant grasses in prairies in eastern and central Nebraska include native species *Andropogon gerardii* (big bluestem), *Schizachyrium scoparium* (Michx.) Nash (little bluestem), *Sorghastrum nutans* (L.) Nash, (Indiangrass), *Panicum virgatum* L. (switchgrass), *Bouteloua curtipendula* (Michx.) Torr. (sideoats grama), *Koeleria macrantha* (Ledeb.) J.A. Schultes (june grass), *Elymus canadensis* L. (Canada wildrye), *Sporobolus compositus* (Poir.) Merr. (tall dropseed), and introduced species *Poa pratensis* L. (Kentucky bluegrass) and *Bromus inermis* (smooth brome) (Steinauer et al. 2003). *A. gerardii* is a suitable larval host for *D. cristata* (Yaro and Krysan 1986). Of these species, *A. gerardii* and *K. macrantha* have not been evaluated as potential *D. v. virgifera* or *D. barberi* larval hosts, but all of the other grass species present in this system can support at least some *D. v. virgifera* larval development (Clark and Hibbard 2004, Oyediran et al. 2004a, Wilson and Hibbard 2004). However, in terms of generating adult *D. v. virgifera*, these grasses seem to be poor hosts relative to corn (Clark and Hibbard 2004, Oyediran et al. 2004a, Wilson and Hibbard 2004). Of the major prairie grass species present in eastern and central Nebraska, only *Elymus canadensis* has been evaluated for its ability to support *D. barberi* larval development, and it seems to be a very poor host (Branson and Ortman 1971). Additionally, larval behavioral assay data suggest that *Bouteloua curtipendula* may not be a good host for *D. barberi* (Oyediran et al. 2004b). While *D. barberi* and *D. v. virgifera* may have the potential to complete larval development in prairie systems, previous research showed limited oviposition outside of corn in modern agroecosystems, suggesting that it may be unlikely that prairie habitats significantly contribute to pest *Diabrotica* population growth. More research is needed to determine the extent of pest *Diabrotica* oviposition in prairies. Whether or not *Diabrotica* larvae develop in prairies, adult *Diabrotica* feeding on pollen outside of corn may be beneficial and could potentially contribute to greater survival, longevity, and reproduction (Lance and Fisher 1987, Naranjo and Sawyer 1987, Elliott et al. 1990). *Diabrotica* that feed outside of corn but return to corn to lay eggs may be more able to contribute to population growth.

Because *D. u. howardi* most likely cannot overwinter in this region, Nebraska prairies would serve only as sink habitat for *D. u. howardi.* However, grasslands and noncrop areas are present in regions where *D. u. howardi* occurs throughout the year, but, because *D. u. howardi* has a broad larval and adult host range (Sell 1916, Krysan 1986) and is a pest of many crops (Arant 1929, Forbes 1996), use of grasslands as a resource by this species may not significantly affect long-term population growth.

The first *Bt* transgenic rootworm resistant corn hybrids were sold in the U.S. Corn Belt in 2003. When transgenic rootworm-resistant corn is used as a corn rootworm management tool, the U.S. Environmental Protection Agency mandates that an insect resistance management (IRM) plan be implemented to delay the development of resistance and to extend the use of *Bt* toxins as management tactics (EPA 2005). The registered product YieldGard Rootworm, (Monsanto Company) only has activity against *D. barberi* and *D. v. virgifera. D. v. virgifera* larvae exposed to YieldGard Rootworm corn emerge as adults at least 7–10 d later than *D. v. virgifera* fed on nontransgenic corn roots, and subsequent longevity and fecundity may be reduced (L.J.M., unpublished data). Reduced reproductive potential from sublethal exposure to the *Bt* toxin may be magnified if emergence occurs later in the season and beetles remain in corn after emergence. This is because the quality of corn as an adult diet deteriorates during grain fill, which can also reduce fecundity (Elliott et al. 1990). However, the fitness of beetles emerging from transgenic corn could possibly be increased by moving out of corn and using late season blooming plants as food hosts. This may increase longevity and fecundity in these individuals and could potentially affect the rate of evolution of resistance to the toxins present in the transgenic plants. Additional research is needed to evaluate how *Diabrotica* fitness is affected by feeding on noncorn pollen sources and the potential impact on resistance management.

