2018

Seasonality of *Colaspis crinicornis* (Coleoptera: Chrysomelidae) and its Injury Potential to Corn in Southeastern Nebraska

Kentaro Miwa

University of Nebraska-Lincoln

Lance Meinke

University of Nebraska--Lincoln, lmeinke1@unl.edu

Follow this and additional works at: <https://digitalcommons.unl.edu/entomologyfacpub>

Part of the [Entomology Commons](#)

Miwa, Kentaro and Meinke, Lance, "Seasonality of *Colaspis crinicornis* (Coleoptera: Chrysomelidae) and its Injury Potential to Corn in Southeastern Nebraska" (2018). *Faculty Publications: Department of Entomology*. 653.

<https://digitalcommons.unl.edu/entomologyfacpub/653>

This Article is brought to you for free and open access by the Entomology, Department of at DigitalCommons@University of Nebraska - Lincoln. It has been accepted for inclusion in Faculty Publications: Department of Entomology by an authorized administrator of DigitalCommons@University of Nebraska - Lincoln.

Published in *Journal of Economic Entomology*, 111:1 (2018), pp 209–217.

doi 10.1093/jee/tox325

Copyright © 2017 Kentaro Miwa and Lance J. Meinke; published by Oxford University Press
on behalf of Entomological Society of America. Used by permission.

Submitted 25 April 2017; accepted 1 November 2017; published 18 December 2017.

Seasonality of *Colaspis crinicornis* (Coleoptera: Chrysomelidae) and its Injury Potential to Corn in Southeastern Nebraska

Kentaro Miwa and Lance J. Meinke

Department of Entomology, University of Nebraska–Lincoln, 109B Entomology Hall,
Lincoln, NE 68583

Corresponding author — L. J. Meinke, email lmeinke1@unl.edu

Abstract

Colaspis crinicornis Schaeffer (Coleoptera: Chrysomelidae) primarily occurs in the Great Plains, United States. Although *C. crinicornis* has historically been considered a non-pest and is rarely found in agricultural systems, population densities of this species have been increasing in corn, *Zea mays* L., and soybean, *Glycine max* (L.) Merrill, over the last decade in southeastern Nebraska. As part of a comprehensive project to understand the life history and pest potential of *C. crinicornis*, a field study was conducted to: understand adult seasonality of *C. crinicornis* using emergence cages and whole-plant-count sampling in cornfields and sweep-net sampling in soybean fields; confirm voltinism and the overwintering stage; and evaluate the potential of larvae to cause economic injury to corn roots. Data indicate that *C. crinicornis* is univoltine in southeastern Nebraska and overwinters as medium–large larvae at least 20 cm deep in the soil. Adults were present from June through August with peak emergence in July. The *C. crinicornis* lifecycle is similar to related *Colaspis* species. Root injury to corn was minor at population densities encountered in the field, and therefore, *C. crinicornis* is unlikely to cause economic loss. *C. crinicornis* may be an example of an insect species that has exploited open niches in crops that have been created by changes in agricultural and pest management practices. The lifecycle and

polyphagous nature of the insect, annual crop rotation, the shift to minimum tillage, and replacement of insecticides with *Bacillus thuringiensis* Berliner (Bt) traits may have collectively facilitated establishment and increased survival in agroecosystems.

Keywords: *Colaspis brunnea*, *Colaspis crinicornis*, grape colaspis

Colaspis F. (Coleoptera: Chrysomelidae) is the largest New World genus in the subfamily Eumolpinae, containing over 200 species with 28 species found in the United States (Riley et al. 2002). In spite of the species richness within the genus, little is known about the biology and ecology of most species. Exceptions are the small number of species known to be pests of agricultural crops (Lindsay 1943, Echols 1963, Rolston and Rouse 1965, Ostmark 1975, Balsbaugh 1982, Flynn and Reagan 1984, Oliver 1987, Lopez et al. 2002).

Colaspis crinicornis Schaeffer (Coleoptera: Chrysomelidae) primarily occurs in the Great Plains region of the United States (Blake 1974, Riley et al. 2003) and is not new to Nebraska as museum specimens indicate that it has been present in the state for at least a century (Miwa and Meinke 2015a). It has been collected only at relatively low densities in natural habitats (K. Miwa, personal observation) and has historically been considered a non-pest species that is rarely found in agricultural systems. However, population densities of this species have been increasing in corn, *Zea mays* L., and soybean, *Glycine max* (L.) Merrill, over the last decade in southeastern Nebraska and some parts of Iowa (Bradshaw et al. 2011; L. J. Meinke, personal observation).

C. crinicornis is often observed along with some of the other pest chrysomelid species in crops in southeastern Nebraska (L. J. Meinke, personal observation). In cornfields, *C. crinicornis* can be found with *Diabrotica virgifera virgifera* LeConte (Coleoptera: Chrysomelidae) and *Diabrotica barberi* Smith and Lawrence (Coleoptera: Chrysomelidae), which are two of the major insect pests of corn in the U.S. Corn Belt (Krysan 1986). *C. crinicornis* is also commonly found with a related pest species, *Colaspis brunnea* (F.) (Coleoptera: Chrysomelidae), in both corn and soybean fields (Miwa and Meinke 2015b) and *Ceratoma trifurcata* (Forster) (Coleoptera: Chrysomelidae) in soybean.

The increasing *C. crinicornis* population densities in crops led to grower inquiries about the species, and therefore, a proactive

approach was taken to initiate a comprehensive study to understand the life history and pest potential of *C. crinicornis*. Developmental biology, immature stage descriptions, and impact of diet on *C. crinicornis* life history traits were characterized in an initial study (Miwa and Meinke 2015a). *C. crinicornis* diel patterns of activity were described in a follow-up paper (Miwa and Meinke 2015b). This paper presents results of a field study that was conducted in southeastern Nebraska with the following objectives: characterize adult seasonality of *C. crinicornis* using emergence cages and whole-plant-count sampling in cornfields, and sweep-net sampling in soybean fields; confirm voltinism and the overwintering stage; and evaluate the potential of larvae to cause economic injury to corn roots.

