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Butterfly and floral community dynamics at a native prairie agrofuel research site

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BUTTERFLY AND FLORAL COMMUNITY DYNAMICS AT A NATIVE PRAIRIE AGROFUEL RESEARCH SITE

An Abstract of a Thesis

Submitted

in Partial Fulfillment

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Benjamin J. Hoksch

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ABSTRACT

Agrofuel production in the Midwest has historically relied upon monoculture food crops (*Zea mays* and *Glycine max*) that do little to support biodiversity or maintain soil and water quality. The University of Northern Iowa's Tallgrass Prairie Center is investigating the feasibility of establishing and harvesting diverse mixes of native prairie vegetation for use as a sustainable agrofuel. In 2009 48 research plots were established on three soil types, on land with a >30 year history of row crop production in Black Hawk County, IA. Each plot was seeded with one of four native prairie agrofuel crops: (1) Switchgrass1, (2) Grasses5 (5 warm season grass species), (3) Prairie16 (16 species of grasses, legumes, and forbs), or (4) Prairie32 (32 species of grasses, legumes, forbs, and sedges). Research plots were actively managed with a three-year rotation (establishment/no management, burn, harvest), and in 2013 and 2014 a significant portion of the research site experienced early summer flooding. I monitored floral and butterfly communities present on site from 2010 through 2014 and hypothesized that more diverse floral communities would support more abundant and diverse butterfly communities. Butterflies were ~3.6 times more abundant, ~1.4 times more species rich, and more diverse in Prairie16 and Prairie32 than Switchgrass1 and Grasses5; however butterfly abundance, richness and diversity in Prairie16 and Prairie32 did not diverge as the site matured as predicted. Sown flowers were more species rich and diverse, but not more abundant in Prairie32 than Prairie16. Flooding frequency and duration was a strong predictor of sown floral abundance, richness and diversity; which in turn influenced butterfly abundance. My research suggests that the widespread adoption of diverse

assemblages of native prairie plants as agrofuel crops would provide higher quality habitat for butterflies than native, grass-only agrofuel feedstocks.

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This Study by: Benjamin J. Hoksch
Entitled: Butterfly and Floral Community Dynamics at a Native Prairie Agrofuel Research Site

has been approved as meeting the thesis requirement for the

Degree of Master of Science

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CHAPTER 1
INTRODUCTION

Escalating worldwide energy demand has stimulated much research aimed at developing renewable agrofuels. In the United States, corn ethanol and soy biodiesel currently dominate the transportation agrofuel market, but their energetic and environmental benefits have recently been under scrutiny (Hill 2009; Tilman et al. 2006). Alternative agrofuel crops such as native perennial prairie plantings have the potential not only to furnish a beneficial energy source, but also to promote improved habitat conditions for wildlife (Blank et al. 2014; Fargione et al. 2009; Myers et al. 2012; Robertson et al. 2011) and provide valuable ecosystem services (USDA 2011). This is especially true in Iowa's agricultural landscape, where less than 0.1% of native tallgrass prairie habitat remains (Samson and Knopf 1994).

In the Midwestern USA, more than 90% of native grasslands have been lost to the intensification of agriculture over the last 150 years (Samson and Knopf 1994). The conversion of small, diverse family farms to larger farms that produce corn (*Zea mays*) and soybean (*Glycine max*) monocultures (Jackson 2002) has resulted in the homogenization of the landscape and subsequent degradation of remaining tallgrass prairie remnants (Mutel 2007; Smith 1998). Significant declines in prairie butterfly populations have followed (Debinski and Kelly 1998; Pleasants and Oberhauser 2012; Swengel et al. 2011), including most recently the iconic monarch butterfly (*Danaus plexippus*). Today, agricultural expansion continues to threaten restored and remnant prairie. The future conservation of prairie butterflies and their associated plant communities is dependent on proper management of existing prairie (Schlicht and Orwig 1998; Swengel and Swengel 2001; Vogel et al. 2010) and the expansion of suitable habitat through prairie restoration (Davros et al. 2006; Schultz and Crone 2005; Shepherd and Debinski 2005).

The first prairie restoration was undertaken at the University of Wisconsin, Madison by Norman Fassett and Aldo Leopold in the early 1930's (Blewett and Cottam 1984). Since then, practitioners have learned much about re-establishing native vegetation yet still find it difficult to restore disturbed sites (Allison 2002; Howe 1994; Kindscher and Tieszen 1998). The low plant diversity found in restorations is often due to the unavailability or high cost of seed from native plants, failed establishment or disappearance of seeded species, competition with invasive species, altered disturbance regimes (Smith et al. 2010), and the slow re-colonization rate of native plants from the surrounding landscape (Foster et al. 2007). Butterflies respond to plant diversity due to their requirements for larval host plants and adult nectar sources (Ries et al. 2001), and consequently the diversity of butterflies found in restorations is often lower than that of remnants (Schlicht and Orwig 1998; Shepherd and Debinski, 2005). Therefore, targeted research involving the establishment of prairie communities is valuable in understanding the long-term conservation of biodiversity in the Midwest.

One potential way to increase the extent of quality grassland habitat in the Midwest is the large-scale establishment of perennial prairie polycultures as agrofuel feedstocks (Fargione et al. 2009; Hill et al. 2006; Myers et al. 2012). Agrofuel feedstocks can be derived from a variety of plant materials, and once harvested may be incinerated, converted to liquid fuel, or gasified to produce energy (USDA 2011). Traditional Midwest agrofuel systems rely on high input monoculture annual crops that do little to maintain biodiversity on the landscape. Targeted research on alternative prairie polycultures as a renewable energy source has indicated energetic and environmental benefits over corn ethanol (Hill et al. 2006; Tilman et al. 2006). Proposed prairie agrofuel crops have consisted of monocultures as well as diverse mixtures of native plant functional groups (Myers et al. 2012; Tilman et al. 2006). The adoption of prairie polycultures as agrofuel feedstocks could increase the amount of restored grassland on the landscape and provide quality habitat for prairie butterflies (Landis and Werling 2010; Myers et al. 2012).

To avoid competition with conventional food production on prime farmland, model prairie agrofuel systems have been recommended for establishment on marginal agricultural land (Hill et al. 2006; Tilman et al. 2006), including floodplains. However, the effects of periodic flooding on grasslands present management concerns for the maintenance of biodiversity. Flood events have been shown to decrease forb abundance in grassland mesocosms (Insausti et al. 1999) and could encourage the spread of native and non-native invasive species (Pysek and Prach 1993). Flooding directly affects the survival rate of immature life stages of butterflies (Joy and Pullin 1997; Nichols and Pullin 2003; Severns 2011) and could indirectly affect adult survival by decreasing the abundance, richness and diversity of larval host plants and/or adult nectar sources. Current climate change models predict an increase in flood events (Easterling et al.

2000), which could impact the trajectories of intact and restored plant and butterfly communities. Considering the cost and effort involved in establishing prairie, it is important to understand how prairie communities will respond to current and changing disturbance regimes so practitioners may most effectively reach their management goals.

In 2008, the University of Northern Iowa's Tallgrass Prairie Center began investigating the feasibility of cultivating and harvesting native perennial prairie vegetation for use as an agrofuel feedstock. In 2009 48 research plots were established on three soil types, on land with a >30 year history of row crop production in Black Hawk County, IA. Each plot was seeded with one of four native prairie agrofuel crops: (1) Switchgrass1, (2) Grasses5 (5 warm season grass species), (3) Prairie16 (16 species of grasses, legumes, and forbs), or (4) Prairie32 (32 species of grasses, legumes, forbs, and sedges).

I report here the results from a five-year study (2010-2014) monitoring the floral and butterfly communities present on site. Research plots were specifically managed for agrofuel production with a three-year rotation (establishment/no management, burn, harvest). In 2013 and 2014 a significant portion of the research site experienced early summer flooding and I took advantage of this "natural experiment" to address questions about flooding.

My specific research questions are:

1. How do annual butterfly and floral abundance, species richness, community diversity and community composition vary among agrofuel crops and soil?

2. Are there positive relationships between the total abundance and richness of forbs in bloom and the total abundance and richness of butterflies?
3. How do early summer floods affect floral abundance, richness, diversity, or community composition in forb-rich crops (Prairie16 and Prairie32)? Do butterflies decrease in abundance, richness or diversity in altered plant communities following flooding?

CHAPTER 2

METHODS

Site History

We conducted research at the Cedar River Natural Resource Area in southeastern Black Hawk County, Iowa, USA (42˚23' N and 92˚13´ W). The site lies within the Cedar River floodplain and consists of a mosaic of open fields surrounded by floodplain timber, wetlands and ponds (Fig. 1). The 1800's historic vegetation was described as floodplain prairie, bordered by timber to the east and south (ISU GISSRF, 2014). Portions of the site had a >50 year crop history prior to the 1970's, when many low lying areas were set aside and succession to timber occurred.

The site was acquired by the Black Hawk County Conservation Board in 1973 and tillable ground was leased to a neighboring farmer for corn/soybean production. In 2008 the lease was transferred to the University of Northern Iowa's Tallgrass Prairie Center, and a long-term study investigating the feasibility of utilizing native perennial prairie vegetation as an agrofuel feedstock was initiated.

Experimental Design

In 2009, seven fields (3.7-6.1 ha) were divided into 48 research plots (0.30-0.56 ha) and randomly assigned one of four crops: (1) Switchgrass1 (*Panicum virgatum* monoculture), (2) Grasses5 (5 warm season grass species), (3) Prairie16 (16 species of grasses, legumes, and forbs), or (4) Prairie32 (32 species of grasses, legumes, forbs, and

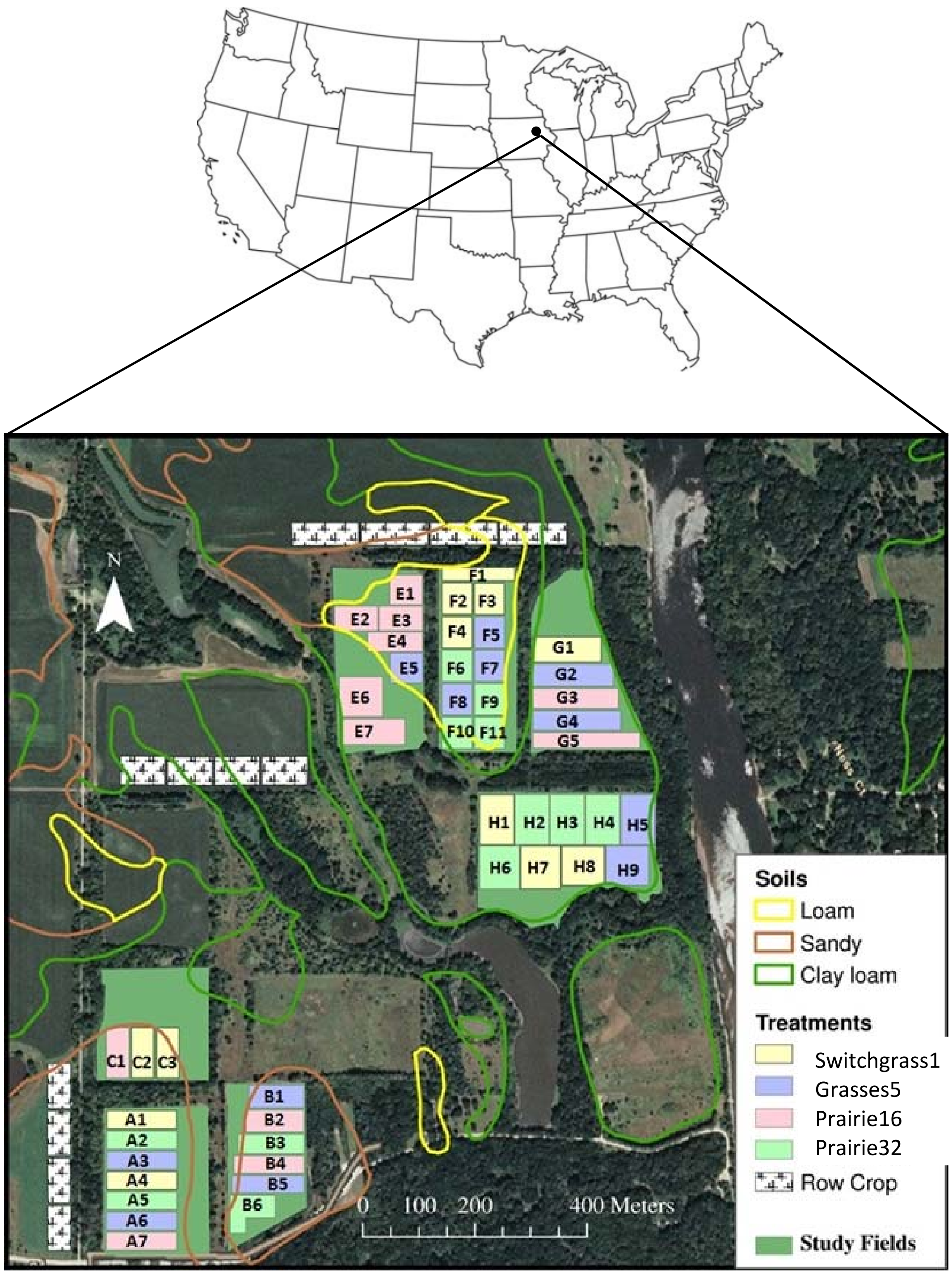


Figure 1. Map of study site at Cedar River Natural Resource Area in Black Hawk County, Iowa, USA (42˚23’ N and 92˚13´ W). Each treatment is replicated 4 times on each of three soil types. Map courtesy of Jim Mason.

sedges). Henceforth, I refer to the Switchgrass1 and Grasses5 crops collectively as "grass crops," and to the Prairie16 and Prairie32 plots collectively as "forb-rich crops." Each of the four crops were replicated four times on three soil types: (1) Flagler sandy loam (no flooding, drainage class (DC) = "somewhat excessively drained," corn suitability rating (CSR) = 50 (2) Waukee loam (no flooding, DC = "well-drained," CSR = 79), and (3) Spillville-Coland complex (occasional flooding, DC = somewhat poorly drained, CSR = 60) (USDA 2015). Henceforth, we refer to these soil types as sandy loam, loam, and clay loam respectively. Considering the site's below county average CSRs and location in the Cedar River floodplain we consider it to be marginal agricultural land.

Site Establishment and Management

To reduce weed competition and to ensure uniform site management, all fields were sprayed with glyphosate and planted in June of 2008 to Round-up® Ready soybeans. The site was sprayed again with glyphosate during the growing season and was harvested in October. At this time tillable ground outside the 48 research plots (non-plot area) was seeded to the Prairie32 crop with double the forb-seeding rate (Table A1, Fig. 1) In May of 2009, plots were seeded from lowest to highest diversity crop with a no-till native grass drill. A 3 m buffer of cool season vegetation (Dan Patch horse pasture mix, Des Moines Forage & Turf Seed Corp., Ankeny, Iowa) was seeded around each plot to limit the spread of plants between plots and aid in boundary perception during surveys. These lanes were mowed periodically throughout the growing season each year. The entire site was mowed to a height of 10cm in July 2009 to decrease annual weed competition and promote the establishment of native species (Williams et al. 2007).

No management was used on plots in 2010 or 2013. In April 2011 and 2014, plots were burned to stimulate native plant growth and reduce tree encroachment. In April 2012, >50 tons of biomass were harvested from plots using a flail-mower and were bailed. Material was later pelletized and then burned by Cedar Falls Utilities (Cedar Falls, IA) in a coal-fired power plant to test electrical production (Table 1). Non-plot areas were harvested in late summer of 2013 and sold to a local farmer for livestock feed.

Table 1. Research plot management from 2008-2014

Year	Management
2008	Soybean cultivation w/ 2x glyphosate application
2009	Seeding (May) and establishment mowing (July)
2010	No management
2011	Prescribed burn (April)
2012	Harvest (April)
2013	No management
2014	Prescribed burn (April)

Environmental Effects

In the year 2012 the Midwest experienced a considerable drought, with the city of Cedar Rapids (40 miles to the southeast), recording 25.13 inches of precipitation; a difference of 9.48 in from the annual average of 34.61 in (NOAA 2015).

The Cedar River crested above flood stage multiple times in May/June 2013 and 2014, inundating some of the research fields during our sampling periods (Fig. 2). The three soil types included in our study had distinct drainage classification and susceptibility to flooding (USDA 2015). During the 2013-2014 floods, plots on each soil type experienced highly variable frequency, depth, and duration of inundation, resulting in a wide-range of degree of disturbance and provided the setting for our "natural experiment".

The clay loam soil was the most poorly drained, lowest in elevation, and extensively flooded when river levels reached above 12.7 feet (3.87m), which occurred four times in 2013 and two times in 2014. In 2013 and 2014, water levels reached 1.8m and 1.3m respectively, and plots remained submerged for up to one week. The loam soil was the intermediate in drainage and elevation, and flooded moderately when river levels reached above 16.9 feet (4.15m), which occurred once in both 2013 and 2014. In 2013 and 2014, water depths reached up to 50cm and 30cm respectively, and plots remained submerged for up to two days (Figure 3). The sandy loam soil was the most well drained, highest in elevation and experienced little to no flooding (Figure 2).