Conclusions. Results from this study add to our understanding of four *Diabrotica* species and especially behavioral similarities and differences exhibited by each in the corn–remnant prairie system studied. Data support the conclusion that *D. v. virgifera, D. barberi*, and *D. u. howardi* primarily use the remnant prairie as sink habitat, where annual mortality probably exceeds annual recruitment. Habitat type and plant phenology, especially as it affects pollen availability, seem to be key factors that influence habitat choice by each species. The contrast in plant phenology between corn and noncorn areas especially influenced the level of noncorn habitat use by *D. v. virgifera* and *D. barberi*, key pests of corn in the United States. Use of crop and noncrop parts of the agroeco-

system by pest species suggests that a more holistic approach rather than a single field view may be appropriate when managing corn rootworms.

References Cited

Arant, F. S. 1929. Biology and control of the southern corn rootworm. Ala. Polytech. Inst. Agric. Exp. Sta. Bull. 230.

Boetel, M. A., D. D. Walgenbach, G. L. Hein, B. W. Fuller, and M. E. Gray. 1992. Oviposition site selection of the northern corn rootworm (Coleoptera: Chrysomelidae). J. Econ. Entomol. 85: 246–249.

Branson, T. F., and E. E. Ortman. 1971. Host range of larvae of the northern corn rootworm: further studies. J. Kans. Entomol. Soc. 44: 50–52.

Branson, T. F., and J. L. Krysan. 1981. Feeding and oviposition behavior and life cycle strategies of *Diabrotica*: an evolutionary view with implications for pest management. Environ. Entomol. 10: 826–831.

Cabrera Walsh, G. 2003. Host range and reproductive traits of *Diabrotica speciosa* (Germar) and *Diabrotica viridula* (F.) (Coleoptera: Chrysomelidae), two species of South American pest rootworms, with notes on other species of Diabroticina. Environ. Entomol. 32: 276–285.

Campbell, L. A. 2003. Seasonality and adult habitat use by four *Diabrotica* species at prairie-corn interfaces. MS thesis, University of Nebraska-Lincoln, Lincoln, NE.

Cinereski, J. E., and H. C. Chiang. 1968. The pattern of movements of adults of the northern corn rootworm inside and outside of corn fields. J. Econ. Entomol. 61: 1531–1536.

Clark, T. L., and B. E. Hibbard. 2004. Comparison of non-maize hosts to support western corn rootworm (Coleoptera: Chrysomelidae) larval biology. Environ. Entomol. 33: 681–689.

Clark, T. L., L. J. Meinke, and J. E. Foster. 2001. Molecular phylogeny of *Diabrotica* beetles (Coleoptera: Chrysomelidae) inferred from analysis of combined mitochondrial and nuclear DNA sequences. Insect Mol. Biol. 10: 303–314.

Coats, S. A., J. J. Tollefson, and J. A. Mutchmor. 1986. Study of migratory flight in the western corn rootworm (Coleoptera: Chrysomelidae). Environ. Entomol. 15: 1–6.

Darnell, S. J., L. J. Meinke, and L. J. Young. 2000. Influence of corn phenology on adult western corn rootworm (Coleoptera: Chrysomelidae) distribution. Environ. Entomol. 29: 587–595.

Eben, A., and A. Espinosa de los Monteros. 2004. Ideas on the systematics of the genus *Diabrotica* Wilcox and other related leaf beetles, pp. 59–73. *In* P. Jolivet, J. A. Santiago-Blay, and M. Schmitt (eds.), New developments in the biology of Chrysomelidae. SPB Academic Publishing, The Hague, The Netherlands.

Elliott, N. C., R. D. Gustin, and S. L. Hanson. 1990. Influence of adult diet on the reproductive biology and survival of the western corn rootworm, *Diabrotica virgifera virgifera.* Entomol. Exp. Appl. 56: 15–21.

Environmental Protection Agency [EPA]. 2005. Pesticides: regulating pesticides: plant incorporated protectants. http://www.epa.gov/pesticides/biopesticides/pips/ index.htm.

Forbes, S. A. 1996. Insect injuries to the seed and root of Indian corn. Univ. Ill. Agric. Exp. Stn. Bull. 44: 209–296.

Hill, R. E., and Z. B. Mayo. 1980. Distribution and abundance of corn rootworm species as influenced by topography and crop rotation in eastern Nebraska. Environ. Entomol. 9: 122–127.