Materials and Methods

Adult Seasonality

Adult seasonality of *C. crinicornis* was studied in commercial corn and soybean fields during the summer of 2010 and 2011 in Nemaha County, NE. Data were collected in three cornfields and three soybean fields in 2010, and four cornfields and three soybean fields in 2011 using the sampling procedures described in the following paragraphs. Agronomic practices and ecological conditions in these fields were typical of those found in southeastern Nebraska counties. Background information for each field is presented in **Table 1**. Meteorological data were collected at automated stations near the study sites. Average high and low temperatures during June–August ranged from 30.22–31.61°C to 18.61–20.50°C, respectively in 2010, and 29.11–32.22°C to 17.83–22.39°C, respectively in 2011 (see Miwa, 2014 for complete seasonal environmental data). A subset of beetles collected during this study was identified by E. G. Riley at Texas A&M University, and vouchers were deposited in the University of Nebraska State Museum.

Cornfields: Emergence Cages

Single plant emergence cages modified from Fisher (1980) were used to collect newly-emerged *C. crinicornis* as well as *C. brunnea*, *D. v. virgifera*, and *D. barberi* in cornfields. Each cage was 76 cm long ×

Table 1. Agronomic practices used in commercial corn and soybean fields sampled during 2010 and 2011 in Nemaha County, NE

Field	Previous crop	Tillage	Variety	Bt toxin types ^a	Insecticide seed treatment ^b	Seeds planted (per hectare)	Planting date	Irrigation
2010								
Cornfield 1	Corn	Light fall disking	DKC62-97	Cry1Ab, Cry3Bb1	Clothianidin	89,000	May 11	Yes
Cornfield 2	Soybean	None	DKC62-98	Cry1Ab	Clothianidin	89,000	May 9	Yes
Cornfield 3	Soybean	None	Pioneer 1625	Cry1F	Thiamethoxam	79,000	May 9	No
Soybean field 1	Corn	None	Asgrow 3731	N/A	None	370,000	May 15	No
Soybean field 2	Corn	None	Asgrow 3731	N/A	None	300,000	May 17	No
Soybean field 3	Corn	None	Asgrow 3731	N/A	None	370,000	May 19	Yes
2011								
Cornfield 4	Corn	Light fall disking	DKC62-97	Cry1Ab, Cry3Bb1	Clothianidin	89,000	April 28	Yes
Cornfield 5	Corn	Spring disking	Pioneer 1625	Cry1F	Thiamethoxam	79,000	May 3	No
Cornfield 6	Soybean	None	DKC62-98	Cry1Ab	Clothianidin	89,000	April 30	Yes
Cornfield 7	Soybean	None	Pioneer 1395	Cry1F, Cry34/35Ab1	Thiamethoxam	79,000	May 5	No
Soybean field 4	Corn	None	Asgrow 3432	N/A	None	370,000	May 12	Yes
Soybean field 5	Corn	None	Asgrow 3432	N/A	None	370,000	May 15	No
Soybean field 6	Corn	None	Asgrow 3731	N/A	None	370,000	May 13	Yes

a. Cry1Ab and Cry1F target Lepidoptera species; Cry3Bb1 and Cry34/35Ab1 target *Diabrotica* species.

b. Rate of all insecticide seed treatments was 250 µg/seed; all seeds had fungicide seed treatments.

38 cm wide, partially buried in the soil, and secured around the base of the plant stalk to prevent beetle escape but allow normal plant growth. Cages were in place from early to mid-June through late August or early September. In each field, 16 cages were placed at 30 to 40-m intervals within four rows (four cages per row), and the sampling rows were approximately 23 m apart. Beetles trapped in collecting jars placed on top of each cage were recovered every 3 to 4 d during peak *C. crinicornis* emergence periods and every 7 d after the frequency of emergence declined. The sex of *C. crinicornis* was determined using a dissecting microscope in the laboratory based on the characters described by Chapin (1979).

Cornfields: Whole-Plant-Count Sampling

To monitor the number of adult *C. crinicornis* and *C. brunnea* present on corn plants, whole-plant-count sampling was performed in each cornfield on days emergence cages were checked for beetles

in 2010 and 2011. Each field was divided into four sections, and 10 plants were randomly selected in each section. Beetles present on plants were visually counted, examining both sides of every leaf, the whorl, the tassel, ears, silks, and the stem. Sampling was carefully conducted to prevent beetles from dropping to the ground or flying away as they often exhibited these behaviors when disturbed (K. Miwa, personal observation). Data were collected by one sampler approximately at the same time of day (i.e., late morning or early afternoon) in each field to minimize sampling variation throughout the summer.

Soybean Fields: Sweep-Net Sampling

In soybean fields, sweep-net sampling was conducted to collect *C. crinicornis* and *C. brunnea* on the same days sampling was conducted in cornfields during 2010 and 2011. A 38-cm diameter sweep net with a 61-cm-long handle (BioQuip Products Inc., Rancho Dominguez, CA) was used. Each field was divided into four sections, and 20 consecutive sweeps were made in each of the four sections on each sampling day. Miwa and Meinke (2015b) describe the sweeping technique used in this study. Sampling was conducted by the same person in the late morning or early afternoon on each sampling day. Beetles collected were stored in plastic bags in a freezer and processed later in the laboratory.

Voltinism and Overwintering Stage

To determine whether or not *C. crinicornis* has multiple generations per year in southeastern Nebraska and to confirm the overwintering stage, an experiment was conducted in a cornfield at the University of Nebraska Eastern Nebraska Research and Extension Center (ENREC) in Saunders County in 2012. A corn hybrid (Dekalb DKC62-97) expressing *Bacillus thuringiensis* Berliner (Bt) toxins (Cry1Ab and Cry3Bb1) was planted in a field previously planted with corn using 76-cm row spacing and 30-cm within-row seed spacing. Seeds were coated with a neonicotinoid seed treatment (250 μ g clothianidin per seed). Shortly after planting, 10 plastic pots (31 cm in diam. $\times$ 36 cm high; Nursery Supplies Inc., Chambersburg, PA) were buried in the ground at 10-m intervals within a corn row, and each pot was filled with soil to approximately 2 cm from the top. To minimize the chance

of accidental introduction of any stages of *C. crinicornis* naturally present, soil placed into pots was taken from a dry pile free of plants at another location at ENREC that was fallowed the previous year. Two corn seeds (DKC62-97) were planted in each pot, and one plant was allowed to grow to maturity.