Floral Surveys

The floral resources of research plots were measured from June 1 to September 30, 2010-2014. Each plot was surveyed five times during the season in periods: June, June/July, July, August and September. Each day we selected one plot of each crop in each of two or three soil types to survey. Thus four to six days were needed to complete a

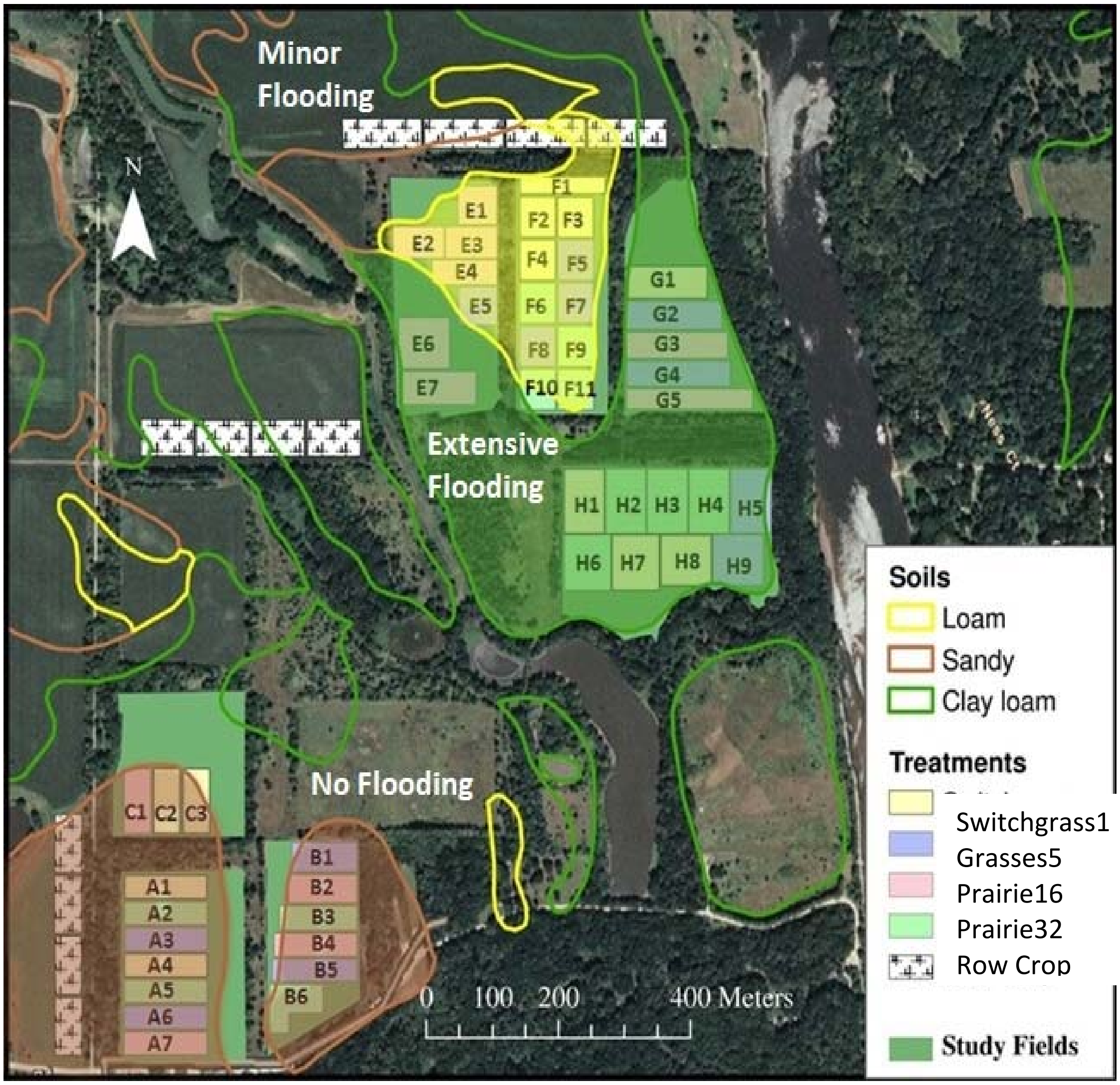
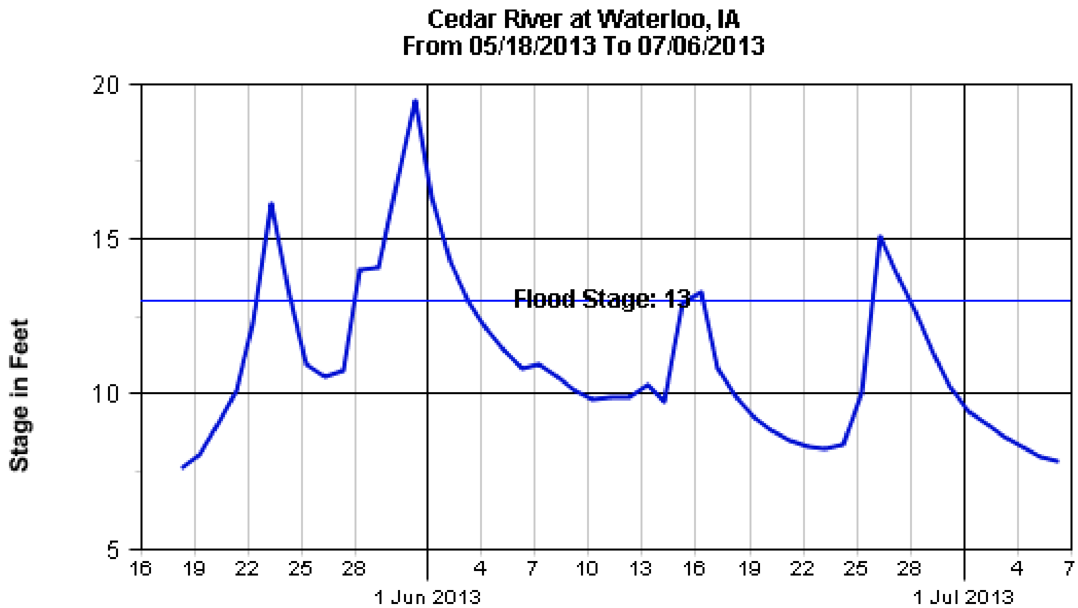


Figure 2. Map of study site at Cedar River Natural Resource Area depicting flooding severity on each of the three soil types. Green shading indicates extensive flooding. Yellow shading indicates a moderate level of flooding. Brown shading indicates little to no flooding. The Cedar River is located directly to the east of the site.

survey period and temporal bias was avoided. Floral resources were quantified in twenty 1 m² quadrats along a permanent 50 m transect established in the center of each plot. Quadrats were placed at 2 m intervals along each transect and randomized by: starting



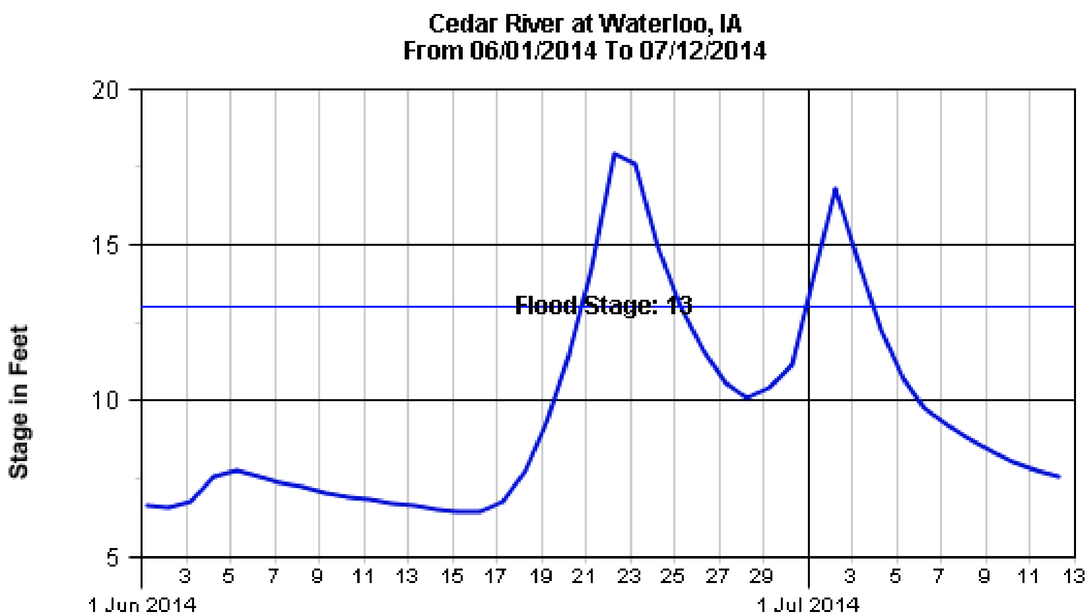


Figure 3. 2013 (top) and 2014 (bottom) water levels from USGS gage at the Cedar River at Waterloo, IA, about 16 miles upstream from the study site. The clay loam and loam soils flood when water levels reach above 12 and 15 feet, respectively. (USGS 2015)

location (0-9 m), left or right of line, and distance from line (1-3 m). Within each quadrat we estimated the number of inflorescences of each flowering forb species.

<u>Butterfly Surveys</u>

Butterfly surveys were conducted from June 1 to September 30, 2010-2014. Each plot was surveyed twice in each of the five periods. A random sample of 16 to 24 plots were surveyed each day, four plots from each crop with at least one plot from each soil type. All plots were surveyed over two to three days to minimize temporal variation. Previously established 50 m transects were walked at a rate of 10 $m\text{-}min^{-1}$ and butterflies that occurred within a 3 m window around the observer were recorded. Surveys took place between 10:00 and 19:00 on warm (18-36°C) days with winds <21.5 km/hr. Individuals were identified on the wing when possible. If not, individuals were captured and identified in hand, or sacrificed and placed in glassine envelopes for identification in the lab.

<u>Data Analysis</u>

For each of the five years (2010-2014) I summed butterfly counts from each sampling period to create a pooled dataset representing the entire year's surveys. I calculated yearly total butterfly abundance, the abundance of each species, total species richness, and Shannon's diversity index for each plot. I averaged the abundance of each flower species in each survey period within years to get the average number of flowers/m^2 for each plot. I then summed all data from each period to create a pooled dataset representing the entire year's surveys for a single plot, a snapshot of an entire growing season's floral assemblage in 1m^2. From this pooled dataset I calculated for each year the average abundance, species richness, and Shannon's diversity index for: sown and unsown forbs in each plot. Sown forbs refer to any species that is listed in Table A1 that occurs in any crop, and unsown forbs refer to any species that occurred in a plot that is not listed in Table A1.

I fit generalized linear mixed models (GLMM; Bolker et al. 2009) with butterfly and flower abundance, species richness, and community diversity as response variables; agrofuel crop, soil type, and year as fixed factors; and plot ID as a random factor accounting for spatial variation among plots. Data was analyzed in two combinations of years: 2010-2014 and 2010-2012. I restricted my primary analysis of soil type to a subset (2010-2012) of the data because soil effects were confounded with flooding in 2013 and 2014. In analyses assessing the effects of flooding, I used only the forb-rich crops, used the full (2010-2014) data set, and looked for evidence of a significant soil × year interaction.

I tested for variation in butterfly community composition by soil type, year and plot within the pooled forb-rich crops using permutational multivariate analysis of variance (PERMANOVA). I used the Bray-Curtis dissimilarity measure to create a distance matrix, completed 9,999 permutations, and performed a posteriori par-wise comparison test of significant main effects and/or interaction terms. Non-metric multidimentional scaling (NMDS) was used to visualize patterns of variation in butterfly community composition between the soil type and year groups (Anderson et al. 2008). A portion of the individual butterfly species were analyzed employing the same GLMM described above to test for differences across years. Species were excluded if their 5-year average was less than 1 individual encountered per plot. Of the 41 butterfly species observed during the study, 6 species were analyzed and include: *Colias* sp. (*C. eurytheme* and *C. philodice*), *Vanessa atalanta, Phyciodes tharos, Danaus plexippus* and *Everes comyntas.*

I investigated butterfly response to floral resources by exploring the relationship between (1) log (x +1)-transformed average butterfly and log (x +1)-transformed total floral abundance and (2) total butterfly and floral species richness within each of the 5 years and over all years using least squares regression. Statistical analyses were performed using the lme4 and lmerTest packages in R (R Core Team 2013) and PRIMER 6 (version 6.1.13) with PERMANOVA+ (version 1.0.3) (PRIMER-E Ltd., Plymouth PL1 3DH, UK) software.

CHAPTER 3

RESULTS

Butterfly and Floral Abundance, Richness and Community Diversity

Butterflies

Over the course of the study I recorded 4769 butterflies representing 41 species (Table 2). Average butterfly abundance, richness, and community diversity varied significantly by the main effects of agrofuel crop and year (Table 3, Figure 4, Figure A1). Average abundance ($F_{3, 36}$ = 168.74, P < 0.001), richness ($F_{3, 36}$ = 50.67, P < 0.001), and community diversity ($F_{3, 36}$ = 7.71, P < 0.001) were significantly greater in the forb-rich crops than the grass crops (Table A2, all P < 0.05) each year, but there were no significant differences between Prairie16 and Prairie32 nor between Switchgrass1 and Grasses5 for average abundance, species richness, or community diversity (Table A2, all P > 0.05).

Butterfly abundance ($F_{4,144}$ = 172.72, P < 0.001) was greater in 2010 than all other years (Table A2, all P < 0.001); greater in 2012 than 2011 (Table A2, P < 0.001); and similar (Table A2, P = 0.70) and significantly lower than all other years in 2013 and 2014 (Figure 4). Richness ($F_{4,144}$ = 69.98, P < 0.001) and diversity ($F_{4,144}$ = 14.05, P < 0.001) were greatest in 2010 and lowest in 2013 (Table A2, all P < 0.05)

During the 2010-2012 growing seasons, average abundance but not species richness nor community diversity varied significantly by soil type (Table 4). Average

Table 2. Butterfly species and number observed. Butterflies classified as habitat specialists (hs), habitat generalists (hg), or woodland species (w) (Vogel et al. 2010).

Common name	Scientific name	Guild	2010	2011	2012	2013	2014	Total
	Family *Hesperiidae*							
Delaware skipper	*Anatrytone logan*	hs	0	0	1	0	0	1
Least skipper	*Ancyloxypha numitor*	hg	2	0	0	0	0	2
Sachem	*Atalopedes campestris*	hg	3	0	3	3	0	9
Silver spotted skipper	*Epargyreus clarus*	hg	0	1	0	0	0	1
Funeral's duskywing	*Erynnis funeralis*	hg	2	0	0	0	0	4
Unidentified duskywing	*Erynnis sp.*	hg	0	0	1	0	0	1
Fiery skipper	*Hylephila phyleus*	hg	21	2	5	0	1	29
Checkered skipper	*Pyrgus communis*	hg	5	0	0	1	0	6
Peck's skipper	*Polites peckius*	hg	0	0	1	0	0	1
Tawny-edged skipper	*Polites themistocles*	hg	0	0	1	0	0	1
	Family *Nymphalidae*							
Hackberry emperor	*Asterocampa celtis*	w	7	11	44	0	0	62
Tawny emperor	*Asterocampa clyton*	w	1	0	0	0	0	1
Meadow fritillary	*Boloria bellona*	hs	0	0	2	2	0	4
Monarch	*Danaus plexippus*	hg	176	31	32	11	43	293
Variegated fritillary	*Euptoieta claudia*	hg	3	1	8	1	2	15
Buckeye	*Junonia coenia*	hg	67	2	19	15	3	106
American snout	*Libytheana carinenta*	w	0	0	0	0	2	2
Viceroy	*Limenitis archippus*	hs	8	9	2	0	5	24
Red spotted purple	*Limenitis arthemis*	w	0	0	0	0	1	1
Morning cloak	*Nymphalis antiopa*	w	1	0	2	0	1	4
Pearl crescent	*Phyciodes tharos*	hg	113	31	32	11	43	293
Eastern comma	*Polygonia comma*	w	8	0	0	0	2	10
Question mark	*Polygonia interrogationis*	*w*	2	2	5	0	1	10
Great spangled fritillary	*Speyeria cybele*	hs	8	28	52	0	22	110
Regal fritillary	*Speyeria idalia*	hs	2	0	0	0	0	2
Red admiral	*Vanessa atalanta*	hg	443	15	18	1	33	510
Painted lady	*Vanessa cardui*	hg	19	0	11	6	10	46
American lady	*Vanessa virginiensis*	hg	3	0	0	0	1	4
	Family *Papilionidae*							
Giant swallowtail	*Papilio cresphontes*	w	0	1	6	0	1	8
Tiger swallowtail	*Papilio glaucus*	w	2	2	1	1	2	8
Black swallowtail	*Papilio polyxenes*	hg	12	4	1	12	0	29
	Family *Pieridae*							
Orange/clouded sulphur	*Colias eurytheme/philodice*	hg	923	163	345	112	89	1632
Little yellow	*Eurema lisa*	hg	17	0	2	0	1	20
Dainty sulphur	*Nathalis iole*	hg	3	0	18	1	0	22
Cabbage white	*Pieris rapae*	hg	25	27	7	8	11	78
Checkered white	*Pontia protodice*	hg	1	0	0	0	0	1
	Family *Lycaenidae*							
Summer azure	*Celastrina neglecta*	hg	15	29	4	0	9	57
Eastern tailed blue	*Everes comyntas*	hg	206	359	375	248	217	1405
Gray copper	*Lycaena dione*	hs	0	0	0	0	1	1
Bronze copper	*Lycaena hyllus*	hs	1	2	0	0	0	3
Gray hairstreak	*Strymon melinus*	hg	12	0	1	0	0	13
Individuals observed			2111	699	1000	484	473	4767
Species observed			31	19	28	15	23	41

Table 3. Generalized linear mixed models comparing butterfly average abundance (log-transformed), species richness, and diversity (Shannon's) by agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
Average Abundance				
Agrofuel crop	3	6.77	168.74	<0.001
Soil type	2	0.63	15.63	0.000
Year	4	6.93	172.72	<0.001
Agrofuel crop × soil type	6	0.10	2.58	0.035
Agrofuel crop × year	12	0.27	6.65	<0.001
Soil × year	8	0.14	3.56	<0.001
Agrofuel crop × soil × year	24	0.08	1.96	0.009
Species richness				
Agrofuel crop	3	112.40	50.67	<0.001
Soil type	2	1.43	0.65	0.530
Year	4	155.23	69.98	<0.001
Agrofuel crop × soil type	6	3.27	1.47	0.215
Agrofuel crop × year	12	6.33	2.86	0.002
Soil × year	8	3.22	1.45	0.180
Agrofuel crop × soil × year	24	1.87	0.84	0.680
Community Diversity				
Agrofuel crop	3	1.12	7.71	<0.001
Soil type	2	0.47	3.26	0.041
Year	4	2.04	14.05	<0.001
Agrofuel crop × soil type	6	0.18	1.23	0.295
Agrofuel crop × year	12	0.14	0.98	0.474
Soil × year	8	0.13	0.93	0.495
Agrofuel crop × soil × year	24	0.10	0.68	0.868

butterfly abundance ($F_{3,36} = 18.16$, $P < 0.001$) was greatest on sandy loam (Table A3, all $P < 0.05$) and significantly lower on loam than clay loam (Table A3, $P = 0.007$). Butterfly abundance did not vary significantly between soils in 2010, was greatest on sandy loam in 2011, and was greater on sandy loam and clay loam than loam 2012 (soil type × year interaction: $F_{4,72} = 4.19$, $P = 0.004$).