Krysan, J. L. 1986. Introduction: biology, distribution, and identification of pest *Diabrotica*, pp. 1–23. *In* J. L. Krysan and T. A. Miller (eds.), Methods for the study of pest *Diabrotica.* Springer, New York.

Krysan, J. L. 1999. Selected topics in the biology of *Diabrotica*, pp. 479–513. *In* M. L. Cox (ed.), Advances in Chrysomelidae biology 1. Backhuys Publishers, Leiden, The Netherlands.

Krysan, J. L., and R. F. Smith. 1987. Systematics of the *virgifera* species group of *Diabrotica* (Coleoptera: Chrysomelidae: Galerucinae). Entomography 5: 375–484.

Lance, D. R. 1993. Effects of a nonpheromonal attractant on movement and distribution of adult *Diabrotica virgifera virgifera* (Coleoptera: Chrysomelidae). Environ. Entomol. 22: 654–662.

Lance, D. R., and J. R. Fisher. 1987. Food quality of various plant tissues for adults of the northern corn rootworm (Coleoptera: Chrysomelidae). J. Kans. Entomol. Soc. 60: 462–466.

Lance, D. R., N. C. Elliott, and G. L. Hein. 1989. Flight activity of *Diabrotica* spp. at the borders of cornfields and its relation to ovarian stage in *D. barberi.* Entomol. Exp. Appl. 50: 61–67.

Lawrence, W. S., and C. E. Bach. 1989. Chrysomelid beetle movements in relation to host-plant size and surrounding non-host vegetation. Ecology 70: 1679–1690.

Levine, E., and M. E. Gray. 1994. Use of cucurbitacin vial traps to predict corn rootworm (Coleoptera: Chrysomelidae) larval injury in a subsequent crop of corn. J. Entomol. Sci. 29: 590–600.

Littell, R. C., G. A. Milliken, W. W. Stroup, and R. D. Wolfinger. 1996. SAS® system for mixed models. SAS Institute, Cary, NC.

Ludwig, K. A., and R. E. Hill. 1975. Comparison of gut contents of adult western and northern corn rootworms in northeast Nebraska. Environ. Entomol. 4: 435–438.

Mabry, T. R., J. L. Spencer, E. Levine, and S. A. Isard. 2004. Western corn rootworm (Coleoptera: Chrysomelidae) behavior is affected by alternating diets of corn and soybean. Environ. Entomol. 33: 860–871.

McKone, M. J., K. K. McLauchlan, E. G. Lebrun, and A. C. McCall. 2001. An edge effect caused by adult corn-rootworm beetles on sunflowers in tallgrass prairie remnants. Conserv. Biol. 15: 1315–1324.

Metcalf, R. L., and R. L. Lampman. 1989. Estragole analogues as attractants for corn rootworms (Coleoptera: Chrysomelidae). J. Econ. Entomol. 82: 123–129.

Metcalf, R. L., and E. R. Metcalf. 1992. Plant kairomones in insect ecology and control. Chapman & Hall, New York.

Metcalf, R. L., R. L. Lampman, and P. A. Lewis. 1998. Comparative kairomonal chemical ecology of Diabroticite beetles (Coleoptera: Chrysomelidae: Galerucinae: Lu-

perini: Diabroticina) in a reconstituted tallgrass prairie ecosystem. J. Econ. Entomol. 91: 881–890.

Moeser, J., and S. Vidal. 2005. Nutritional resources used by the invasive maize pest *Diabrotica virgifera virgifera* in its new Southeast European distribution range. Entomol. Exp. Appl. 114: 55–63.

National Climatic Data Center, National Oceanic and Atmospheric Administration [NCDC]. 2003. www.ncdc. noaa.gov/oa/ncdc.html.

Naranjo, S. E. 1990. Comparative flight behavior of *Diabrotica virgifera virgifera* and *Diabrotica barberi* in the laboratory. Entomol. Exp. Appl. 55: 79–90.

Naranjo, S. E. 1991. Movement of corn rootworm beetles, Diabrotica spp. (Coleoptera: Chrysomelidae), at cornfield boundaries in relation to sex, reproductive status, and crop phenology. Environ. Entomol. 20: 230–240.

Naranjo, S. E. 1994. Flight orientation of *Diabrotica virgifera virgifera* and *D. barberi* (Coleoptera: Chrysomelidae) at habitat interfaces. Ann. Entomol. Soc. Am. 87: 383–394.