A single plant emergence cage described earlier was placed over each pot when plants were approximately 30 cm tall. Ten adult *C. crinicornis* (five males and five females) collected in the field were released in each cage on 15 June 2012. Adults were infested because eggs and neonate larvae of *C. crinicornis* were known to be fragile and mortality was high when handled (Miwa and Meinke 2015a). Corn leaves from new growth on plants in the same field were placed every 2 to 3 d inside each cage as adult food for 30 d after beetle release. Previous research indicated that leaf-feeding would support egg development in *C. crinicornis* (Miwa and Meinke 2015a). Foam was plugged into the hole that connected the cage and collecting jar to prevent released beetles from being captured. The foam plug was removed on 16 July 2012 after released beetles died. At the time of pot placement, garden cloth (Landscape Fabric, Scotts, Marysville, OH) was used to completely cover the soil surrounding the pot inside each cage. This directed released beetles to oviposit in soil within pots and prevent collection of any *C. crinicornis* adults that could emerge from soil surrounding the pots. Collecting jars were checked once or twice a week through October to determine if *C. crinicornis* adults would emerge in the same year under field conditions. To determine the overwintering stage of *C. crinicornis*, any individuals present in each pot were excavated on 6 December 2012 and 11 December 2012 by systematically removing small amounts of soil one depth layer at a time. The depth at which each individual was found was recorded. Morphological measurements of the larvae were taken, using an Olympus SZX16 stereomicroscope, Olympus DP26 camera, and Olympus cellSens v. 1.6 imaging software (Olympus, Tokyo, Japan).

Root Injury Evaluation

All plants that were caged in four Nemaha County cornfields during 2011 were excavated at the end of the adult *C. crinicornis* emergence period to evaluate root injury and to correlate root injury with

emerged adult density. In addition, part of the same cornfield that was used for the voltinism and overwintering stage study mentioned earlier was used to evaluate *C. crinicornis* root injury at ENREC in 2012. To record adult emergence, 30 single-plant emergence cages were used in 2012 in a similar manner as previously described for 2010 and 2011 studies in Nemaha County. Because no published method had been available to evaluate *Colaspis* larval injury in corn, the node-injury scale developed by Oleson et al. (2005) to quantify *Diabrotica* larval injury on corn roots was used for *C. crinicornis* during 2011 and 2012. In this root rating system, a root rating of 0.00 indicated no injury while 3.00, the highest possible value, indicated that three or more complete nodes were pruned to within 3.8 cm of the plant stalk (Oleson et al. 2005).

Statistical Analyses

Total Seasonal Counts From Emergence Cages, Whole-Plant-Count Sampling, and Sweep-Net Sampling

All statistical analyses were conducted using SAS software 9.2 (SAS Institute 2008, Cary, NC), and the .05 level of significance. Analysis of variance was conducted in a completely randomized design in PROC GLIMMIX for the total seasonal counts from emergence cages in corn, whole-plant-count sampling in corn, and sweep-net sampling in soybean. Data were log ($y+0.5$) transformed before each analysis to better meet the normality and variance assumptions of ANOVA. Non-transformed data are presented in the results. The total seasonal emergence of the four chrysomelid species captured in emergence cages in cornfields was compared among species within fields with 16 replications. The seasonal total *C. crinicornis* and *C. brunnea* from whole-plant-count sampling (four replications of 10 plants) and sweep-net sampling (four replications of 20 sweeps) were analyzed between species within fields. Mean separation was determined by LSMEANS comparisons in all analyses mentioned above.

C. crinicornis Male and Female Emergence Patterns

To compare the temporal emergence patterns between males and females of *C. crinicornis*, a nonlinear model adapted from Boetel et al. (2003) was fit to the cumulative emergence data for each of the two

fields with the highest total seasonal emergence (Cornfields 1 and 4). The following logistic curve was fit to the data using PROC NLMIXED: Cumulative emergence = $(a + a1*sex) / \{1 + \exp[(b+b1*sex) + (c + c1*sex)*days]\}$, where a is the final emergence count or the upper asymptote of the curve, and the inflection point or the maximum increase in cumulative emergence is found at $X = b/-c$ and $Y = a/2$. The parameters a , b , and c refer to males, and $a1$, $b1$, and $c1$ are the adjustments to a , b and c , respectively, for females. Exp in the model represents the natural log constant. In the logistic curve, linear emergence represents the period during which the majority of beetle emergence occurs, and the beginning and end of the linear emergence indicate the initiation and termination of beetle emergence, respectively. The quantities calculated and compared between males and females in PROC NLMIXED were days before linear emergence, days to the maximum linear emergence (peak emergence), days to the end of linear emergence, total days of linear beetle emergence, and total emergence count. These quantities were calculated from functions of a , b and c , and the specific functions may be found in Boetel et al. (2003).

Root Injury Ratings

To understand the relationship between the number of *C. crinicornis* and feeding injury on corn roots, correlation between root ratings and the number of adult *C. crinicornis* that emerged from each plant was analyzed in PROC CORR. Only the plants with < three total individuals from *C. brunnea*, *D. v. virgifera*, and *D. barberi* collections were included in the analyses to minimize the potential impact of feeding by the other species on root injury ratings.

Results

Adult Seasonality

Cornfields: Emergence Cages

In all cornfields sampled during 2010 and 2011 in Nemaha County, *C. crinicornis* was captured with single plant emergence cages (**Table 2**). *Colaspis brunnea* and *D. barberi* were recorded in six of the seven fields sampled, but only one field produced *D. v. virgifera* (Table 2). Time of emergence of all species collected overlapped with peak

Table 2. Mean total $\pm$ SEM adult *C. crinicornis*, *C. brunnea*, *D. v. virgifera*, and *D. barberi* that emerged per single plant emergence cage in cornfields during 2010 and 2011 in Nemaha County, NE