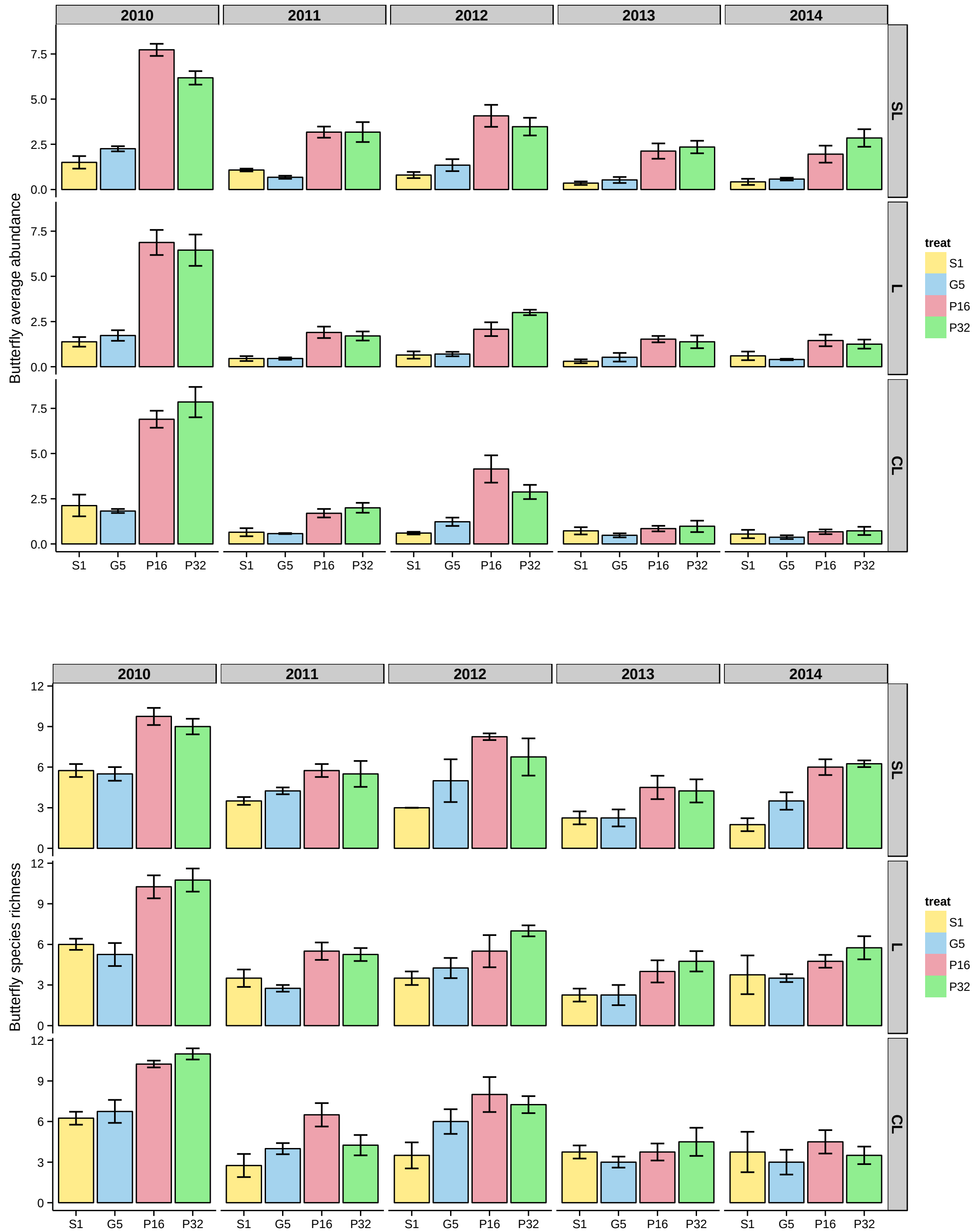


Figure 4. Butterfly average abundance (top) and species richness (bottom) by agrofuel crop, year and soil type (2010-2014)

Table 4. Generalized linear mixed models comparing butterfly average abundance (log-transformed), species richness, and diversity (Shannon's) by agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
Average Abundance				
Agrofuel crop	3	7.54	273.41	<0.001
Soil type	2	0.50	18.16	<0.001
Year	2	6.59	239.41	<0.001
Agrofuel crop × soil type	6	0.02	0.60	0.727
Agrofuel crop × year	6	0.20	7.28	<0.001
Soil type × year	4	0.12	4.19	0.004
Agrofuel crop × soil type × year	12	0.06	2.22	0.016
Species Richness				
Agrofuel crop	3	56.66	45.64	<0.001
Soil type	2	2.06	1.66	0.201
Year	2	159.53	128.52	<0.001
Agrofuel crop × soil type	6	1.34	1.08	0.387
Agrofuel crop × year	6	7.65	6.16	<0.001
Soil type × year	4	3.28	2.64	0.038
Agrofuel crop × soil type × year	12	1.34	1.08	0.384
Shannon's Diversity Index				
Agrofuel crop	3	0.35	5.54	<0.001
Soil type	2	0.12	1.85	0.169
Year	2	1.29	20.41	<0.001
Agrofuel crop × soil type	6	0.11	1.77	0.126
Agrofuel crop × year	6	0.13	2.07	0.064
Soil type × year	4	0.04	0.65	0.628
Agrofuel crop × soil type × year	12	0.07	1.10	0.372

Butterfly community composition in forb-rich crops (Table 5, Figure 5) varied significantly by soil ($F_{2, 119} = 9.42$, $P < 0.001$) and year ($F_{4,119} = 24.832$, $P < 0.001$), and displayed a significant soil × year interaction ($F_{8,119} = 1.95$, $P < 0.001$), though it explained relatively little of the variation in community composition. The main effect of

Table 5. PERMANOVA comparing butterfly community composition within the forb-rich crops by year and soil type (2010-2014).

Source of variation	df	Pseudo-F	*P*
Community Composition			
Soil type	2	9.42	<0.001
Year	4	24.83	<0.001
Plot (soil)	21	1.06	0.303
Soil type × year	8	1.95	<0.001
Residual	84		
Total	119		

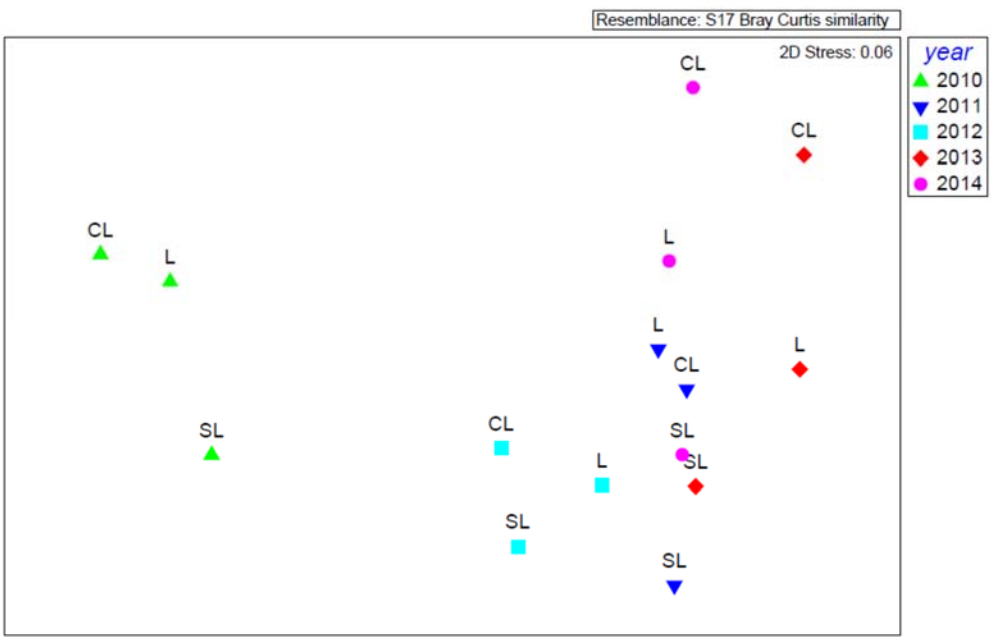


Figure 5. NMDS of 2010-2014 butterfly community composition. Letters represent soil type: CL = clay loam, L = loam, and SL = sandy loam; and colors/shapes, years.

year can be visualized along the horizontal NMDS axes through the separation of 2010 from the other years (Figure 5). The main effect of soil can be visualized along the vertical NMDS axes through the separation of loam and clay loam from sandy loam.

Butterfly communities were similar to one another on loam and clay loam, and somewhat distinct from those from those on sandy loam, in 2010 (no management) and 2011 (prescribed burn), but there was little variation among soils in 2012 (harvest). Communities in the no management (2010 and 2013) did not have similar butterfly assemblages. Butterfly communities in the burn years (2011 and 2014) were not distinct from other years.

Of all butterflies encountered during the study 25 species were habitat generalists, 7 were habitat specialists, and 9 were woodland species (adapted from Vogel et al. 2010). Habitat generalists represented 93.7% of all individuals encountered, and six species (*Colias eurytheme/C. philodice, Everes comyntas, Vanessa atalanta, Danaus plexippus* and *Phyciodes tharos)* accounted for 86.7% of total individuals. In the first burn year (2011) butterfly abundance dropped by 77% and richness by 35% from 2010. Of the six-habitat generalist species analyzed (*Colias eurytheme/C. philodice* ($F_{4,72}$ = 81.08, P < 0.001, Table A4, Figure 6)*, Everes comyntas* ($F_{4,72}$ = 13.84, P <0.001, Table A6, Figure 7)*, Vanessa atalanta* ($F_{4,90}$ = 181.52, P < 0.001,Table A8, Figure 8)*, Danaus plexippus* ($F_{4,72}$ = 31.16, P < 0.001, Table A10, Figure 9) and *Phyciodes tharos* ($F_{4,120}$ = 30.70, P < 0.001, Table A12, Figure 10)) all decreased in abundance in 2011 (Table A5, Table A7, Table A9, Table A11, Table A13, all $P < 0.001$) except for *E. comyntas,* which increased in abundance (Table A7, $P < 0.001$). In 2012, the year of haying on site, total butterfly abundance increased by 46% and richness by 29%. All six species analyzed remained similarly abundant in 2011 relative to 2012, besides *Colias sp.,* which significantly increased in abundance (Table A5, $P < 0.001$). Results after 2013 are possibly confounded by flood events. In 2013, a year of no management, total butterfly abundance decreased by 52% and richness by 46%. All species analyzed significantly decreased in abundance (Table A5, Table A7, Table A9, Table A11, Table A13, all *P* < 0.001), except for *P. tharos,* which remained stable (Table A113, *P* > 0.05), *and Colias sp.* (Table A5, *P* < 0.001), which significantly increased. 2014 was a burn year, and while total butterfly abundance decreased by 2% richness increased by 35%. This year both *D. plexippus* and *V. atalanta* significantly increased (Table A11, Table A9, all *P* < 0.05), *P. tharos* significantly decreased (Table A13, *P* < 0.001), and both *Colias sp.* and *E. comyntas* remained stable (Table A5, Table A7, P > 0.05). The remainder of the butterfly species encountered during the study could not be statistically assessed for their response to management, however a notable increase in the habitat specialist *Speyeria cybele* and decrease in the generalist species *Junonia coenia* was apparent in both burn years.

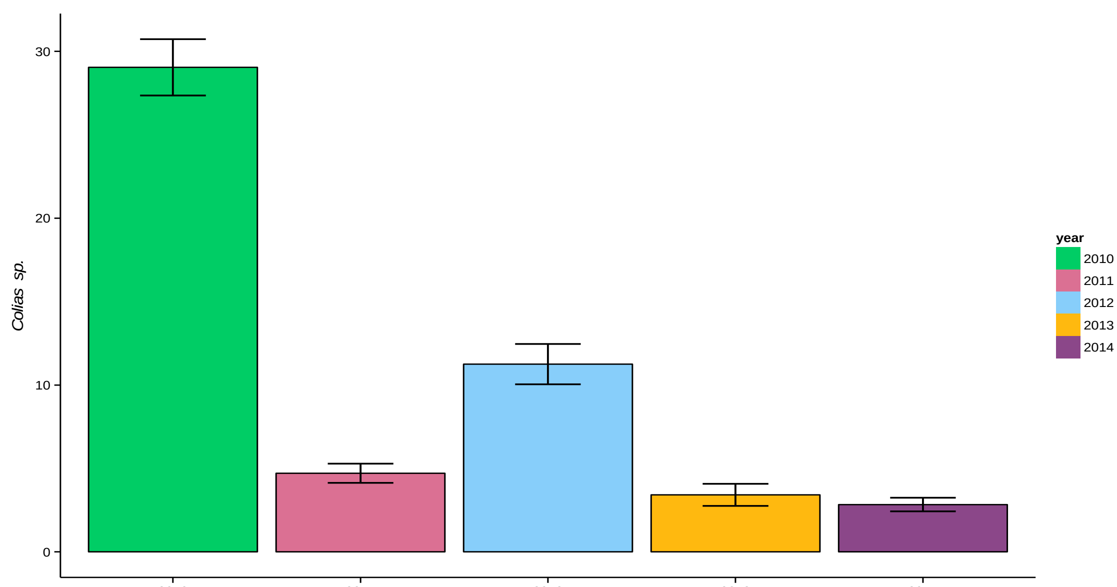


Figure 6. Average abundance per plot of *Colias eurytheme/philodice* in a forb rich crop (2010-2014).

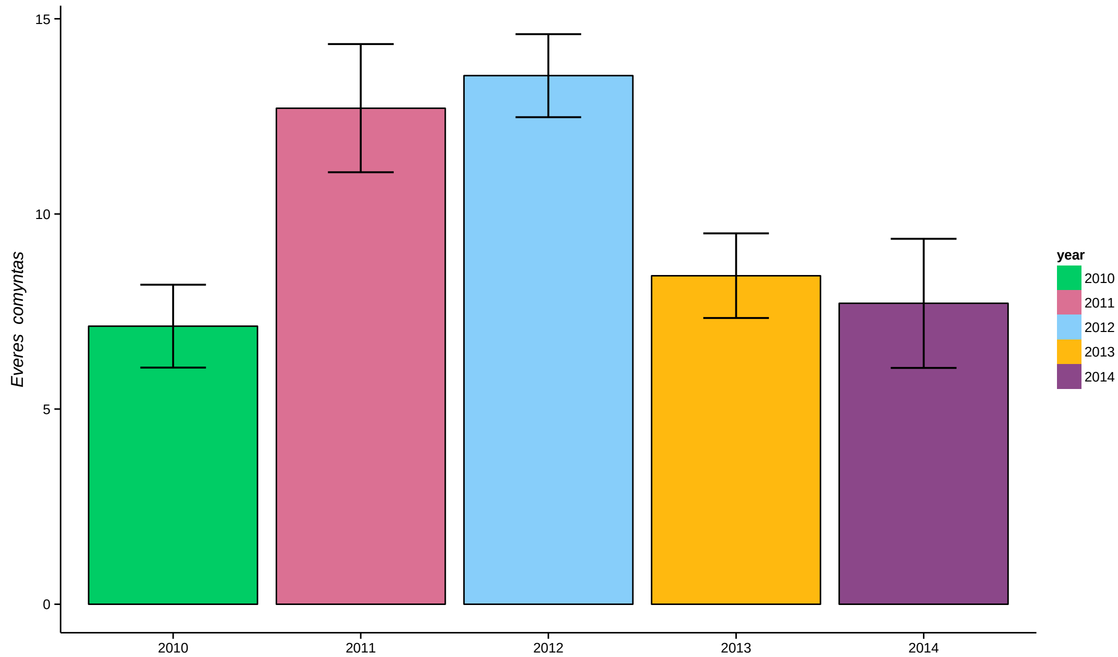


Figure 7. Average abundance per plot of *Everes comyntas* in a forb rich crop (2010-2014).

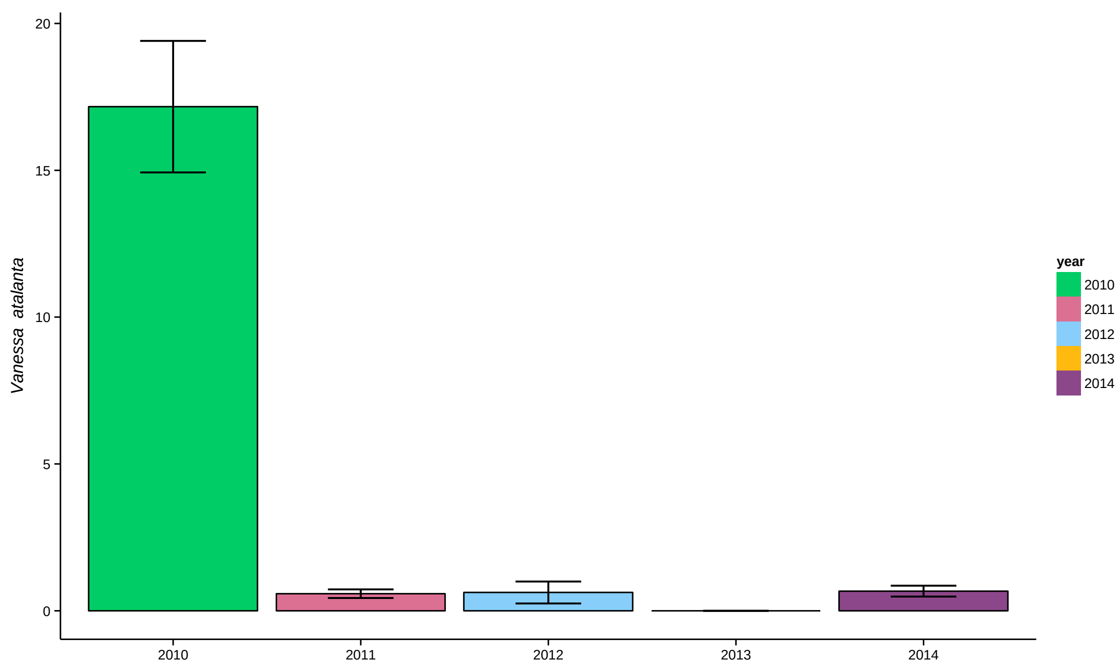


Figure 8. Average abundance per plot of *Vanessa atalanta* in a forb rich crop (2010-2014).