Naranjo, S. E., and A. J. Sawyer. 1987. Reproductive biology and survival of *Diabrotica barberi* (Coleoptera: Chrysomelidae): effect of temperature, food, and seasonal time of emergence. Ann. Entomol. Soc. Am. 80: 841–848.

Naranjo, S. E., and A. J. Sawyer. 1988. Impact of host plant phenology on the population dynamics and oviposition of northern corn rootworms, Diabrotica barberi (Coleoptera: Chrysomelidae), in field corn. Environ. Entomol. 17: 508–521.

Oyediran, I. O., B. E. Hibbard, and T. L. Clark. 2004a. Prairie grasses as hosts of the western corn rootworm (Coleoptera: Chrysomelidae). Environ. Entomol. 33: 740–747.

Oyediran, I. O., B. E. Hibbard, T. L. Clark, and B. W. French. 2004b. Selected grassy weeds as alternate hosts of northern corn rootworm (Coleoptera: Chrysomelidae). Environ. Entomol. 33: 1497–1504.

Prystupa, B., C. R. Ellis, and P.E.A. Teal. 1988. Attraction of adult *Diabrotica* (Coleoptera: Chrysomelidae) to corn silks and analysis of the host-finding response. J. Chem. Ecol. 14: 635–651.

Pulliam, H. R., and B. J. Danielson. 1991. Sources, sinks, and habitat selection: a landscape perspective on population dynamics. Am. Nat. 137: S50–S66.

Samson, F., and F. Knopf. 1994. Prairie conservation in North America. BioScience. 44: 418–421.

SAS Institute. 2001. User's manual, version 8.02. SAS Institute, Cary, NC.

Sell, R. A. 1916. Notes on the twelve-spotted cucumber beetle. J. Econ. Entomol. 9: 551–556.

Shaw, J. T., J. H. Paullus, and W. H. Luckmann. 1978. Corn rootworm oviposition in soybeans. J. Econ. Entomol. 71: 189–191.

Shaw, J. T., W. G. Ruesink, S. P. Briggs, and W. H. Luckmann. 1984. Monitoring populations of corn rootworm beetles (Coleoptera: Chrysomelidae) with a trap baited with cucurbitacins. J. Econ. Entomol. 77: 1495–1499.

Siegfried, B. D., and C. A. Mullin. 1990. Effects of alternative host plants on longevity, oviposition, and emergence of western and northern corn rootworms (Coleoptera: Chrysomelidae). Environ. Entomol. 19: 474–480.

Smith, R. F. 1966. Distributional patterns of selected western North American insects: The distribution of Diabroticites in western North America. Bull. Entomol. Soc. Am. 12: 108–110.

Steel, R.G.D., J. H. Torrie, and D. A. Dickey. 1997. Principles and procedures of statistics: a biometrical approach. WCB/McGraw-Hill, Boston, MA.

Steinauer, G., B. Whitney, K. Adams, M. Bullerman, and C. Helzer. 2003. A guide to prairie and wetland restoration in eastern Nebraska. Prairie Plains Resource Institute and Nebraska Game and Parks Commission. Aurora, NE.

Stoner, K.J.L., and A. Joern. 2004. Landscape vs. local habitat scale influences to insect communities from tallgrass prairie remnants. Ecol. Appl. 14: 1306–1320.

White, R. 1977. Sexual characters of species of *Diabrotica* (Coleoptera: Chrysomelidae). Ann. Entomol. Soc. Am. 70: 168.

Wiesenborn, W. D., and J. L. Krysan. 1980. A survey for *Diabrotica cristata* (Coleoptera: Chrysomelidae) on relict prairies of eastern South Dakota and southwestern Minnesota. Proc. S.D. Acad. Sci. 59: 130–137.

Wilcox, J. A. 1972. Coleopterorum Catalogus. Supplementa. (edita secunda). (Galerucinae, Luperini) Pars 78, Fasc. 2: 296–431.

Wilson, T. A., and B. E. Hibbard. 2004. Host suitability of nonmaize agroecosystem grasses for the western corn rootworm (Coleoptera: Chrysomelidae). Environ. Entomol. 33: 1102–1108.

Yaro, N., and J. L. Krysan. 1986. Host relationship of *Diabrotica cristata* (Coleoptera: Chrysomelidae). Entomol. News 97: 11–16.