Field	<i>C. crinicornis</i>	<i>C. brunnea</i>	<i>D. v. virgifera</i>	<i>D. barberi</i>
2010				
Cornfield 1	15.86 $\pm$ 1.78a	0.13 $\pm$ 0.09b	0.25 $\pm$ 0.11b	0.13 $\pm$ 0.09b
Cornfield 2	2.56 $\pm$ 0.63a	0.25 $\pm$ 0.19b	0b	0.13 $\pm$ 0.09b
Cornfield 3	3.81 $\pm$ 1.00a	0.38 $\pm$ 0.15b	0b	5.00 $\pm$ 0.99a
2011				
Cornfield 4	14.06 $\pm$ 2.15a	0.69 $\pm$ 0.25b	0b	0.06 $\pm$ 0.06b
Cornfield 5	3.13 $\pm$ 0.69a	0b	0b	2.38 $\pm$ 0.46a
Cornfield 6	0.69 $\pm$ 0.20b	4.38 $\pm$ 1.41a	0b	0b
Cornfield 7	1.63 $\pm$ 0.59a	0.44 $\pm$ 0.22b	0b	0.48 $\pm$ 0.22b

Within years, means followed by the same lower-case letters within rows (among species within field) for each year are not significantly different ($P > 0.05$; LSMEANS test).

emergence of each species occurring in July. *C. crinicornis* densities were highly variable among fields, but it was the first or second most abundant species in every field (Table 2). *C. crinicornis* seasonal adult emergence per cage was highest in Cornfield 1 during 2010 (15.86 ± 1.78), and Cornfield 4 during 2011 (14.06 ± 2.15).

In 2010, initial *C. crinicornis* beetles were captured in emergence cages during late June in all cornfields with the first beetles recorded on 24 June in Cornfield 1 (Fig. 1a). Emergence was generally highest in July and declined in early August with the last beetle collected on 10 August in Cornfield 3 (Fig. 1a). In 2011, a similar pattern was

Fig. 1. Seasonal adult *C. crinicornis* emergence patterns derived from single plant emergence cage collections in cornfields during (a) 2010 and (b) 2011 in Nemaha County, NE.

observed with initial and final adult emergence recorded on 21 June (Cornfield 6) and 15 August (Cornfields 4 and 5), respectively (Fig. 1b).

In Cornfield 1 and Cornfield 4, total female emergence was significantly greater than that of males, consisting of 60.5% and 58.5% of total emergence, respectively (**Table 3**). However, no significant difference was found between males and females in days before linear emergence, days to the maximum linear emergence, days to the end of linear emergence, or total days of linear emergence (Table 3; **Fig. 2a** and **b**).

Table 3. Variables compared between males and females derived from nonlinear regression analysis of *C. crinicornis* cumulative emergence in Cornfield 1(2010) and Cornfield 4 (2011) in Nemaha County, NE

Curve character	Male	Female	P
Cornfield 1			
Days before linear emergence	7.58 $\pm$ 0.54	8.77 $\pm$ 0.32	0.0685
Days to maximum linear emergence	17.85 $\pm$ 0.27	18.12 $\pm$ 0.16	0.3970
Days to end of linear emergence	28.13 $\pm$ 0.64	27.47 $\pm$ 0.36	0.3814
Days of linear emergence	20.56 $\pm$ 1.06	18.70 $\pm$ 0.60	0.1408
Total emergence count	100.55 $\pm$ 0.94	153.79 $\pm$ 0.89	< 0.0001
Cornfield 4			
Days before linear emergence	10.48 $\pm$ 1.09	12.58 $\pm$ 0.74	0.1259
Days to maximum linear emergence	24.96 $\pm$ 0.69	26.19 $\pm$ 0.47	0.1539
Days to end of linear emergence	39.44 $\pm$ 1.43	39.79 $\pm$ 0.97	0.8396
Days of linear emergence	28.96 $\pm$ 2.14	27.21 $\pm$ 1.45	0.5064
Total emergence count	95.90 $\pm$ 2.14	135.39 $\pm$ 2.14	< 0.0001

The last date before emergence began was set as the starting point for analyses.

Fig. 2. Best fit cumulative emergence curves of male and female *C. crinicornis* derived from nonlinear regression analysis: (a) Cornfield 1 during 2010 and (b) Cornfield 4 during 2011 in Nemaha County, NE.

Cornfields: Whole-Plant-Count Sampling

Adult *C. crinicornis* were recorded at all sites while *C. brunnea* was found in six of the seven fields during 2010–2011 (**Table 4**). However, the number of *C. brunnea* recorded during whole-plant-count sampling was small, and the majority of *Colaspis* beetles found on corn plants were *C. crinicornis* (96.7% in 2010 and 83.2% in 2011). Even though *C. crinicornis* densities were variable among sites, total seasonal counts of *C. crinicornis* were significantly higher than those of *C. brunnea* in six of the seven fields (Table 4). During sampling each year, most adults of each species were found in the whorl, at the base of a leaf, or in silks especially on warm, sunny days. Adults were more active on cooler, cloudy days.

In 2010, *C. crinicornis* was initially observed on corn plants in mid-to late June with the first individual found on 18 June in Cornfield 1 (**Fig. 3a**). Beetle densities were highest in July with a peak of 14.0 ± 2.0 beetles per 10 plants in Cornfield 1 on 2 July (Fig. 3a) and then declined through late July into early August (Fig. 3a). In 2011, a similar pattern was observed with initial and last beetles observed in the field on 21 June (Cornfield 6) and 8 August (Cornfield 7), respectively (Fig. 3b). The highest beetle density per 10 plants recorded in 2011 was 8.0 ± 1.5 on July 1 in Cornfield 4 (Fig. 3b).

Table 4. Mean total $\pm$ SEM *C. crinicornis* and *C. brunnea* from whole-plant-count sampling in cornfields during 2010 and 2011 in Nemaha County, NE

Field	<i>C. crinicornis</i>	<i>C. brunnea</i>
2010		
Cornfield 1	$57.25 \pm 4.59a$	$1.00 \pm 1.00b$
Cornfield 2	$8.5 \pm 1.19a$	$1.75 \pm 0.85b$
Cornfield 3	$14.25 \pm 1.80a$	0b
2011		
Cornfield 4	$45.50 \pm 4.17a$	$4.75 \pm 2.59b$
Cornfield 5	$31.75 \pm 2.14a$	$4.75 \pm 0.63b$
Cornfield 6	$14.00 \pm 2.80a$	$10.00 \pm 1.29a$
Cornfield 7	$15.25 \pm 3.20a$	$1.50 \pm 0.65b$

Within years, means followed by the same lower-case letters within rows (among species within field) for each year are not significantly different ($P > 0.05$: LSMEANS test).