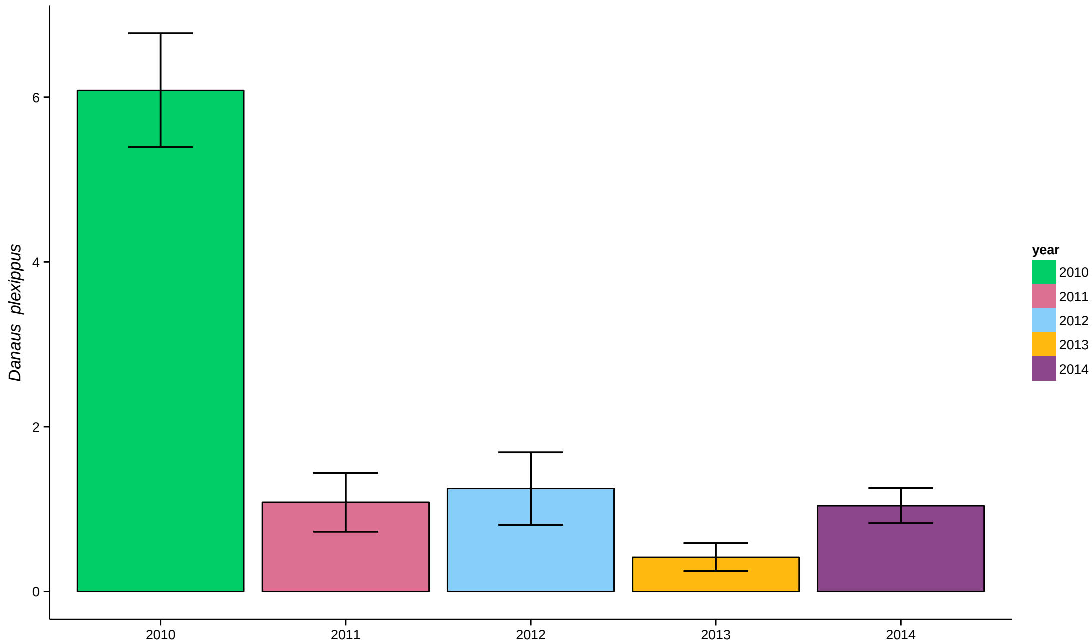


Figure 9. Average abundance per plot of *Danaus plexippus* in a forb rich crop (2010-2014).

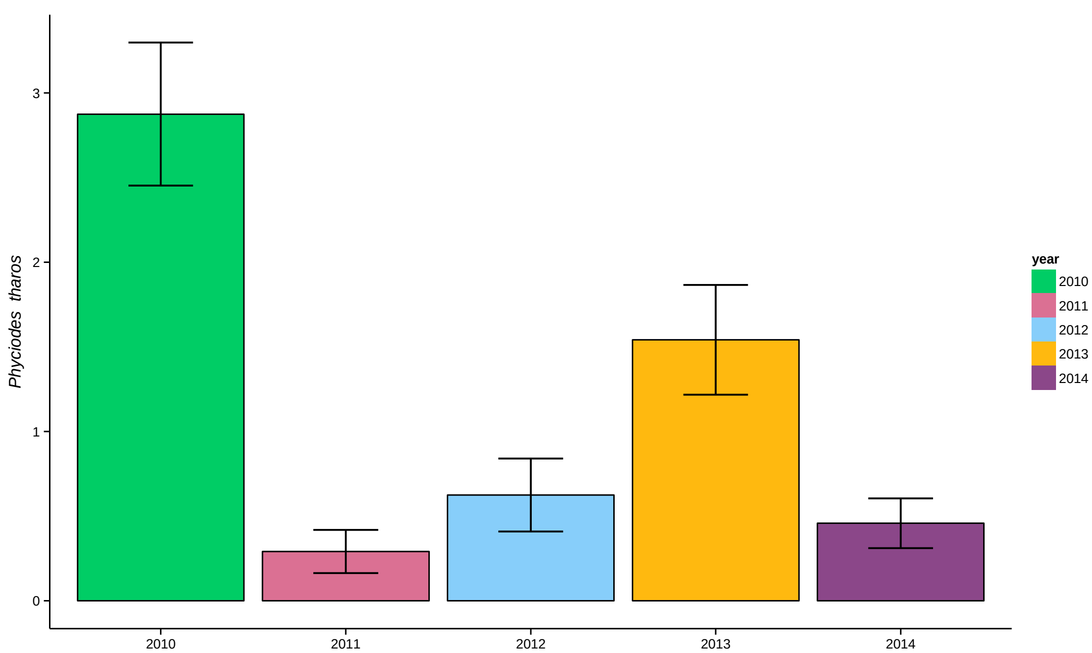


Figure 10. Average abundance per plot of *Phyciodes tharos* in a forb rich crop (2010-2014).

Habitat specialists occurred in greater proportional abundance in 2011, 2012, and 2014 than 2010 and 2013. *Speyeria cybele* was the most abundant habitat specialist encountered during surveys and represented 76% of total habitat specialist observations (Table 2, Table 6). Habitat specialist species richness ranged from 1 to 4 species

Table 6. Butterfly habitat characteristics and percentage of total abundance by year.

Year/Management	Habitat generalists	Habitat specialists	Woodland
2010/No Management	98.1	0.9	1.0
2011/Burn	92.2	5.6	2.5
2012/Hay	88.4	5.7	6.6
2013/No Management	99.4	0.4	0.2
2014/Burn	92.0	5.9	2.3
TOTAL	93.7	3.0	2.2

annually. Proportional abundance of habitat specialists was lowest in years of no management (2010 = 0.9%: *S. cybele*, *Speyeria idalia*, *L. archippus* and *Lycaena hyllus*; 2013 = 0.4%: *Boloria bellona)* and highest in years of burning (2014 = 5.9%: *L. archippus*, *Lycaena dione and S. cybele*; 2011 = 5.6%: *L. archippus, L. hyllus, S. cybele*) or haying (2012= 5.7%: *Anatrytone logan, B. bellona, L. archippus* and *S. cybele)*. Habitat generalists followed the opposite trend with their lowest abundance recorded in 2012, the same year woodland species abundance peaked (Table 2, Table 6).

<u>Sown Flowers</u>

Over the course of the study we recorded 60 forb species in bloom: 19 sown and 41 unsown (Table 7). All sown forb species (Table A1) except *Amorpha canescens* were

Table 7. Average floral abundance/m^2 (mean, SE) of all sown forbs and the nine most abundant unsown forbs by year. Unsown forbs exceeding 0.5 inflorescences/m^2 averaged over the entire study were included.

	2010		2011		2012		2013		2014	
	x̄	SE	x̄	SE	x̄	SE	x̄	SE	x̄	SE
Sown species										
Artemisia ludoviciana	0.004	0.003	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Amorpha canescense	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Astragalus canadensis	0.33	0.10	0.78	0.22	0.17	0.07	0.01	0.01	0.01	0.01
Baptisia leucantha	0.00	0.00	0.00	0.00	0.05	0.03	0.009	0.004	0.04	0.02
Dalea purpurea	0.008	0.004	0.01	0.009	0.04	0.02	0.003	0.003	0.03	0.02
Desmodium canadense	14.7	3.53	22.9	4.3	0.78	0.32	1.66	0.57	5.20	1.75
Echinacea pallida	0.23	0.06	0.16	0.06	0.03	0.01	0.04	0.02	0.07	0.03
Erynigium yuccifolium	0.09	0.04	0.42	0.13	0.02	0.01	0.05	0.02	0.43	0.16
Helianthus grosseseratus	0.32	0.12	0.42	0.17	0.10	0.07	0.20	0.14	0.20	0.14
Helopsis helianthoides	20.1	3.81	18.1	4.04	3.09	1.00	2.79	1.20	1.01	0.49
Lespedeza capitata	0.007	0.004	0.13	0.05	0.10	0.03	0.03	0.02	0.07	0.02
Monarda fistulosa	0.32	0.12	0.18	0.07	0.06	0.03	0.04	0.02	0.01	0.007
Oligoneuron rigidum	0.93	0.15	1.05	0.19	1.37	0.29	0.86	0.29	0.58	0.18
Phlox pilosa	0.51	0.28	0.37	0.15	0.10	0.05	0.09	0.05	0.01	0.007
Ratibida pinnata	2.01	0.49	1.12	0.30	0.37	0.12	1.19	0.39	0.45	0.13
Silphium laciniatum	0.00	0.00	0.02	0.02	0.02	0.01	0.07	0.03	0.02	0.01
Symphyotrichum laeve	1.00	0.38	0.46	0.20	0.07	0.05	0.01	0.01	0.09	0.06
Symphyotrichum novae-angliae	0.08	0.07	0.02	0.02	0.04	0.02	0.03	0.02	0.06	0.03
Tradescantia bracteata	0.01	0.01	0.12	0.04	0.04	0.03	0.33	0.11	0.77	0.22
Zizia aurea	0.00	0.00	0.27	0.11	0.005	0.004	0.03	0.02	0.03	0.01
Unsown species										
Chamaecrista fasciculata	0.27	0.13	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00
Erigeron strigosus	2.09	0.56	0.76	0.32	0.32	0.24	0.03	0.02	0.001	0.001
Melilotus alba	2.38	0.84	0.00	0.00	0.05	0.03	0.18	0.12	0.00	0.00
Melilotus officinalis	13.6	3.79	0.00	0.00	0.14	0.06	0.46	0.22	0.00	0.00
Medicago sativa	0.14	0.05	0.14	0.11	0.003	0.003	0.00	0.00	0.00	0.00
Securigera varia	0.02	0.02	0.14	0.14	0.003	0.003	0.53	0.53	0.00	0.00
Solidago canadensis	0.45	0.10	0.35	0.08	0.34	0.10	0.22	0.06	0.45	0.14
Symphyotrichum pilosum	0.70	0.18	0.06	0.06	0.00	0.00	0.37	0.30	1.87	0.72
Trifolium pretense	0.36	0.13	0.00	0.00	0.001	0.001	0.00	0.00	0.00	0.00

recorded in bloom during at least one survey (Table 7); however, flowering *A. canescens* individuals were observed outside transects in 2014.

Sown floral abundance varied significantly by year; richness by year and treatment; and community diversity by treatment (Table 8, Figure 11, Figure 12). Sown floral abundance ($F_{2,24} = 0.56$, $P = 0.46$) did not vary between Prairie16 or Prairie32 (Table A14, $P = 0.569$). Abundance ($F_{4,96} = 101.36$, $P < 0.001$) was similar in 2010 and 2011 (Table A14, $P < 0.001$), and greater in those years than 2012-2014 (Table A14, all $P < 0.001$). Abundance did not vary between the 2012-2014 growing seasons. Sown floral richness and community diversity were greater in Prairie32 than Prairie16 (Table A14, $P < 0.001$).

Richness was greatest in 2011 (Table A14, all $P < 0.05$), and greater in 2010 than 2012-2014 (Table A14, all $P < 0.001$). Richness did not vary significantly between the 2012-2014 growing seasons. During the 2010-2012 growing seasons, sown floral abundance, richness, and community diversity varied significantly among soil types (Table 9, Figure 11, Figure 12). Sown floral abundance ($F_{2,24} = 17.62$, $P < 0.001$) was greatest in clay loam, and significantly greater in loam than sandy loam (Table A15, all $P < 0.05$). Sown richness ($F_{2,72} = 28.53$, $P < 0.001$) and diversity ($F_{2,24} = 9.45$, $P < 0.001$) were lower on loam than both sandy loam and clay loam (Table A15, all $P < 0.05$), which were similar to one another (Table A15, $P > 0.05$).

Sown floral abundance and richness displayed significant two and three way interactions (Table 9, Figure 11, Figure 12). Sown floral abundance ($F_{4,48} = 14.69$, $P <$

Table 8. Generalized linear mixed models comparing sown floral average abundance (log-transformed), species richness, and diversity (Shannon's) by agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
Average Abundance				
Agrofuel crop	1	0.17	0.56	0.460
Soil type	2	0.38	1.26	0.301
Year	4	30.74	101.36	<0.001
Agrofuel crop × soil type	2	0.23	0.77	0.474
Agrofuel crop × year	4	0.71	2.34	0.060
Soil × year	8	2.98	9.82	<0.001
Agrofuel crop × soil × year	8	0.27	0.89	0.532
Species Richness				
Agrofuel crop	1	765.08	384.54	<0.001
Soil type	2	40.51	20.36	<0.001
Year	4	36.43	18.31	<0.001
Agrofuel crop × soil type	2	9.92	4.99	0.008
Agrofuel crop × year	4	9.24	4.65	0.002
Soil × year	8	20.83	10.47	<0.001
Agrofuel crop × soil × year	8	8.75	4.40	<0.001
Community Diversity				
Agrofuel crop	1	6.64	71.98	<0.001
Soil type	2	0.74	8.05	<0.001
Year	4	0.10	1.03	0.394
Agrofuel crop × soil type	2	0.13	1.44	0.257
Agrofuel crop × year	4	0.19	2.04	0.095
Soil × year	8	0.44	4.81	<0.001
Agrofuel crop × soil × year	8	0.07	0.78	0.623

0.001) was similar on all soils in 2010 and 2011 (all $P > 0.05$), but flowers were most abundant on clay loam and least abundant on sandy loam in 2012, a drought year (all $P <$ 0.001). In 2010 and 2012, abundance ($F_{4,48} = 4.28$, $P = 0.005$) in Prairie32 was lower than Prairie16, and in all other years no significant differences were found between crops (all $P > 0.05$). Sown richness ($F_{4,72} = 4.90$, $P = 0.002$) significantly increased from 2010

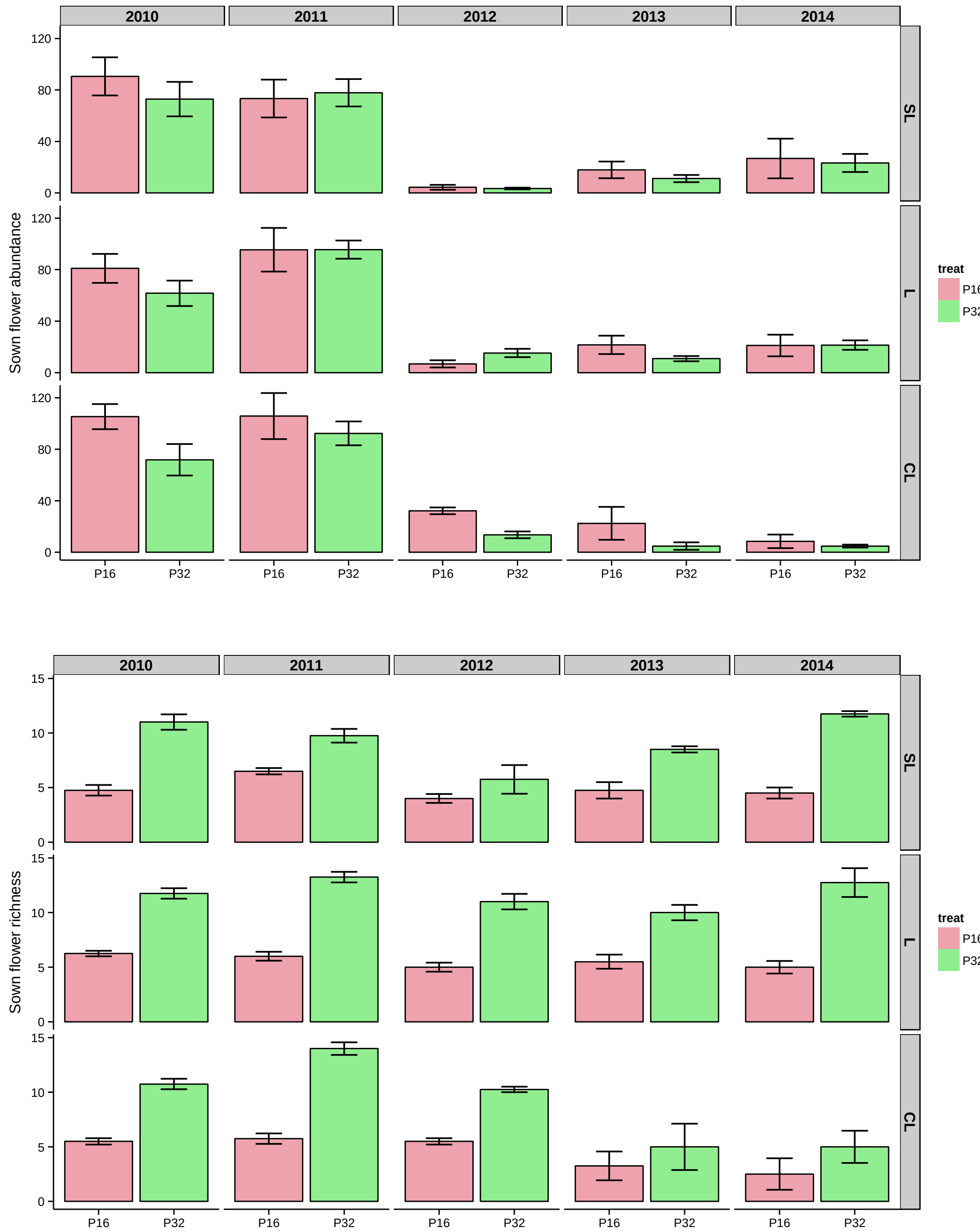


Figure 11. Average floral abundance (top) and richness (bottom) by year, soil type in Prairie16 and Prairie32 (2010-2014).