Fig. 3. Adult *C. crinicornis* densities observed over time during whole-plant-count sampling in cornfields during (a) 2010 and (b) 2011 in Nemaha County, NE.

Soybean Fields: Sweep-Net Sampling

In all soybean fields in 2010 and 2011, both *C. crinicornis* and *C. brunnea* were collected during sweep-net sampling. *C. crinicornis* consisted of 52.7 % and 85.9% of all *Colaspis* collected in 2010 and 2011, respectively. In 2010, total seasonal captures of both *C. crinicornis* and *C. brunnea* were low with *C. crinicornis* as common as *C. brunnea* at every site (**Table 5**). In 2011, mean total captures of *C. crinicornis* were variable among fields, and two of three fields had significantly greater mean total captures of *C. crinicornis* than *C. brunnea* (Table 5).

Table 5. Mean total $\pm$ SEM *C. crinicornis* and *C. brunnea* from sweep-net sampling in soybean fields during 2010 and 2011 in Nemaha County, NE

Field	<i>C. crinicornis</i>	<i>C. brunnea</i>
2010		
Soybean field 1	3.75 $\pm$ 1.11a	1.50 $\pm$ 0.29a
Soybean field 2	2.50 $\pm$ 0.29a	1.25 $\pm$ 0.25b
Soybean field 3	6.00 $\pm$ 1.87a	3.50 $\pm$ 0.29a
2011		
Soybean field 4	7.00 $\pm$ 1.29a	4.50 $\pm$ 1.32a
Soybean field 5	29.75 $\pm$ 3.12a	2.25 $\pm$ 1.31b
Soybean field 6	27.25 $\pm$ 5.98a	3.75 $\pm$ 0.48b

Within years, means followed by the same lower-case letters within rows (between species within field) for each year are not significantly different ($P > 0.05$: LSMEANS test).

Fig. 4. Adult *C. crinicornis* densities observed over time during sweep-net sampling in soybean fields during (a) 2010 and (b) 2011 in Nemaha County, NE.

In 2010, sweep-net captures of *C. crinicornis* began in late June with the first collections made on 24 June in Soybean fields 1 and 2 and the last on 27 July in Soybean fields 1 and 3 (**Fig. 4a**). No single peak capture period was observed in 2010, but most beetles were collected in July. Densities remained low throughout the summer with the highest capture of 1.25 ± 0.48 *C. crinicornis* per 20 sweeps recorded 9 July in Soybean field 2 (Fig. 4a). In 2011, the initial captures were recorded on 28 June in all soybean fields, and the last collection was made on 2 August in Soybean field 5 (Fig. 4b). Unlike 2010, peak captures of *C. crinicornis* were recorded on 5 July in Soybean field 5 (9.75 ± 1.44 beetles per 20 sweeps) and on 28 July in Soybean field 6 (11.00 ± 3.54 beetles per 20 sweeps) (Fig. 4b).

Voltinism and Overwintering

In 2012, no *C. crinicornis* adults of a new generation were captured in emergence cages at ENREC after field-collected adults were released and allowed to oviposit in cages placed over corn. When buried pots were systematically searched in December of 2012, no *C. crinicornis* adults or pupae were found in the soil although a total of 16 larvae were found in 5 of the 10 pots. All larvae were in quiescence in ovoid shaped-cells that were slightly larger than larvae and constructed of soil. The mean depth in the soil at which larvae were observed was 25.00 ± 0.57 cm (range: 20–29 cm, $n = 16$). The mean head capsule width and body length of the larvae were 0.98 ± 0.04 (range: 0.69–1.21 mm, $n = 16$), and 4.51 ± 0.22 mm (range: 3.39–6.20 mm, $n = 16$), respectively.

Root Injury Evaluation

In 2011, when plants under emergence cages that produced primarily *C. crinicornis* adults were evaluated for root injury, the mean root rating was 0.040 ± 0.005 (range: 0–0.1, $n = 36$) while the mean total number of *C. crinicornis* emerged per cage was 7.44 ± 1.34 (range: 1–36, $n = 36$). In 2012, the mean root rating was 0.031 ± 0.005 (range: 0–0.1, $n = 28$), and the mean total adult emergence was 9.32 ± 1.36 per cage (range: 2–34). A significant correlation was not found between the number of adult *C. crinicornis* captured in emergence cages and root injury ratings in either 2011 ($r^2 = 0.16785$; $P = 0.3286$) or 2012 ($r^2 = -0.08369$; $P = 0.6720$).

Discussion

Data from this study collectively support the conclusion that *C. crinicornis* is univoltine in southeastern Nebraska and overwinters in the soil as larvae. The emergence cage data indicated that adult emergence from the soil in cornfields began in late June and continued through early August with highest emergence in July. Whole-plant-count and sweep-net sampling confirmed that some adults of *C. crinicornis* forage in corn and soybean from late June through early August with highest densities in July. Adults have been shown to mate and oviposit soon after emergence, and newly-hatched larvae begin feeding on roots after a short egg period (Miwa and Meinke 2015a). The voltinism and overwintering stage experiment revealed that *C. crinicornis* overwinters as medium- to large-sized larvae at least 20 cm below the soil surface. These results were reinforced by casual sampling. Overwintering larvae were found on roots of volunteer and planted corn within 15 cm of the soil surface from April through June while pupae were found in the top 5 cm of soil in June and July (K. Miwa, personal observation). A conceptual model that presents the probable life history of *C. crinicornis* in southern Nebraska is shown in **Fig. 5**.

The general life history of *C. crinicornis* in southeastern Nebraska appears to be similar to known life histories of related *Colaspis* species (Lindsay 1943, Echols 1963). Both *C. brunnea* and *Colaspis pini* Barber (Coleoptera: Chrysomelidae) have been reported to be univoltine and overwinter as larvae in the soil. Bigger (1928) and Rolston and

Fig. 5. Probable life history of *C. crinicornis* in southeastern Nebraska.

Rouse (1965) also demonstrated that *C. brunnea* larvae moved downward in the soil to overwinter. Even though population densities of *C. brunnea* were generally low during this study, adult emergence of *C. crinicornis* and *C. brunnea* overlapped, indicating that temporal occurrence of these species is similar in Nebraska. Furthermore, Rolston and Rouse (1965) reported that a partial second generation of *C. brunnea* could occur in Arkansas under favorable conditions. Because the geographical distribution of *C. crinicornis* expands to southern Texas and Mexico (Riley et al. 2003), it is possible that the temporal occurrence of *C. crinicornis* life history events may be different in regions farther south of Nebraska.

Emergence data collected in cornfields and nonlinear regression analyses indicated that males and females of *C. crinicornis* have similar adult emergence patterns. Peak emergence occurred approximately at the same time for males and females. Because no significant differences between the sexes were found in days before linear emergence (although a near significant trend was evident in Cornfield 1 in 2010), days to the end of linear emergence, and total days of linear beetle emergence; it appears that protandry does not play a large role in the reproductive biology of this species. This is in contrast to *D. v. virgifera* in which male emergence begins before female emergence, allowing many males to become sexually mature before peak densities of females emerge (Quiring and Timmins 1990, Darnell et al. 2000, Nowatzki et al. 2002). In addition, more *C. crinicornis* females than males were captured in emergence cages, and this agrees with a report by Chapin (1979) that females were more common than males when collections of several *Colaspis* species were made in Louisiana.

Seasonal emergence and whole-plant-count data in cornfields followed a similar pattern in 2010 (Figs. 1a and 3a) but less so in 2011 (Figs. 1b and 3b). The reasons for this are unclear, but environmental factors may have played a role. Planting dates were 1 to 2 wk later in 2010 than 2011, but corn phenology was slightly advanced in 2010 until mid-July (Miwa 2014). Higher rainfall during May and June in 2010 (252.5 cm) versus 2011 (135.1 cm) may have contributed to this trend (Miwa 2014). In both years, beetle densities as measured by whole plant counts greatly declined during late July when corn reached the R2–R3 growth stage (Ritchie et al. 1993, Miwa 2014). Continued adult emergence from corn coupled with the reduction in whole plant counts during late July (Figs. 1 and 3) suggests that adult *C. crinicornis* may have moved out of corn to forage on other plant species that were more attractive as feeding sites during this period. Adult *C. crinicornis* were found in vegetation adjacent to cornfields in August after beetles were no longer observed during whole-plant-count sampling. Common plants on which beetles were found included *Digitaria* species and *Setaria* species.

Sweep-net sampling was an effective collecting method for *C. crinicornis* in soybean fields early in the adult emergence period when plant height was relatively short and most of the habitat could be sampled. Sweep-net sampling has also been effective for collecting *C. brunnea* in short vegetation (Lindsay 1943, Rolston and Rouse 1965, Eaton 1978). However, in this study, the effectiveness of sweep-net sampling was limited as plants became taller during both years. During 2010, plant height was over 100 cm by early July and approximately 150 cm by the end of the sampling period. In 2011, soybean height approached 80 cm by mid-July and reached 120 cm by the end of the sampling period. When sweeping in tall vegetation, a smaller proportion of total habitat was sampled, which may have underestimated beetle densities that were actually present. This may have contributed to the relatively low number of beetles collected throughout the 2010 season (Table 5; Fig. 4a) and the dramatic decline in beetles captured after mid-July 2011 in Soybean fields 5 and 6 (Fig. 4b). In both years, it was common for tall soybean plants to lodge after heavy storms or strong wind, making it difficult for the sampler to use a sweep net. Therefore, changes in plant height over time and weather conditions may have led to inconsistency in results.

In Cornfields 1 and 4, total seasonal emergence of *C. crinicornis* was at least 20 times greater than that of the second most common chrysomelid species (Table 2). High densities of *C. crinicornis* have also been found in other parts of southeastern Nebraska (K. Miwa, personal observation), including the field used for root injury evaluation at ENREC in 2012. Because *C. crinicornis* has remained relatively uncommon in natural habitats, this suggests that something has changed over time to facilitate increased survival in an agricultural setting.

Changes in agronomic and pest management practices have occurred during the last two decades that may have contributed to increased presence of *C. crinicornis* in the agricultural system of southeast Nebraska. Historically in the study area, aerial applications of insecticides were frequently made to manage European corn borer, *Ostrinia nubilalis* (Hubner; Lepidoptera: Crambidae) and other lepidopteran pests in both continuous and first-year corn. Soil insecticides were applied at planting in continuous corn to manage *D. v. virgifera* and *D. barberi* larvae. Both foliar and soil-applied insecticides may have greatly reduced densities of nontarget adult and larval *C. crinicornis* in corn, respectively. These insecticide tactics have largely been replaced by transgenic corn hybrids that express Bt toxins (Hutchison et al. 2010, Wangila et al. 2015) and include neonicotinoid seed treatments (Andersch and Schwarz 2003, Elbert et al. 2008). In this study, the two fields with the highest *C. crinicornis* emergence (Cornfields 1 and 4) were planted to Bt corn expressing *Diabrotica*-active traits with neonicotinoid seed treatments (Tables 1 and 2), suggesting the toxins had minimal impact on *C. crinicornis* survival. Neonicotinoid seed treatments have also been reported to have little effect on emergence of the related species *C. brunnea*, (Kaeb 2006). Exposure of young *C. crinicornis* larvae to seed-applied neonicotinoids may be low during the first year of their life cycle because seed treatments are designed to target early to mid-season soil insects (Andersch and Schwarz 2003, Elbert et al. 2008) and *C. crinicornis* eggs hatch during mid- or late summer. Furthermore, because *C. crinicornis* larvae externally feed on surface tissues and hairs of secondary roots (Miwa and Meinke 2015a), many small larvae feeding on new growth of large root systems may stay out of soil zones where maximum exposure to insecticides could occur. During the second year, overwintering larvae are larger and exposure to insecticides may be minimal as substantial feeding may not be required by some larvae before pupation.

The shift of farmers away from conventional tillage to adoption of minimum or no-tillage practices has also probably contributed to increased densities of *C. crinicornis* in southeast Nebraska. Higher survival of *C. brunnea* has been reported in minimum or no-tillage environments than in conventional tillage (Tonhasca 1994) so a similar response to tillage would be expected with *C. crinicornis*. Minimum tillage practices were used at most field sites in this study (Table 1) so less soil disturbance would likely reduce mortality of *C. crinicornis* larvae during fall and spring.