Figure 12. Average floral community diversity (Shannon's) by year and soil type in Prairie16 and Prairie32 (2010-2014).

to 2011, and again from 2011 to 2012 (Table A15, all $P < 0.001$). Sown floral richness ($F_{4,72} = 6.77$, $P = 0.001$) was higher in the Prairie32 on all soils and in all years, except in 2012 on sandy loam, when the Prairie16 and Prairie32 crops were similar.

Of the 19 species documented in bloom, 12 experienced peak abundance in 2010 or 2011 (*Artemisia ludoviciana, Astragalus canadensis, Desmodium canadense, Echinacea pallida, Helianthus grosseseratus, Heliopsis helianthoides, Lespedeza capitata, Monarda fistulosa, Phlox pilosa, Ratibida pinnata, Symphyotrichum laeve, Symphyotrichum novae-angliae,* and *Zizia aurea*). The two most abundant sown species in these years were *Desmodium canadense* and *Heliopsis helianthoides*, which represented ~43% and ~44% of all sown inflorescences. Seven species were more

Table 9. Generalized linear mixed models comparing sown floral average abundance (log-transformed), species richness, and Shannon's diversity index by agrofuel crop (Prairie16 and Prairie32 only), soil type and year (2010-2012).

Source of variation	df	MS	Pseudo-F	*P*
Abundance				
Agrofuel crop	1	0.12	1.29	0.267
Soil type	2	1.69	17.62	<0.001
Year	2	36.68	382.26	<0.001
Agrofuel crop × soil type	2	0.43	4.45	0.023
Agrofuel crop × year	2	0.18	1.86	0.167
Soil type × year	4	1.41	14.69	<0.001
Agrofuel crop × soil type × year	4	0.41	4.28	0.005
Richness				
Agrofuel crop	1	517.35	566.52	<0.001
Soil type	2	26.06	28.53	<0.001
Year	2	32.10	35.15	<0.001
Agrofuel crop × soil type	2	11.72	12.84	<0.001
Agrofuel crop × year	2	6.93	7.59	0.001
Soil type × year	4	4.47	4.90	0.002
Agrofuel crop × soil type × year	4	6.18	6.77	0.001
Diversity				
Agrofuel crop	1	1.59	30.30	<0.001
Soil type	2	0.50	9.45	<0.001
Year	2	0.16	3.08	0.055
Agrofuel crop × soil type	2	0.05	0.94	0.404
Agrofuel crop × year	2	0.06	1.08	0.348
Soil type × year	4	0.07	1.25	0.302
Agrofuel crop × soil type x year	4	0.02	0.40	0.811

abundant in 2014 than 2010 (*Baptisia leucantha*, *Dalea purpurea*, *Eryngium yuccifolium*, *Lespedeza capitata*, *Silphium laciniatum*, *Tradescantia bracteata* and *Zizia aurea*) and of these, three did not flower in 2010 (*Baptisia leucantha*, *Silphium laciniatum* and *Zizia aurea*) (Table 7).

<u>Unsown Flowers</u>

Unsown floral abundance, richness, and diversity varied significantly by both treatment and year over the 2010-2014 growing seasons (Table 10, Figure A2, Figure A3). Unsown flowers were less abundant ($F_{3,48} = 8.19$, $P < 0.001$) in Grasses5 than Switchgrass1 and the forb-rich crops (Table A16, all $P < 0.05$). Unsown floral abundance was greater in Prairie16 than Prairie32 (Table A16, P = 0.023). Unsown flowers were most abundant in 2010 (Table A16, all $P < 0.001$), and did not vary over the 2011-2014 growing seasons (Table A16, all $P < 0.05$). During the 2010-2012 growing seasons, unsown floral abundance (Table 11, $F_{3,48} = 6.21$, $P = 0.001$) was greatest on the loam soil in 2010 (F= 9.49, $P < 0.001$). *Melilotus officinalis* and *M. alba* were most abundant on this soil and represented 60% of total unsown inflorescenses from 2010-2102 (Table 7). Unsown richness ($F_{3,48} = 9.31$, $P < 0.001$) in 2010-2014 was greater in the forb-rich crops than Grasses5, and greater in Prairie16 than Switchgrass1 (Table A16, all $P < 0.05$).

Richness ($F_{3,48} = 9.31$, $P < 0.001$) was similar between Switchgrass1 and Grasss5 in 2010-2014 (Table A16, $P = 0.219$). Unsown richness was greatest in 2010 (Table A16, all $P < 0.001$) and did not vary between the 2011-2014 growing seasons (Table A16, all $P > 0.05$). Unsown community diversity was greater in the forb-rich crops than the grass crops (Table A16, all $P < 0.05$) and did not vary between the Switchgrass1 and Grasses5 nor between Prairie16 and Prairie32 (Table A16, all $P > 0.05$).

Of the 42 species documented in bloom, only nine species averaged 0.05 flowers/m^2 or more over the entire study and six of them experienced peak abundance in

Table 10. Generalized linear mixed models comparing unsown floral average abundance (log-transformed), species richness, and diversity (Shannon's) by agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
Average Abundance				
Agrofuel crop	3	2.70	8.19	<0.001
Soil type	2	1.44	4.38	0.018
Year	4	31.31	95.02	<0.001
Agrofuel crop × soil type	6	0.32	0.97	0.457
Agrofuel crop × year	12	0.59	1.79	0.053
Soil × year	8	4.12	12.51	<0.001
Agrofuel crop × soil × year	24	0.42	1.29	0.176
Species Richness				
Agrofuel crop	3	8.58	8.80	<0.001
Soil type	2	1.57	1.61	0.211
Year	4	169.04	173.28	<0.001
Agrofuel crop × soil type	6	0.26	0.27	0.948
Agrofuel crop × year	12	11.55	11.84	<0.001
Soil × year	8	2.15	2.21	0.028
Agrofuel crop × soil × year	24	1.13	1.16	0.286
Community Diversity				
Agrofuel crop	3	0.56	6.60	<0.001
Soil type	2	0.14	1.65	0.204
Year	4	4.53	53.59	<0.001
Agrofuel crop × soil type	6	0.02	0.28	0.945
Agrofuel crop × year	12	0.28	3.35	<0.001
Soil × year	8	0.67	7.97	<0.001
Agrofuel crop × soil × year	24	0.09	1.11	0.339

2010 or 2011 (Table 7). Two species (*Solidago canadensis* and *Symphyotrichum pilosum,* Table 7) were more abundant in 2014 than they were in 2010.

Table 11. Generalized linear mixed models comparing unsown floral average abundance (log-transformed), species richness, and diversity (Shannon's) by agrofuel crop, soil type and year (2010-2012).

Source of variation	df	MS	F	*P*
Abundance				
Agrofuel crop	3	2.46	6.21	0.001
Soil type	2	2.95	7.45	0.002
Year	2	54.67	137.83	<0.001
Agrofuel crop × soil type	6	0.19	0.47	0.824
Agrofuel crop × year	6	0.73	1.87	0.010
Soil type × year	4	3.76	9.49	<0.001
Agrofuel crop × soil type × year	12	0.32	0.99	0.467
Richness				
Agrofuel crop	3	11.01	9.31	<0.001
Soil type	2	1.07	0.91	0.411
Year	2	271.77	229.86	<0.001
Agrofuel crop × soil type	6	0.22	0.19	0.978
Agrofuel crop × year	6	19.54	16.53	<0.001
Soil type × year	4	2.92	2.47	0.049
Agrofuel crop × soil type × year	12	1.78	1.51	0.135
Diversity				
Agrofuel crop	3	0.89	9.06	<0.001
Soil type	2	0.49	4.97	0.011
Year	2	6.63	67.56	<0.001
Agrofuel crop × soil type	6	0.06	0.61	0.725
Agrofuel crop × year	6	0.32	3.24	0.006
Soil type × year	4	1.04	10.62	<0.001
Agrofuel crop × soil type × year	12	0.12	1.27	0.250

Butterfly-Flower Regression Analysis

There were highly significant positive linear relationships between butterfly and floral abundance and between butterfly and floral species richness during each year and over the complete 5-year study (Table 12, Figure 13).

Table 12. Butterfly and floral (abundance and richness) linear regression output by year.

Year	Intercept	Slope	P	r^2
Butterfly abundance by flower abundance (log (x+1)-transformed)				
2010	0.551	0.290	<0.001	0.591
2011	0.389	0.161	<0.001	0.620
2012	0.650	0.257	<0.001	0.414
2013	0.338	0.200	<0.001	0.508
2014	0.301	0.174	<0.001	0.371
All Years	0.423	0.323	<0.001	0.485
Butterfly richness by flower richness				
2010	4.660	0.327	<0.001	0.615
2011	3.530	0.140	<0.001	0.203
2012	4.430	0.239	0.001	0.181
2013	2.490	0.215	<0.001	0.305
2014	3.110	0.204	<0.001	0.211
All Years	3.240	0.301	<0.001	0.385

Effects of Flooding on Floral and Butterfly Communities

Sown floral abundance, richness, and community diversity in forb-rich crops displayed significant two-way interactions involving soil (Table 8, Figure 11, Figure 12). Abundance and richness ($F_{8,96} = 9.82$, P < 0.001; $F_{8,120} = 10.47$, $P < 0.001$) were significantly greater in both 2010 and 2011 on all soil types than any other year × soil type combination (all $P < 0.05$). Sown floral abundance in 2012 was greatly reduced across all soil types in response to drought (all $P < 0.001$). Flower abundance was correlated with drainage class and was greater on clay loam than loam, and greater on loam than sandy loam in 2012 (Table A4, all $P < 0.05$).

In the flood years of 2013 and 2014, sown floral abundance, richness, and diversity ($F_{8,96} = 9.82$, $P < 0.05$; $F_{8,120} = 10.47$, $P < 0.001$; $F_{8,,96} = 4.81$, $P < 0.001$) were

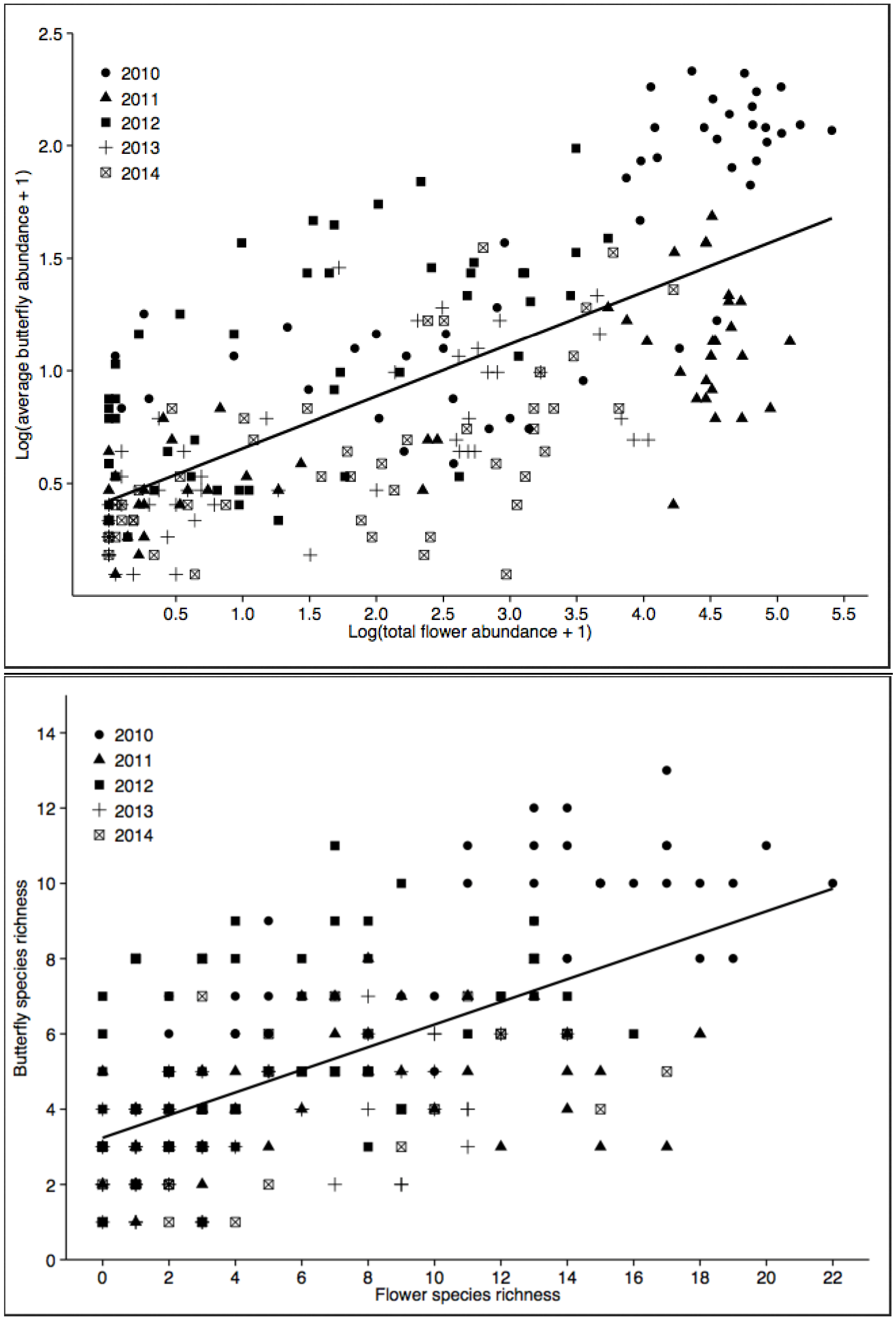


Figure 13. (Top) Relationship between butterfly average abundance and flower abundance over the 2010-2014 growing seasons. (Bottom) Relationship between butterfly richness and floral richness over the 2010-2014 growing seasons.

significantly lower on the extensively flooded clay loam than the moderately flooded loam (all $P < 0.05$), and were similar on loam and the not flooded sandy loam (all $P > 0.05$). Sown floral abundance, richness and diversity on the extensively flooded clay loam decreased significantly in 2013 and 2014 relative to 2012 (all $P < 0.001$) while unsown floral abundance ($F_{8,192} = 12.51$, $P < 0.001$, Table 10, Figure A2) increased (Table A16, $P < 0.001$). Sown floral abundance on the unflooded sandy loam increased in 2013 and 2014 relative to 2012 ($F_{8,96} = 9.82$, $P < 0.001$; all $P < 0.001$).

Butterfly abundance in the forb-rich plots displayed significant soil × year, agrofuel crop × soil, and agrofuel crop × soil × year interactions (Table 3, Figure 4). Butterfly average abundance decreased in 2013 and 2014 relative to 2012 across soil types (Table A2, all $P < 0.001$). Butterfly average abundance ($F_{8,144} = 3.56$, $P < 0.001$) in 2013 was similar between the moderately flooded loam and the extensively flooded clay loam ($P = 0.417$), which were both significantly lower than the not flooded sandy loam (all $P < 0.05$). In 2014 butterfly average abundance decreased in a stepwise manner with clay loam being significantly less than loam, which was significantly less than sandy loam (Table A2, all $P < 0.05$). There was great variation in butterfly community composition among soils in 2013 and 2014 compared to the 2010-2012 period (Figure 5). Butterfly communities were similar within and distinct among soil types during the 2013-2014 flood years (Figure 5).

CHAPTER 4
DISCUSSION

I studied butterfly use of four candidate native prairie agrofuel crops on three soil types beginning with crop establishment and continuing over a period of five years. I explored the successional changes in both communities and found variation in butterfly and floral abundance, species richness, and community diversity among crops and soil types and over time. I also documented evidence for the influence of flooding events on both communities in 2013 and 2014.

As expected, from 2010-2014 butterflies were ~3.6 times more abundant, ~1.4 times more species rich, and more diverse in the forb-rich crops than the grass crops. Sown flowers were more species rich and diverse, but not more abundant in Prairie32 than Prairie16. During early establishment (2010), floral abundance was greater in P16 than P32, and butterflies were similarly abundant and species rich in both forb-rich crops (Myers et al. 2012). I hypothesized that as succession proceeded, flower and butterfly abundance, richness, and community diversity would eventually be greater in Prairie32 than Prairie16. However, I did not observe the butterfly community responding to documented differences in the richness or diversity of the forb-rich crops. As the study progressed, I found that the floral community on site shifted, somewhat consistent with Schramm (1990). Early-successional, short-lived non-native (*Melilotus officinalis* and *Trifolium pratense*) and native forbs (*Heliopsis helianthoides, Ratibida pinnata,* and *Erigeron strigosus*) decreased in abundance, while long-lived native perennial prairie species (*Baptisia leucantha* and *Tradescantia bracteata*) increased. *Desmodium canadense* co-dominated the forb community with *Heliopsis helianthoides* in the first two years of the study, and the species *Phlox pilosa* and *Zizia aurea* decreased as the site matured, inconsistent with Schramm (1990). I hypothesize that the floral communities on site will continue to shift towards domination by long-lived perennial species, and if flower communities in Prairie16 and Prairie32 diverge in abundance as they have richness and diversity, the butterfly community will respond similarly.