The annual corn-soybean rotation that is common in southeast Nebraska may also favor *C. crinicornis* population growth over time as it can utilize both corn and soybean. Adult emergence was recorded from both continuous and first-year cornfields during this study (Tables 1 and 2), and emergence has been observed from continuous and first-year soybean fields (Miwa 2014), suggesting that *C. crinicornis* oviposits, develops, and survives in all of these rotation patterns. In addition, corn and soybean leaves have been shown to be equally adequate food sources for adult survival and reproduction in the laboratory (Miwa and Meinke 2015a).

Data from this study also suggest that interspecific larval competition in the root zone may affect *C. crinicornis* population densities. The highest *C. crinicornis* emergence was recorded in cornfields where emergence of the three pest species was low (Fields 1 and 4, Table 2). In contrast, *C. crinicornis* only existed at a moderate density when *D. barberi* was also found at similar densities (Fields 3 and 5, Table 2). The two *Colaspis* species did not coexist at high or moderate densities in any field, with one species always significantly more common than the other in all fields (Table 2). Crop rotation is generally more widely practiced in the study area than some parts of Nebraska, where continuous corn is the predominant cropping pattern. *D. v. virgifera* densities were low throughout the study area (Table 2) but can be very high in continuous corn (Meinke et al. 2009). Interspecific competition between *D. v. virgifera* and *C. crinicornis* may be one of the reasons that high densities of *C. crinicornis* have not been found in areas with high *Diabrotica* densities (K. Miwa, personal observation). The interaction between *Diabrotica* and *Colaspis* species is a complex issue influenced by numerous factors and warrants further study.

The root injury data demonstrated extremely low injury levels caused by *C. crinicornis* larvae, and even plants with total emergence over 30 beetles had ratings of 0.1 or lower. Root injury ratings below 0.25 are unlikely to cause economic loss even under high environmental stress conditions (Oleson et al. 2005). The low root injury ratings are consistent with known feeding behavior and life history of *C. crinicornis*. First-year external feeding on primarily root hairs of full-grown plants with well-developed root systems may enable plants to withstand injury caused by young larvae. Overwintering larvae only feed for a short period before pupation so injury potential is of short duration. In contrast, *D. v. virgifera* and *D. barberi* larvae can severely injure corn by tunneling and pruning roots (Palmer and Kommedahl 1969, Chiang 1973, Riedell and Kim 1990) thereby facilitating removal of much more root biomass than *C. crinicornis*. Nevertheless, larval feeding on roots by *C. brunnea*, occasionally causes above-ground symptoms such as stunting, wilting, and discoloration of corn seedlings especially when plants are grown in stressful environments (Lindsay 1943, Ratcliffe and Steffey 2001). As *C. brunnea* and *C. crinicornis* have similar life history and larval feeding behaviors, a combination of *C. crinicornis* larval feeding on roots and adverse environmental conditions could potentially lead to similar symptoms in young corn plants when *C. crinicornis* densities are high. However, even though adults have been observed to feed on corn leaves, corn silks, and soybean leaves both in the field and laboratory (Miwa and Meinke 2015a,b), injury in both crops appears to be minor. At the population densities observed to date, *C. crinicornis* does not appear to be an economic threat to either corn or soybean production. This is an example of an insect species that appears to have adapted to changing agroecosystems by exploiting open niches. As agricultural practices continue to change, *C. crinicornis* densities and range expansion may fluctuate. This work serves as a baseline reference point if *C. crinicornis* densities continue to increase and pest status is attained in the future.

Acknowledgments — We thank E. G. Riley at Texas A&M University for identifying specimens. We are also grateful for C. A. Francis, B. C. Ratcliffe, and B. D. Siegfried at the University of Nebraska-Lincoln for providing comments and suggestions on earlier versions of the manuscript. We would like to acknowledge the Nemaha County growers who allowed us to collect data in their fields.

References

- Andersch, W., and M. Schwarz. 2003. Clothianidin seed treatment (Poncho®) – the new technology for control of corn rootworms and secondary pests in US-corn production. *Pflanzenschutz-Nachrichten Bayer* 56: 147–172.
- Balsbaugh, E. D. Jr. 1982. *Colaspis chapalensis* Blake (Coleoptera: Chrysomelidae), a new pest of corn in Mexico. *J. Kansas Entomol. Soc.* 55: 577–580.
- Blake, D. H. 1974. The costate species of *Colaspis* in the United States (Coleoptera: Chrysomelidae). *Smithsonian Contribution to Zoology*. No. 181. Smithsonian Institution Press, Washington, DC.
- Bigger, J. H. 1928. Hibernation studies of *Colaspis brunnea* (Fab.). *J. Econ. Entomol.* 21: 268–273.
- Boetel, M. A., B. W. Fuller, and P. D. Evenson. 2003. Emergence of adult northern and western corn rootworms (Coleoptera: Chrysomelidae) following reduced soil insecticide applications. *J. Econ. Entomol.* 96: 714–729.
- Bradshaw, J. D., E.W. Hodgson, M. E. Rice, J. K. VanDyk, and D. Adams. 2011. Soybean insects guide, *Colaspis* beetles. Department of Entomology, Iowa State University. http://www.ent.iastate.edu/soybeaninsects/colaspis_beetles
- Chapin, J. B. 1979. A review of the Louisiana species of *Colaspis* (Coleoptera: Chrysomelidae). *Coleopt. Bull.* 33: 445–450.
- Chiang, H. C. 1973. Bionomics of the northern and western corn rootworms. *Annu. Rev. Entomol.* 18: 47–72.
- 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.
- Eaton, A. T. 1978. Studies on distribution patterns, ovipositional preference, and egg and larval survival of *Colaspis brunnea* (Fab.) in North Carolina Coastal Plain soybean fields. Ph.D. dissertation. North Carolina State University, Raleigh, NC.
- Echols, H. W. 1963. The biology, distribution, and insecticidal control of pine colaspis (*Colaspis pini* Barber) in Louisiana. M.S. thesis. Louisiana State University, Baton Rouge, LA.
- Elbert, A., M. Haas, B. Springer, W. Thielert, and R. Nauen. 2008. Applied aspects of neonicotinoid uses in crop protection. *Pest Manag. Sci.* 64: 1099–1105.
- Fisher, J. R. 1980. A modified emergence trap for quantitative adult corn rootworm studies (Coleoptera: Chrysomelidae). *J. Kansas Entomol. Soc.* 53: 363–366.
- Flynn, J. L. and T. E. Reagan. 1984. Pollination interference in seed corn from silk feeding by *Colaspis louisianae* Blake (Coleoptera: Chrysomelidae). *J. Econ. Entomol.* 77: 1185–1188.
- Hutchison, W. D., E. C. Burkness, P. D. Mitchell, R. D. Moon, T. W. Leslie, S. J. Fleischer, M. Abrahamson, K. L. Hamilton, K. L. Steffey, M. E. Gray, et al. 2010. Area-wide suppression of European corn borer with Bt maize reaps savings to non-Bt maize growers. *Science* 330: 222–225.
- Kaeb, B. C. 2006. Management of grape colaspis, *Colaspis brunnea* (Coleoptera: Chrysomelidae), in seed corn production. M.S. thesis. Iowa State University, Ames, IA.