Previous research has shown positive relationships between butterfly abundance and forb abundance (Waltz and Covington 2001), percent cover (Vogel et a. 2007), and number of ramets in bloom (Reeder et al. 2005; Shepherd and Debinski 2005). Other studies have shown positive associations between floral richness and the richness (Ries et al. 2001; Waltz and Covington 2001) or abundance (Ries et al. 2001) of butterflies, while some authors have reported no significant relationships (Hawkins and Porter 2003; Shepherd and Debinski 2005). I found that sown floral abundance and richness were strong predictors of butterfly abundance and richness when comparing the forb-rich and grass crops. However, butterfly richness and diversity did not respond to elevated sown forb abundance and diversity in Prairie32 compared to Prairie16, as predicted in Myers et al. 2012. The majority of the butterflies encountered on site were species whose larval host plants (Schlicht et al. 2007) were found in the landscape surrounding the agrofuel plots. The similarity of the butterfly communities in the forb-rich crops could be due to a lack of overlap between the larval host plant requirements of the butterfly species encountered on site and sown forb species, as well as a lack of habitat connectivity to established populations of butterflies that rely on specific larval host plants that occur in Prairie32 but not Prairie16 (Shepherd and Debinski 2005). My results indicate that butterfly communities will respond to agrofuel crop diversity and that the adoption of species-rich, forb containing assemblages of native plants as agrofuel feedstocks will provide higher quality habitat for butterflies than native, grass-only agrofuel feedstocks. My results suggest that butterflies were responding to the presence of abundant nectar sources in the forb-rich plots, but that they did not distinguish between the forb-rich crops even though Prairie32 contained greater species richness and diversity of sown forbs.

Habitat generalists dominated the butterfly community for the duration of the study, and their proportional abundance ranged from 88.4% to 99.4% annually. Most research in the Midwestern USA on prairie butterflies focuses on declining habitat specialist species (Swengel et al. 2011; Schlicht and Orwig 1998) while few have focused on habitat generalists. However, recent research has indicated declines in once common habitat generalist species from Europe (Van Dyck et al. 2009), and, more recently, the charismatic North American Monarch (*Danaus plexippus)* (Brower et al. 2012; Pleasants and Oberhauser 2012; Monarch Joint Venture 2015).

The Monarch (*Danaus plexippus*), has gained recent nationwide attention due to its decline over much of its range (Monarch Joint Venture 2015). This highly migratory species is dependent on species in the genus *Asclepias* as its larval host plants, and as an adult feeds on nectar of a wide range of forbs. Monarchs were commonly recorded during surveys on site, and a decline in yearly abundance from 2010-2014 parallels documented declines of overwintering population numbers in Mexico (Monarch Joint Venture, 2015).

Recent evidence has linked the reduction of common milkweed, *Asclepias syriaca*, and resulting reduction in the population of *Danaus plexippus* adults overwintering in Mexico to the increased cultivation of genetically modified glyphosate tolerant corn and soybeans in the Midwest (Pleasants and Oberhauser 2012). These crops currently dominate the agrofuel marketplace. The widespread adoption of diverse assemblages of native prairie plants as agrofuel crops that include milkweeds and abundant nectar sources at different seasonal periods could provide quality habitat for migrating and breeding *D. plexippus* individuals, and other generalist butterfly species in the Midwest.

Habitat specialist butterflies represented only 3% of all individuals observed. Of all habitat specialist species encountered, only one species, *Anatryone logan,* arrived to find potential larval host plants, *Andropogon gerardii* and *Panicum virgatum*, (Schlicht et al. 2007) within the seeded species. However, larval host plants of the remaining habitat specialists (*Boloria bellona- Viola sp.; Limentis archippus- Salicaceae sp.; Lycaena dione- Rumex sp.; Lycaena hyllus- Rumex sp. and Speyeria cybele- Viola sp.)* (Schlicht et al. 2007), except *Speyeria idalia*, were observed within fields or on field edges. *Speyeria idalia* was encountered in two surveys in 2010, and opportunistically during a floral survey in 2014. While its larval host plant (*Viola pedata*) was never observed on site, the use of the agrofuel crops by adults suggests that the targeted establishment of *Viola pedata* could provide habitat for a declining habitat specialist butterfly (Debinski and Kelly 1998). Land managers seeking to establish a harvestable agrofuel crop and provide habitat for specific butterfly species could seed desired larval host plants in addition to the Prairie16 or Prairie32 crops. Results indicate that diverse agrofuel crops could provide valuable nectar sources for habitat specialist butterflies with larval host plants that occur in the surrounding landscape, or are able to colonize agrofuel crops. The colonization and survival of butterfly populations in prairie reconstructions is dependent on a variety of factors including proximity and connectivity to established populations, (Shepherd and Debinski 2005), the availability of adult nectar sources and larval host plants, and the utilization of management techniques that that do not contribute to significant larval or adult mortality.

While butterfly species may be able to locate a reconstructed site, previous research suggests the persistence of butterfly populations can be dependent on the frequency and intensity of management techniques. Several studies have suggested burning can be detrimental to butterfly populations (Swengel and Swengel 2001; Vogel et al. 2007; Vogel et al. 2010; Swengel et al. 2011), while Panzer and Schwartz (2000) did not find negative effects at the scale of their study. In the case of an actively managed agrofuel site, haying would be common practice, and it has been shown to reduce butterfly abundance (Dover et al. 2010), though is suggested be less detrimental to butterfly richness and density than burning (Swengel and Swengel 2001). Butterfly abundance, richness, and diversity significantly decreased in 2011, the year of the first burn. The burn year of 2014 experienced significant flooding, and in this year butterfly richness increased significantly while abundance and diversity remained stable compared to 2013. This leads me to believe that many butterflies using the agrofuel research site are reproducing in the surrounding landscape. Abundance and richness increased significantly in 2012, a year during which the Midwest experienced a significant drought and the site was hayed for the first time. Drought has been reported to decrease (Pollard et al. 1997) the abundance of adult butterflies and create unpredictable population shifts in others (Ehrlich et al. 1980). Drought effects were not apparent but could have produced a carry-over effect (Ehrlich et al. 1980) that contributed to the steep decline in butterfly abundance, richness, and diversity in 2013. Of all six species analyzed, *P. tharos* was the only species that exhibited characteristic fire sensitivity by decreasing in burn years and increasing in abundance with time since the 2011 burn, though management effects are confounded with flood effects in 2013 and 2014. My research suggests that the effects of yearly management on the butterfly community on site are confounded with natural variation in environmental conditions and butterfly populations (Ehrlich et al. 1980), making it difficult to draw conclusions or make recommendations about how to manage agrofuels for the maintenance of butterfly diversity.

In 2013 and 2014, the site experienced significant early summer flooding that completely inundated many of the research fields with several feet of water. The clay loam was extensively flooded; the loam moderately flooded; and the sandy loam experienced little to no flooding. I found that soil drainage class was a strong predictor of floral abundance and richness, and consequently butterfly abundance, during the flood-impacted years of 2013-2014. Few studies have assessed the survival or flowering of prairie forbs in response to natural flood events. Insausti et al. (1999) reported a decrease in aboveground biomass of forbs in heavily flooded grasslands, and McIndoe et al. (2008) recorded a decrease in mean floristic quality assessment values in intermittently flooded prairies. Many studies have reported decreases in abundance of immature life stages of butterflies to flood events (Joy and Pullin 1997; Nichols and Pullin 2003; Severns 2011). Alternatively Kajzer-Bonk et al. (2013) described a species that may be resistant to flooding in their larval stage. Fiedler and Truxa (2012) failed to detect differences in moth species richness between flooded and not flooded habitats due to influx of species from the surrounding environment. However, I was unable to find any literature describing adult butterfly response to flooding events, or studies linking butterfly abundance to the dynamics in a floral community resulting from a flood event. My results suggest the importance of designing seed mixes that contain flood tolerant species for agrofuel crops that would be established on floodplains. *Tradescantia bracteata, Eryngium yuccifolium, Baptisia leucantha, and Silphium laccinatum* flowered after flood events on the extensively flooded clay loam, and could be good candidates. Known riparian, wetland, or flood tolerant plant species could also be good choices for agrofuel seed mixes, especially if they are known larval host plants of native butterflies, as their life histories could be better adapted to periodical flooding. Further research is needed to determine what species would be best suited for agrofuel production in floodplains, and how butterfly and floral communities respond to flood events in reconstructed prairies.

I found significant effects of soil type on butterfly abundance and community composition; sown floral abundance, richness, and diversity; and unsown floral abundance and diversity during the 2010-2012 growing seasons. Butterflies were ~1.1 times more abundant on sandy loam than clay loam, and ~1.2 times more abundant on clay loam than loam. *Everes comyntas,* which represented ~27% of all individuals encountered, was ~2 times more abundant on sandy loam than both loam and clay loam. This species can use *Desmodium canadense* as a larval host plant, which was ~1.8 times more abundant on sandy loam than loam, and ~3.1 times more abundant on sandy loam than clay loam. The abundance of *E. comyntas* on sandy loam can be likely attributed to the abundance of *D. canadense* in 2010-2012 (Myers et al. 2012). Sown flowers were ~ 1.1 times more abundant on the clay loam than loam, and loam than sandy loam. *Vanessa atalanta* was ~1.2 times more abundant on clay loam than loam, and ~3. 67 times more abundant on loam than sandy loam. Greater than 90% of *V. atalanta* individuals observed from 2010-2012 were encountered in 2010, and their soil preference was attributed to an increased abundance of *H. helianthoides* on the more poorly drained soils in 2010 (Myers et al. 2012). In the drought year of 2012, significant differences between the floral richness of Prairie16 and Prairie32 were not apparent on sandy loam, and could be due to its low water holding capacity relative to loam and clay loam. Results suggest that edaphic conditions structured floral and butterfly communities at an agrofuel research site with uniform management and land-use history. Large scale production of agrofuel crops would likely result in similar seed mixes being established over a variety of soil types, which could create a landscape containing heterogeneous habitats suitable for a variety of butterfly species.

My research demonstrates that the floral abundance, richness, and diversity of prairie agrofuel crops were strong predictors of butterfly abundance, richness, and diversity. I found significant variation in butterfly abundance, richness, diversity and community composition when comparing the forb-rich crops to the grass crops.

However, butterfly richness and diversity did not respond to elevated sown forb abundance and diversity in Prairie32 compared to Prairie16. Of all butterflies encountered during the 5 year study, ~93% were habitat generalists, and as the site matured, significant changes in the proportion of habitat specialists found on site were not apparent. Annual butterfly community responses to agrofuel crop management were highly variable and provided little insight into how to best manage agrofuel crops for butterfly diversity. This was partially due to the influence of drought in 2012 and early summer flooding in 2013 and 2014. Flooding frequency and duration was a strong predictor of sown floral abundance, richness and diversity; which in turn influenced butterfly abundance.

In conclusion, my research suggests that the widespread adoption of diverse assemblages of native prairie plants as agrofuel crops would provide higher quality habitat for butterflies than native, grass-only agrofuel feedstocks. By adapting seed mixes to local soil and hydrologic conditions, as well as larval host plant requirements, land managers can provide habitat for target butterfly species while producing a harvestable crop.

Prairie32 failed to attract a greater abundance, richness, or diversity of butterflies than Prairie16, and more research is needed to determine which flower species were preferred by butterflies so that diverse and cost effective agrofuel seed mixes can be designed that contribute to the maintenance of butterfly biodiversity in the Midwest.

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APPENDIX

ADDITIONAL INFORMATION

Table A1. Seeding rates for four native prairie agrofuel crops in Black Hawk County, Iowa, USA.

Species (common name)	Seeding Rate (seeds m^{-2})			
	Switchgrass1	Grass	Biomass	Prairie
Panicum virgatum (switchgrass)	561	86	43	32
Andropogon gerardii (big bluestem)		151	151	135
Bouteloua curtipendula (side-oats grama)		86	43	32
Schizachyrium scoparium (little bluestem)		151	151	135
Sorghastrum nutans (indiangrass)		86	43	32
Agropyron smithii (western wheatgrass)			43	32
Elymus canadensis (Canada wildrye)			43	32
Elymus virginicus (Virginia wildrye)			43	32
Astragalus canadensis (milk vetch)			38	16
Desmodium canadense (showy tick trefoil)			38	16
Heliopsis helianthoides (ox-eye sunflower)			38	16
Lespedeza capitata (round-headed bush clover)			38	16
Solidago rigida (stiff goldenrod)			38	16
Ratibida pinnata (yellow coneflower)			38	16
Helianthus grosseserratus (saw-tooth sunflower)			38	16
Silphium laciniatum (compass plant)			3	3
Carex bicknellii (copper-shoulder oval sedge)				32
Carex brevior (plains oval sedge)				32
Carex gravida (long-awned bracted sedge)				32
Sporobolus asper (tall dropseed)				32
Amorpha canescens (leadplant)				16
Artemisia ludoviciana (prairie sage)				16
Symphyotrichum laeve (smooth blue aster)				16
Symphyotrichum novae-angliae (New England				16
Baptisia leucantha (white wild indigo)				1
Dalea purpurea (purple prairie clover)				16
Echinacea pallida (pale purple coneflower)				16
Erynigium yuccifolium (rattlesnake master)				16
Monarda fistulosa (wild bergamot)				16
Phlox pilosa (prairie phlox)				3
Tradescantia bracteata (prairie spiderwort)				16
Zizia aurea (golden alexanders)				16

Table A2. Generalized linear mixed model post-hoc tests comparing butterfly average abundance, richness and and Shannon's diversity index within agrofuel crop, soil type and year over the 2010-2014 sampling seasons

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Abundance							
crop 1 - 2	-0.1	0.044	36.0	-1.16	-0.141	0.038	0.3
crop 1 - 3	-0.7	0.044	36.0	-16.46	-0.819	-0.639	<0.001
crop 1 - 4	-0.7	0.044	36.0	-16.48	-0.819	-0.640	<0.001
crop 2 - 3	-0.7	0.044	36.0	-15.30	-0.767	-0.588	<0.001
crop 2 - 4	-0.7	0.044	36.0	-15.31	-0.768	-0.588	<0.001
crop 3 - 4	0.0	0.044	36.0	-0.01	-0.090	0.089	1.0
soil 1 - 2	0.2	0.038	36.0	5.13	0.119	0.274	<0.001
soil 1 - 3	0.2	0.038	36.0	4.49	0.095	0.250	<0.001
soil 2 - 3	0.0	0.038	36.0	-0.63	-0.102	0.054	0.5
year 1 - 2	0.7	0.041	144.0	17.54	0.636	0.798	<0.001
year 1 - 3	0.5	0.041	144.0	12.74	0.440	0.602	<0.001
year 1 - 4	0.9	0.041	144.0	22.16	0.825	0.987	<0.001
year 1 - 5	0.9	0.041	144.0	22.59	0.843	1.005	<0.001
year 2 - 3	-0.2	0.041	144.0	-4.79	-0.277	-0.115	<0.001
year 2 - 4	0.2	0.041	144.0	4.62	0.108	0.267	<0.001
year 2 - 5	0.2	0.041	144.0	5.06	0.126	0.288	<0.001
year 3 - 4	0.4	0.041	144.0	9.42	0.304	0.466	<0.001
year 3 - 5	0.4	0.041	144.0	9.85	0.322	0.484	<0.001
year 4 - 5	0.0	0.041	144.0	0.43	-0.063	0.099	0.7
Richness							
crop 1 - 2	-0.4	0.294	36.0	-1.36	-0.997	0.197	0.183
crop 1 - 3	-2.8	0.294	36.0	-9.51	-3.397	-2.203	<0.001
crop 1 - 4	-2.7	0.294	36.0	-9.17	-3.297	-2.103	<0.001
crop 2 - 3	-2.4	0.294	36.0	-8.15	-2.997	-1.803	<0.001
crop 2 - 4	-2.3	0.294	36.0	-7.81	-2.897	-1.703	<0.001
crop 3 - 4	0.1	0.294	36.0	0.34	-0.497	0.697	0.736
soil 1 - 2	0.1	0.255	36.0	0.44	-0.405	0.630	0.662
soil 1 - 3	-0.2	0.255	36.0	-0.69	-0.692	0.342	0.497
soil 2 - 3	-0.3	0.255	36.0	-1.13	-0.805	0.230	0.267
year 1 - 2	3.6	0.304	144.0	11.79	2.982	4.184	<0.001
year 1 - 3	2.4	0.304	144.0	7.81	1.774	2.976	<0.001
year 1 - 4	4.6	0.304	144.0	15.08	3.982	5.184	<0.001
year 1 - 5	3.9	0.304	144.0	12.75	3.274	4.476	<0.001
year 2 - 3	-1.2	0.304	144.0	-3.97	-1.809	-0.607	<0.001
year 2 - 4	1.0	0.304	144.0	3.29	0.399	1.601	0.001

(table continues)

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
year 2 - 5	0.3	0.304	144.0	0.96	-0.309	0.893	0.339
year 3 - 4	2.2	0.304	144.0	7.26	1.607	2.809	<0.001
year 3 - 5	1.5	0.304	144.0	4.93	0.899	2.101	<0.001
year 4 - 5	-0.7	0.304	144.0	-2.33	-1.309	-0.107	0.021
Diversity							
crop 1 - 2	-0.1	0.070	180.0	-1.57	-0.246	0.028	0.118
crop 1 - 3	-0.3	0.070	180.0	-4.28	-0.435	-0.160	<0.001
crop 1 - 4	-0.3	0.070	180.0	-3.66	-0.392	-0.117	<0.001
crop 2 - 3	-0.2	0.070	180.0	-2.71	-0.325	-0.051	0.007
crop 2 - 4	-0.1	0.070	180.0	-2.09	-0.283	-0.008	0.038
crop 3 - 4	0.0	0.070	180.0	0.62	-0.094	0.180	0.538
soil 1 - 2	-0.1	0.060	180.0	-1.58	-0.214	0.024	0.115
soil 1 - 3	-0.2	0.060	180.0	-2.52	-0.271	-0.033	0.012
soil 2 - 3	-0.1	0.060	180.0	-0.94	-0.176	0.062	0.347
year 1 - 2	0.3	0.078	180.0	4.22	0.174	0.481	<0.001
year 1 - 3	0.2	0.078	180.0	2.35	0.029	0.336	0.020
year 1 - 4	0.5	0.078	180.0	7.02	0.392	0.699	<0.001
year 1 - 5	0.4	0.078	180.0	4.83	0.222	0.529	<0.001
year 2 - 3	-0.1	0.078	180.0	-1.87	-0.299	0.008	0.063
year 2 - 4	0.2	0.078	180.0	2.80	0.064	0.371	0.006
year 2 - 5	0.0	0.078	180.0	0.62	-0.105	0.201	0.538
year 3 - 4	0.4	0.078	180.0	4.67	0.210	0.516	<0.001
year 3 - 5	0.2	0.078	180.0	2.49	0.040	0.347	0.014
year 4 - 5	-0.2	0.078	180.0	-2.19	-0.323	-0.017	0.030

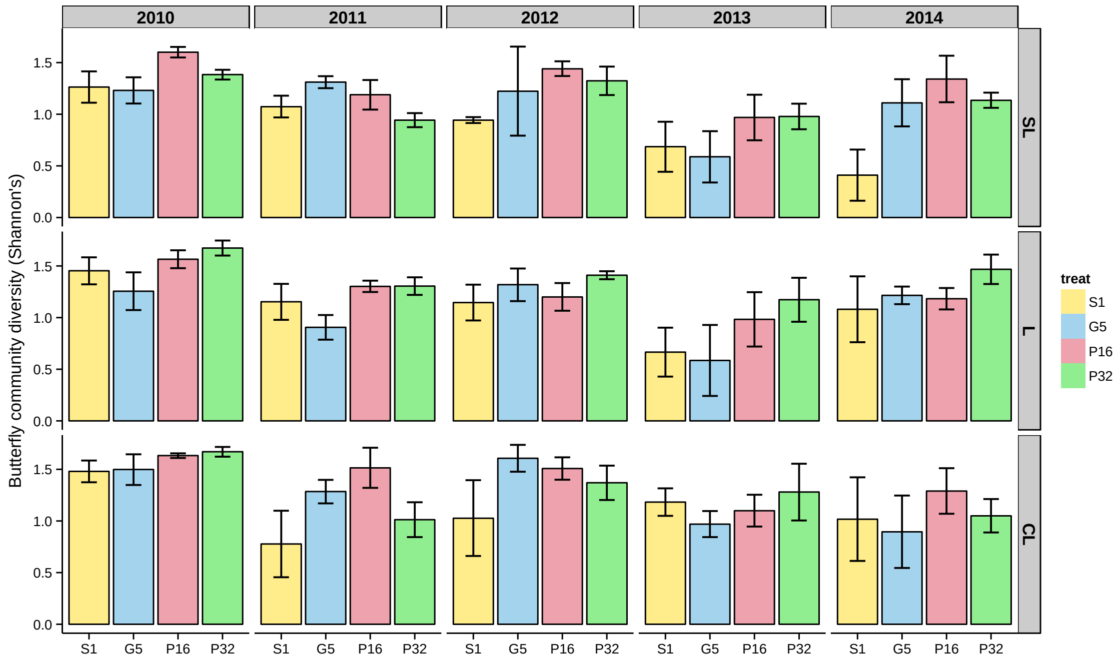


Figure A1. Butterfly community diversity (Shannon's) by agrofuel crop, year and soil type (2010-2014)

Table A3. Generalized linear mixed model post-hoc tests outputs comparing butterfly average abundance, species richness and community diversity (Shannon's) within agrofuel crop, soil type and year (2010-2012).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average Abundance							
crop 1 - 2	-0.1	0.049	36.0	-1.74	-0.182	0.014	0.091
crop 1 - 3	-0.9	0.049	36.0	-18.66	-1.002	-0.806	<0.001
crop 1 - 4	-0.9	0.049	36.0	-18.08	-0.974	-0.778	<0.001
crop 2 - 3	-0.8	0.049	36.0	-16.92	-0.918	-0.722	<0.001
crop 2 - 4	-0.8	0.049	36.0	-16.34	-0.890	-0.694	<0.001
crop 3 - 4	0.0	0.049	36.0	0.58	-0.070	0.126	0.569
soil 1 - 2	0.2	0.042	36.0	5.21	0.134	0.304	<0.001
soil 1 - 3	0.1	0.042	36.0	2.38	0.015	0.185	0.023
soil 2 - 3	-0.1	0.042	36.0	-2.84	-0.204	-0.034	0.007
year 1 - 2	0.7	0.039	72.0	18.33	0.639	0.795	<0.001
year 1 - 3	0.5	0.039	72.0	13.32	0.443	0.599	<0.001
year 2 - 3	-0.2	0.039	72.0	-5.01	-0.274	-0.118	<0.001
Richness							
crop 1 - 2	-0.7	0.433	36.0	-1.54	-1.544	0.211	0.1
crop 1 - 3	-3.6	0.433	36.0	-8.22	-4.433	-2.678	<0.001
crop 1 - 4	-3.2	0.433	36.0	-7.45	-4.100	-2.345	<0.001
crop 2 - 3	-2.9	0.433	36.0	-6.68	-3.767	-2.011	<0.001
crop 2 - 4	-2.6	0.433	36.0	-5.91	-3.433	-1.678	<0.001
crop 3 - 4	0.3	0.433	36.0	0.77	-0.544	1.211	0.4
soil 1 - 2	0.2	0.375	36.0	0.56	-0.552	0.968	0.6
soil 1 - 3	-0.4	0.375	36.0	-1.00	-1.135	0.385	0.3
soil 2 - 3	-0.6	0.375	36.0	-1.56	-1.343	0.177	0.1
year 1 - 2	3.6	0.263	72.0	13.65	3.060	4.107	<0.001
year 1 - 3	2.4	0.263	72.0	9.04	1.851	2.898	<0.001
year 2 - 3	-1.2	0.263	72.0	-4.60	-1.732	-0.685	<0.001
Shannon's Diversity Index							
crop 1 - 2	-0.1	0.085	36.0	-1.72	-0.318	0.026	0.093
crop 1 - 3	-0.3	0.085	36.0	-3.45	-0.465	-0.121	0.001
crop 1 - 4	-0.2	0.085	36.0	-2.32	-0.369	-0.025	0.026
crop 2 - 3	-0.1	0.085	36.0	-1.73	-0.319	0.025	0.092
crop 2 - 4	-0.1	0.085	36.0	-0.60	-0.223	0.121	0.552

(table continues)

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
crop 3 - 4	0.1	0.085	36.0	1.13	-0.076	0.268	0.266
soil 1 - 2	-0.1	0.074	36.0	-0.87	-0.213	0.085	0.390
soil 1 - 3	-0.1	0.074	36.0	-1.66	-0.271	0.027	0.105
soil 2 - 3	-0.1	0.074	36.0	-0.79	-0.207	0.091	0.433
year 1 - 2	0.3	0.059	72.0	5.52	0.209	0.446	<0.001
year 1 - 3	0.2	0.059	72.0	3.07	0.064	0.301	0.003
year 2 - 3	-0.1	0.059	72.0	-2.45	-0.264	-0.027	0.017

Table A4. Generalized linear mixed models comparing *Colias eurytheme/philodice* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
Average abundance				
crop	1	0.082	0.327	0.575
soil	2	1.071	4.264	0.031
year	4	20.363	81.083	<0.001
agrofuel crop:soil	2	0.261	1.039	0.374
agrofuel crop:year	4	0.079	0.312	0.869
soil:year	8	0.414	1.649	0.126
agrofuel crop:soil:year	8	0.298	1.186	0.320

Table A5. Generalized linear mixed model post-hoc tests comparing *Colias eurytheme/philodice* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average abundance							
crop 3 - 4	-0.1	0.111	18.0	-0.57	-0.296	0.169	0.575
soil 1 - 2	0.3	0.136	18.0	2.13	0.004	0.574	0.047
soil 1 - 3	0.4	0.136	18.0	2.79	0.094	0.664	0.012
soil 2 - 3	0.1	0.136	18.0	0.66	-0.195	0.375	0.517
year 1 - 2	1.7	0.145	72.0	11.97	1.444	2.021	<0.001
year 1 - 3	1.0	0.145	72.0	6.71	0.683	1.259	<0.001
year 1 - 4	2.1	0.145	72.0	14.72	1.841	2.417	<0.001
year 1 - 5	2.2	0.145	72.0	15.07	1.891	2.468	<0.001
year 2 - 3	-0.8	0.145	72.0	-5.26	-1.050	-0.473	<0.001
year 2 - 4	0.4	0.145	72.0	2.74	0.108	0.685	0.008
year 2 - 5	0.4	0.145	72.0	3.09	0.159	0.736	0.003
year 3 - 4	1.2	0.145	72.0	8.01	0.870	1.446	<0.001
year 3 - 5	1.2	0.145	72.0	8.35	0.920	1.497	<0.001
year 4 - 5	0.1	0.145	72.0	0.35	-0.238	0.339	0.728

Table A6. Generalized linear mixed models comparing *Everes comyntas* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
crop	1	0.030	0.121	0.733
soil	2	10.751	42.738	0
year	4	3.482	13.844	<0.001
agrofuel crop:soil	2	0.084	0.332	0.722
agrofuel crop:year	4	0.139	0.554	0.697
soil:year	8	0.696	2.767	0.010
agrofuel crop:soil:year	8	0.636	2.528	0.018

Table A7. Generalized linear mixed model post-hoc tests comparing *Everes comyntas* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average abundance							
crop 3 - 4	0.0	0.092	18.0	-0.35	-0.225	0.161	0.733
soil 1 - 2	0.7	0.113	18.0	6.33	0.476	0.949	<0.001
soil 1 - 3	1.0	0.113	18.0	9.00	0.776	1.249	<0.001
soil 2 - 3	0.3	0.113	18.0	2.67	0.064	0.536	0.016
year 1 - 2	-0.6	0.145	72.0	-3.90	-0.853	-0.276	<0.001
year 1 - 3	-0.7	0.145	72.0	-4.83	-0.988	-0.411	<0.001
year 1 - 4	-0.2	0.145	72.0	-1.05	-0.440	0.137	0.298
year 1 - 5	0.2	0.145	72.0	1.42	-0.083	0.494	0.160
year 2 - 3	-0.1	0.145	72.0	-0.93	-0.424	0.153	0.353
year 2 - 4	0.4	0.145	72.0	2.85	0.124	0.701	0.006
year 2 - 5	0.8	0.145	72.0	5.32	0.481	1.059	<0.001
year 3 - 4	0.5	0.145	72.0	3.78	0.259	0.837	<0.001
year 3 - 5	0.9	0.145	72.0	6.25	0.617	1.194	<0.001
year 4 - 5	0.4	0.145	72.0	2.47	0.069	0.646	0.016

Table A8. Generalized linear mixed models comparing *Vanessa atalanta* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
crop	1	0.366	2.373	0.127
soil	2	1.649	10.692	<0.001
year	4	28.000	181.524	<0.001
agrofuel crop:soil	2	0.045	0.288	0.750
treat:year	4	0.125	0.807	0.524
soil:year	8	1.339	8.678	<0.001
treat:soil:year	8	0.185	1.198	0.309

Table A9. Generalized linear mixed model post-hoc tests comparing *Vanessa atalanta* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average abundance							
treat 3 - 4	0.1	0.072	90.0	1.54	-0.032	0.253	0.127
soil 1 - 2	-0.3	0.088	90.0	-3.39	-0.472	-0.124	0.001
soil 1 - 3	-0.4	0.088	90.0	-4.42	-0.562	-0.214	<0.001
soil 2 - 3	-0.1	0.088	90.0	-1.02	-0.265	0.085	0.308
year 1 - 2	2.3	0.113	90.0	20.12	2.055	2.506	<0.001
year 1 - 3	2.4	0.113	90.0	20.99	2.155	2.605	<0.001
year 1 - 4	2.6	0.113	90.0	23.37	2.424	2.874	<0.001
year 1 - 5	2.2	0.113	90.0	19.82	2.022	2.473	<0.001
year 2 - 3	0.1	0.113	90.0	0.87	-0.126	0.324	0.384
year 2 - 4	0.4	0.113	90.0	3.25	0.143	0.594	0.002
year 2 - 5	0.0	0.113	90.0	-0.29	-0.259	0.192	0.770
year 3 - 4	0.3	0.113	90.0	2.37	0.044	0.495	0.020
year 3 - 5	-0.1	0.113	90.0	-1.17	-0.358	0.093	0.246
year 4 - 5	-0.4	0.113	90.0	-3.54	-0.627	-0.176	<0.001

Table A10. Generalized linear mixed model post-hoc tests comparing *Danaus plexippus* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
treat	1	0.001	0.002	0.967
soil	2	0.784	2.585	0.103
year	4	9.445	31.163	<0.001
treat:soil	2	0.206	0.679	0.520
treat:year	4	0.308	1.017	0.404
soil:year	8	0.227	0.747	0.650
treat:soil:year	8	0.181	0.596	0.778

Table A11. Generalized linear mixed model post-hoc tests comparing *Danaus plexippus* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average abundance							
treat 3 - 4	0.0	0.117	18.0	0.04	-0.240	0.250	0.97
soil 1 - 2	0.2	0.143	18.0	1.36	-0.106	0.494	0.19
soil 1 - 3	-0.1	0.143	18.0	-0.90	-0.429	0.171	0.38
soil 2 - 3	-0.3	0.143	18.0	-2.26	-0.623	-0.023	0.04
year 1 - 2	1.3	0.159	72.0	8.33	1.008	1.641	<0.001
year 1 - 3	1.3	0.159	72.0	8.23	0.991	1.624	<0.001
year 1 - 4	1.6	0.159	72.0	10.08	1.284	1.918	<0.001
year 1 - 5	1.2	0.159	72.0	7.81	0.924	1.557	<0.001
year 2 - 3	0.0	0.159	72.0	-0.11	-0.334	0.300	0.92
year 2 - 4	0.3	0.159	72.0	1.74	-0.040	0.594	0.09
year 2 - 5	-0.1	0.159	72.0	-0.53	-0.401	0.233	0.60
year 3 - 4	0.3	0.159	72.0	1.85	-0.023	0.611	0.07
year 3 - 5	-0.1	0.159	72.0	-0.42	-0.384	0.250	0.67
year 4 - 5	-0.4	0.159	72.0	-2.27	-0.678	-0.044	0.03

Table A12. Generalized linear mixed models comparing *Phyciodes tharos* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of variation	df	MS	F	*P*
treat	1	0.034	0.226	0.635
soil	2	0.027	0.179	0.837
year	4	4.634	30.702	< 0.001
treat:soil	2	0.691	4.576	0.013
treat:year	4	0.215	1.422	0.231
soil:year	8	0.362	2.397	0.020
treat:soil:year	8	0.340	2.254	0.028

Table A13. Generalized linear mixed model post-hoc tests comparing *Phyciodes tharos* average abundance within agrofuel crop, soil type and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average abundance							
treat 3 - 4	0.0	0.082	90.0	0.41	-0.129	0.196	0.7
soil 1 - 2	0.0	0.100	90.0	0.25	-0.174	0.225	0.8
soil 1 - 3	0.1	0.100	90.0	0.52	-0.147	0.251	0.6
soil 2 - 3	0.0	0.100	90.0	0.26	-0.173	0.226	0.8
year 1 - 2	1.1	0.130	90.0	8.12	0.795	1.309	<0.001
year 1 - 3	0.9	0.130	90.0	6.92	0.638	1.153	<0.001
year 1 - 4	0.4	0.130	90.0	3.46	0.191	0.705	<0.001
year 1 - 5	0.9	0.130	90.0	7.32	0.691	1.206	<0.001
year 2 - 3	-0.2	0.130	90.0	-1.21	-0.414	0.101	0.2
year 2 - 4	-0.6	0.130	90.0	-4.66	-0.861	-0.346	<0.001
year 2 - 5	-0.1	0.130	90.0	-0.80	-0.361	0.154	0.4
year 3 - 4	-0.4	0.130	90.0	-3.45	-0.705	-0.190	<0.001
year 3 - 5	0.1	0.130	90.0	0.41	-0.204	0.310	0.7
year 4 - 5	0.5	0.130	90.0	3.86	0.243	0.758	<0.001

Table A14. Generalized linear mixed model post-hoc tests comparing sown floral average abundance, species richness and community diversity (Shannon's) within agrofuel crop, soil type and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average Abundance							
crop 3 - 4	0.1	0.159	24.0	0.75	-0.208	0.446	0.5
soil 1 - 2	-0.2	0.194	24.0	-1.27	-0.647	0.155	0.2
soil 1 - 3	0.0	0.194	24.0	0.20	-0.363	0.439	0.8
soil 2 - 3	0.3	0.194	24.0	1.46	-0.117	0.685	0.2
year 1 - 2	-0.1	0.159	96.0	-0.76	-0.437	0.194	0.4
year 1 - 3	2.1	0.159	96.0	13.07	1.763	2.394	<0.001
year 1 - 4	2.0	0.159	96.0	12.74	1.710	2.341	<0.001
year 1 - 5	1.9	0.159	96.0	11.94	1.583	2.214	<0.001
year 2 - 3	2.2	0.159	96.0	13.83	1.884	2.515	<0.001
year 2 - 4	2.1	0.159	96.0	13.50	1.831	2.462	<0.001
year 2 - 5	2.0	0.159	96.0	12.70	1.704	2.335	<0.001
year 3 - 4	-0.1	0.159	96.0	-0.33	-0.368	0.263	0.7
year 3 - 5	-0.2	0.159	96.0	-1.13	-0.495	0.136	0.3
year 4 - 5	-0.1	0.159	96.0	-0.80	-0.443	0.188	0.4
Richness							
crop 3 - 4	-5.0	0.258	120.0	-19.61	-5.560	-4.540	<0.001
soil 1 - 2	-1.5	0.15	120.0	-4.84	-2.150	-0.901	<0.001
soil 1 - 3	0.4	0.315	120.0	1.19	-0.250	0.100	0.24
soil 2 - 3	1.9	0.315	120.0	6.02	1.276	2.525	<0.001
year 1 - 2	-0.9	0.407	120.0	-2.15	-1.681	-0.069	0.03
year 1 - 3	1.4	0.407	120.0	3.48	0.611	2.223	<0.001
year 1 - 4	2.2	0.407	120.0	5.32	1.361	2.973	<0.001
year 1 - 5	1.4	0.407	120.0	3.48	0.611	2.223	<0.001
year 2 - 3	2.3	0.407	120.0	5.63	1.486	3.098	<0.001
year 2 - 4	3.0	0.407	120.0	7.47	2.236	3.848	<0.001
year 2 - 5	2.3	0.407	120.0	5.63	1.486	3.098	<0.001
year 3 - 4	0.8	0.407	120.0	1.84	-0.056	1.556	0.07
year 3 - 5	0.0	0.407	120.0	0.00	-0.806	0.806	1.00
year 4 - 5	-0.8	0.407	120.0	-1.84	-1.556	0.056	0.07
Diversity							
crop 3 - 4	-0.5	0.057	24.0	-8.48	-0.604	-0.368	<0.001
soil 1 - 2	-0.2	0.070	24.0	-3.42	-0.385	-0.095	0.002
soil 1 - 3	0.0	0.070	24.0	0.10	-0.138	0.152	0.922