- 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-Verlag, New York, NY.
- Lindsay, D. R. 1943. The biology and morphology of *Colaspis flavida* (Say). Ph.D. dissertation. Iowa State College, Ames, IA.
- Lopez, A. N., H. A. A. Castillo, D. Carmona, P. L. Manetti, E. Mondino, and A. M. Vincini. 2002. Biological aspects of *Colaspis bridarollii* (Bechyne) (Coleoptera: Chrysomelidae: Eumolpinae) in Argentina. *Coleopts. Bull.* 56: 259–269.
- Meinke, L. J., T. W. Sappington, D. W. Onstad, T. Guillemaud, N. J. Miller, J. Komaromi, N. Levay, L. Furlan, J. Kiss, and F. Toth. 2009. Western corn rootworm (*Diabrotica virgifera virgifera* LeConte) population dynamics. *J. Agric. and Forest Entomol.* 11: 29–46.
- Miwa, K. 2014. Natural history, phylogenetic relationships, and pest potential of *Colaspis* Fabricius Species (Coleoptera: Chrysomelidae) in Nebraska. Ph.D. Dissertation. University of Nebraska-Lincoln, Lincoln, NE.
- Miwa, K., and L. J. Meinke. 2015a. Developmental biology and effects of adult diet on consumption, longevity, and fecundity of *Colaspis crinicornis* (Coleoptera: Chrysomelidae). *J. Insect Sci.* 15: 78.
- Miwa, K., and L. J. Meinke. 2015b. Diel patterns of *Colaspis brunnea* and *Colaspis crinicornis* (Coleoptera: Chrysomelidae) in southeastern Nebraska. *Environ. Entomol.* 44: 1553–1561.
- Nowatzki, T. M., J. J. Tollefson, and D. D. Calvin. 2002. Development and validation of models for predicting the seasonal emergence of corn rootworm (Coleoptera: Chrysomelidae) beetles in Iowa. *Environ. Entomol.* 31: 864–873.
- Oleson, J. D., Y. Park, T. M. Nowatzki, and J. J. Tollefson. 2005. Node injury scale to evaluate root injury by corn rootworms (Coleoptera: Chrysomelidae). *J. Econ. Entomol.* 98: 1–8.
- Oliver, A. D. 1987. Seasonal history and pest status of *Colaspis pini* Barber (Coleoptera: Chrysomelidae) on Christmas tree pines in Louisiana. *J. Entomol. Sci.* 22: 295–296.
- Ostmark, H. E. 1975. Banana pests in the genus *Colaspis* including description of a new species (Coleoptera: Chrysomelidae). *Fla. Entomol.* 58: 1–8.
- Palmer, L. T., and T. Kommedahl. 1969. Root-infesting *Fusarium* species in relation to rootworm infestations in corn. *Phytopath.* 59:1613–1617.
- Quiring, D. T., and P. R. Timmins. 1990. Influence of reproductive ecology on feasibility of mass trapping *Diabrotica virgifera virgifera* (Coleoptera: Chrysomelidae). *J. Appl. Ecol.* 27: 965–982.
- Ratcliffe, S., and K. Steffey. 2001. Grape colaspis: a quick review, pp. 94–95. In K. Steffey (ed.), *The pest management and crop development bulletin*. University of Illinois Extension, Urbana, IL.
- Riedell, W. E., and A. Y. Kim. 1990. Anatomical characterization of western corn rootworm damage in adventitious roots of maize. *J. Iowa Acad. Sci.* 97: 15–17.
- Riley, E. G., S. M. Clark, R. W. Flowers, and A. J. Gilbert. 2002. Family 124. Chrysomelidae Latreille 1802, pp. 617–691. In R. H. Arnett Jr., M. C. Thomas, P. E. Skelley, and J. H. Frank (eds.), *American Beetles*, Vol. 2. CRC Press, Boca Raton, FL.

- Riley, E. G., S. M. Clark, and T. N. Seeno. 2003. Catalog of the leaf beetles of America north of Mexico (Coleoptera: Megalopodidae, Orsodacnidae and Chrysomelidae, exclusive of Bruchinae). Coleopterists Society, Sacramento, CA.
- Ritchie, S. W., J. J. Hanway, and G. O. Benson. 1993. How a corn plant develops. Special Report 48. Iowa State University, Cooperative Extension Service, Ames, IA.
- Rolston, L. H. and P. Rouse. 1965. The biology and ecology of the grape colaspis, *Colaspis flavida* Say, in relation to rice production in Arkansas Grand Prairie. Bulletin No. 694. University of Arkansas Division of Agriculture Cooperative Extension service, Fayetteville, AR.
- SAS Institute. 2008. SAS/STAT user's guide, release 9.2 ed. SAS Institute, Cary, NC.
- Tonhasca, A. 1994. Response of soybean herbivores to two agronomic practices increasing agroecosystem diversity. *Agr. Ecosyst. Environ.* 48: 57–65.
- Wangila, D. S., A. J. Gassmann, J. L. Petzold-Maxwell, B. W. French, and L. J. Meinke. 2015. Susceptibility of Nebraska western corn rootworm (Coleoptera: Chrysomelidae) populations to Bt corn events. *J. Econ. Entomol.* 108: 742–751.