(table continues)

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
soil 2 - 3	0.2	0.070	24.0	3.52	0.102	0.392	0.002
year 1 - 2	0.0	0.087	96.0	0.50	-0.131	0.218	0.621
year 1 - 3	-0.1	0.087	96.0	-1.32	-0.289	0.059	0.192
year 1 - 4	0.0	0.087	96.0	0.03	-0.171	0.177	0.974
year 1 - 5	0.0	0.087	96.0	0.35	-0.143	0.205	0.724
year 2 - 3	-0.2	0.087	96.0	-1.81	-0.333	0.015	0.073
year 2 - 4	0.0	0.087	96.0	-0.46	-0.215	0.133	0.644
year 2 - 5	0.0	0.087	96.0	-0.14	-0.186	0.162	0.887
year 3 - 4	0.1	0.087	96.0	1.35	-0.056	0.292	0.181
year 3 - 5	0.1	0.087	96.0	1.67	-0.028	0.320	0.098
year 4 - 5	0.0	0.087	96.0	0.32	-0.146	0.202	0.748

Table A15. Generalized linear mixed model post-hoc tests comparing sown (P16/32) floral abundance, richness and community diversity (Shannon's) within agrofuel crop, soil and year (2010-2012)

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Abundance							
crop 3 - 4	0.1	0.092	24.0	1.14	-0.085	0.294	0.267
soil 1 - 2	-0.3	0.112	24.0	-2.96	-0.565	-0.101	0.007
soil 1 - 3	-0.7	0.112	24.0	-5.94	-0.899	-0.435	<0.001
soil 2 - 3	-0.3	0.112	24.0	-2.98	-0.567	-0.103	0.007
year 1 - 2	-0.1	0.089	48.0	-1.36	-0.301	0.059	0.182
year 1 - 3	2.1	0.089	48.0	23.24	1.898	2.258	<0.001
year 2 - 3	2.2	0.089	48.0	24.59	2.012	2.379	<0.001
Richness							
crop 3 - 4	-5.4	0.225	72.0	-23.80	-5.810	-4.912	<0.001
soil 1 - 2	-1.9	0.276	72.0	-6.95	-2.467	-1.367	<0.001
soil 1 - 3	-1.7	0.276	72.0	-6.04	-2.217	-1.117	<0.001
soil 2 - 3	0.2	0.276	72.0	0.91	-0.300	0.800	0.368
year 1 - 2	-0.9	0.276	72.0	-3.17	-1.425	-0.325	0.002
year 1 - 3	1.4	0.276	72.0	5.14	0.867	1.967	<0.001
year 2 - 3	2.3	0.276	72.0	8.31	1.742	2.842	<0.001
Diversity							
crop 3 - 4	-0.4	0.067	24.0	-5.50	-0.505	-0.230	<0.001
soil 1 - 2	-0.3	0.082	24.0	-3.82	-0.481	-0.143	<0.001
soil 1 - 3	-0.3	0.082	24.0	-3.71	-0.472	-0.135	0.001
soil 2 - 3	0.0	0.082	24.0	0.11	-0.160	0.177	0.917
year 1 - 2	0.0	0.067	48.0	0.66	-0.090	0.177	0.514
year 1 - 3	-0.1	0.067	48.0	-1.74	-0.248	0.018	0.088
year 2 - 3	-0.2	0.067	48.0	-2.40	-0.292	-0.026	0.020

Table A16. Generalized linear mixed model post-hoc tests comparing unsown floral average abundance, species richness and community diversity (Shannon's) within agrofuel crop, soil and year (2010-2014).

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Average Abundance							
crop 1 - 2	0.6	0.156	48.0	3.52	0.236	0.865	0.001
crop 1 - 3	-0.2	0.156	48.0	-1.24	-0.508	0.120	0.221
crop 1 - 4	0.2	0.156	48.0	1.11	-0.141	0.488	0.274
crop 2 - 3	-0.7	0.156	48.0	-4.76	-1.059	-0.430	<0.001
crop 2 - 4	-0.4	0.156	48.0	-2.41	-0.691	-0.063	0.020
crop 3 - 4	0.4	0.156	48.0	2.35	0.053	0.682	0.023
soil 1 - 2	-0.4	0.135	48.0	-2.94	-0.670	-0.126	0.005
soil 1 - 3	-0.2	0.135	48.0	-1.76	-0.511	0.033	0.084
soil 2 - 3	0.2	0.135	48.0	1.18	-0.113	0.431	0.245
year 1 - 2	1.7	0.117	192.0	14.76	1.498	1.961	<0.001
year 1 - 3	1.9	0.117	192.0	16.63	1.717	2.179	<0.001
year 1 - 4	1.7	0.117	192.0	14.88	1.513	1.975	<0.001
year 1 - 5	1.8	0.117	192.0	15.00	1.527	1.989	<0.001
year 2 - 3	0.2	0.117	192.0	1.87	-0.012	0.450	0.064
year 2 - 4	0.0	0.117	192.0	0.12	-0.217	0.245	0.903
year 2 - 5	0.0	0.117	192.0	0.24	-0.203	0.260	0.809
year 3 - 4	-0.2	0.117	192.0	-1.74	-0.435	0.027	0.083
year 3 - 5	-0.2	0.117	192.0	-1.62	-0.421	0.041	0.106
year 4 - 5	0.0	0.117	192.0	0.12	-0.217	0.245	0.904
Richness							
crop 1 - 2	0.4	0.321	48.0	1.24	-0.246	1.046	0.219
crop 1 - 3	-1.1	0.321	48.0	-3.58	-1.796	-0.504	<0.001
crop 1 - 4	-0.6	0.321	48.0	-1.71	-1.196	0.096	0.093
crop 2 - 3	-1.6	0.321	48.0	-4.82	-2.196	-0.904	<0.001
crop 2 - 4	-1.0	0.321	48.0	-2.96	-1.596	-0.304	0.005
crop 3 - 4	0.6	0.321	48.0	1.87	-0.046	1.246	0.068
soil 1 - 2	-0.4	0.278	48.0	-1.62	-1.010	0.110	0.112
soil 1 - 3	-0.4	0.278	48.0	-1.48	-0.972	0.147	0.145
soil 2 - 3	0.0	0.278	48.0	0.13	-0.522	0.597	0.893
year 1 - 2	4.0	0.202	192.0	19.74	3.582	4.377	<0.001
year 1 - 3	4.2	0.202	192.0	21.08	3.852	4.648	<0.001
year 1 - 4	4.3	0.202	192.0	21.18	3.873	4.669	<0.001
year 1 - 5	4.2	0.202	192.0	21.08	3.852	4.648	<0.001
year 2 - 3	0.3	0.202	192.0	1.34	-0.127	0.669	0.181

(table continues)

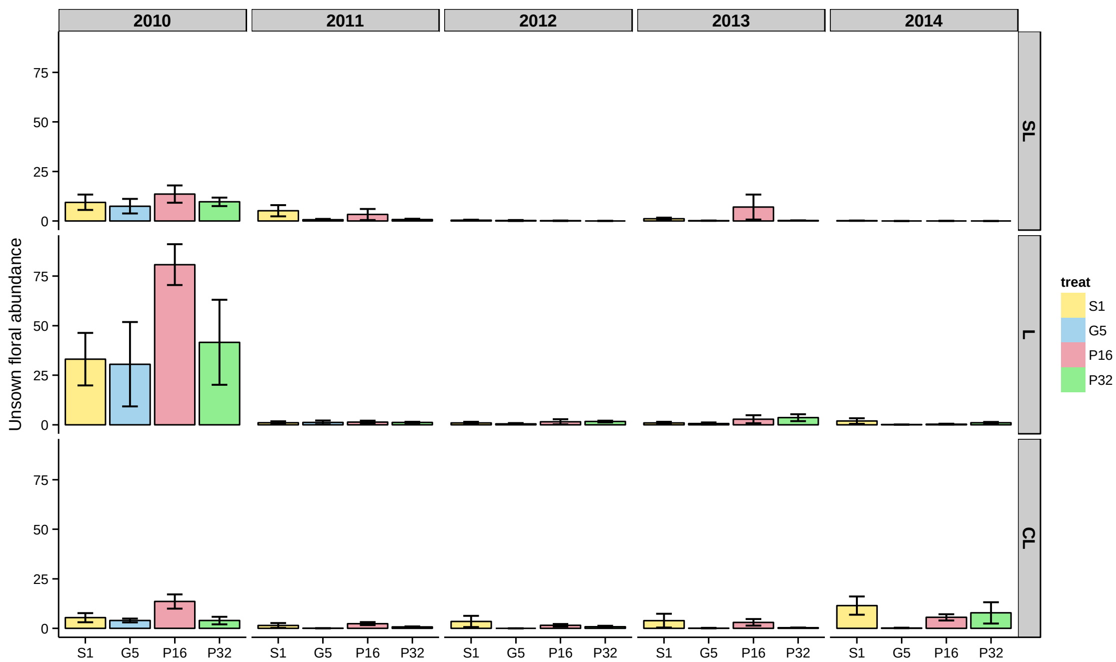
Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
year 2 - 4	0.3	0.202	192.0	1.45	-0.106	0.689	0.150
year 2 - 5	0.3	0.202	192.0	1.34	-0.127	0.669	0.181
year 3 - 4	0.0	0.202	192.0	0.10	-0.377	0.419	0.918
year 3 - 5	0.0	0.202	192.0	0.00	-0.398	0.398	1.000
year 4 - 5	0.0	0.202	192.0	-0.10	-0.419	0.377	0.918
Diversity							
crop 1 - 2	0.1	0.069	48.0	0.95	-0.073	0.204	0.345
crop 1 - 3	-0.2	0.069	48.0	-2.87	-0.336	-0.059	0.006
crop 1 - 4	-0.2	0.069	48.0	-2.26	-0.294	-0.017	0.028
crop 2 - 3	-0.3	0.069	48.0	-3.82	-0.402	-0.125	<0.001
crop 2 - 4	-0.2	0.069	48.0	-3.22	-0.360	-0.083	0.002
crop 3 - 4	0.0	0.069	48.0	0.61	-0.097	0.180	0.546
soil 1 - 2	0.0	0.060	48.0	-0.09	-0.126	0.114	0.925
soil 1 - 3	-0.1	0.060	48.0	-1.62	-0.216	0.024	0.113
soil 2 - 3	-0.1	0.060	48.0	-1.52	-0.211	0.029	0.135
year 1 - 2	0.6	0.060	192.0	10.50	0.506	0.740	<0.001
year 1 - 3	0.7	0.060	192.0	11.16	0.545	0.779	<0.001
year 1 - 4	0.8	0.060	192.0	12.83	0.644	0.878	<0.001
year 1 - 5	0.7	0.060	192.0	11.16	0.545	0.78 0	<0.001
year 2 - 3	0.0	0.060	192.0	0.65	-0.078	0.156	0.513
year 2 - 4	0.1	0.060	192.0	2.33	0.021	0.255	0.021
year 2 - 5	0.0	0.060	192.0	0.66	-0.078	0.156	0.509
year 3 - 4	0.1	0.060	192.0	1.67	-0.018	0.216	0.096
year 3 - 5	0.0	0.060	192.0	0.01	-0.117	0.118	0.994
year 4 - 5	-0.1	0.060	192.0	-1.67	-0.216	0.018	0.097

Table A17. Generalized linear mixed model post-hoc tests comparing unsown floral abundance, richness and community diversity (Shannon's) within agrofuel crop, soil and year (2010-2012)

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
Abundance							
crop 1 - 2	0.4	0.175	48.0	2.44	0.076	0.779	0.018
crop 1 - 3	-0.3	0.175	48.0	-1.80	-0.666	0.038	0.079
crop 1 - 4	0.1	0.175	48.0	0.81	-0.211	0.493	0.423
crop 2 - 3	-0.7	0.175	48.0	-4.24	-1.093	-0.390	<0.001
crop 2 - 4	-0.3	0.175	48.0	-1.64	-0.638	0.066	0.108
crop 3 - 4	0.5	0.175	48.0	2.60	0.104	0.807	0.012
soil 1 - 2	-0.5	0.152	48.0	-3.13	-0.779	-0.170	0.003
soil 1 - 3	0.1	0.152	48.0	0.39	-0.245	0.364	0.697
soil 2 - 3	0.5	0.152	48.0	3.52	0.229	0.838	0.001
year 1 - 2	1.7	0.129	96.0	13.45	1.474	1.985	<0.001
year 1 - 3	1.9	0.129	96.0	15.15	1.693	2.203	<0.001
year 2 - 3	0.2	0.129	96.0	1.70	-0.037	0.474	0.092
Richness							
crop 1 - 2	0.1	0.414	48.0	0.34	-0.694	0.972	0.739
crop 1 - 3	-1.8	0.414	48.0	-4.22	-2.583	-0.917	<0.001
crop 1 - 4	-1.0	0.414	48.0	-2.48	-1.861	-0.195	0.017
crop 2 - 3	-1.9	0.414	48.0	-4.56	-2.722	-1.056	<0.001
crop 2 - 4	-1.2	0.414	48.0	-2.82	-2.000	-0.334	0.007
crop 3 - 4	0.7	0.414	48.0	1.74	-0.111	1.555	0.088
soil 1 - 2	-0.4	0.359	48.0	-1.10	-1.117	0.325	0.275
soil 1 - 3	-0.4	0.359	48.0	-1.22	-1.159	0.284	0.229
soil 2 - 3	0.0	0.359	48.0	-0.12	-0.763	0.680	0.908
year 1 - 2	4.0	0.222	96.0	17.93	3.539	4.420	<0.001
year 1 - 3	4.2	0.222	96.0	19.15	3.809	4.691	<0.001
year 2 - 3	0.3	0.222	96.0	1.22	-0.170	0.711	0.225
Diversity							
crop 1 - 2	0.0	0.082	48.0	-0.28	-0.187	0.142	0.782
crop 1 - 3	-0.3	0.082	48.0	-3.75	-0.472	-0.143	<0.001
crop 1 - 4	-0.3	0.082	48.0	-3.89	-0.483	-0.154	<0.001
crop 2 - 3	-0.3	0.082	48.0	-3.47	-0.449	-0.120	0.001
crop 2 - 4	-0.3	0.082	48.0	-3.61	-0.460	-0.131	<0.001
crop 3 - 4	0.0	0.082	48.0	-0.13	-0.176	0.154	0.894
soil 1 - 2	0.1	0.071	48.0	1.37	-0.045	0.234	0.176
soil 1 - 3	-0.1	0.071	48.0	-1.77	-0.268	0.017	0.083

(table continues)

Source of Variation	Est.	SE	DF	t-value	LCI	UCI	*P*
soil 2 - 3	-0.2	0.071	48.0	-3.15	-0.366	-0.080	0.003
year 1 - 2	0.6	0.064	96.0	9.75	0.496	0.750	<0.001
year 1 - 3	0.7	0.064	96.0	10.36	0.535	0.789	<0.001
year 2 - 3	0.0	0.064	96.0	0.61	-0.088	0.166	0.545



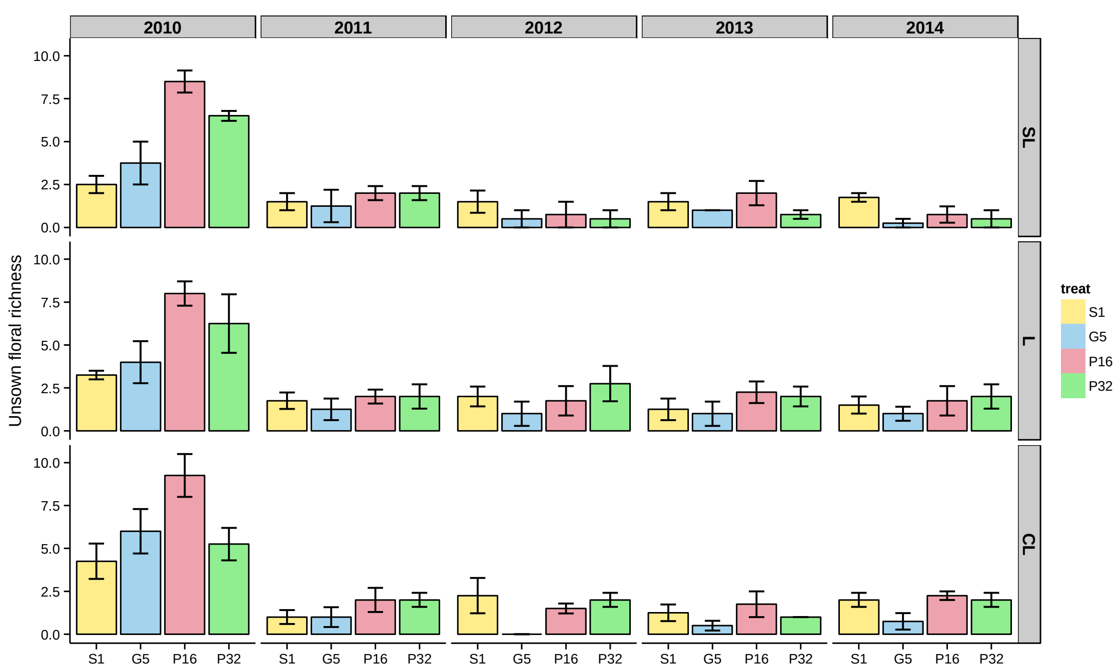


Figure A2 Unsown floral average abundance (top) and species richness (bottom) by agrofuel crop, year and soil type (2010-2014)

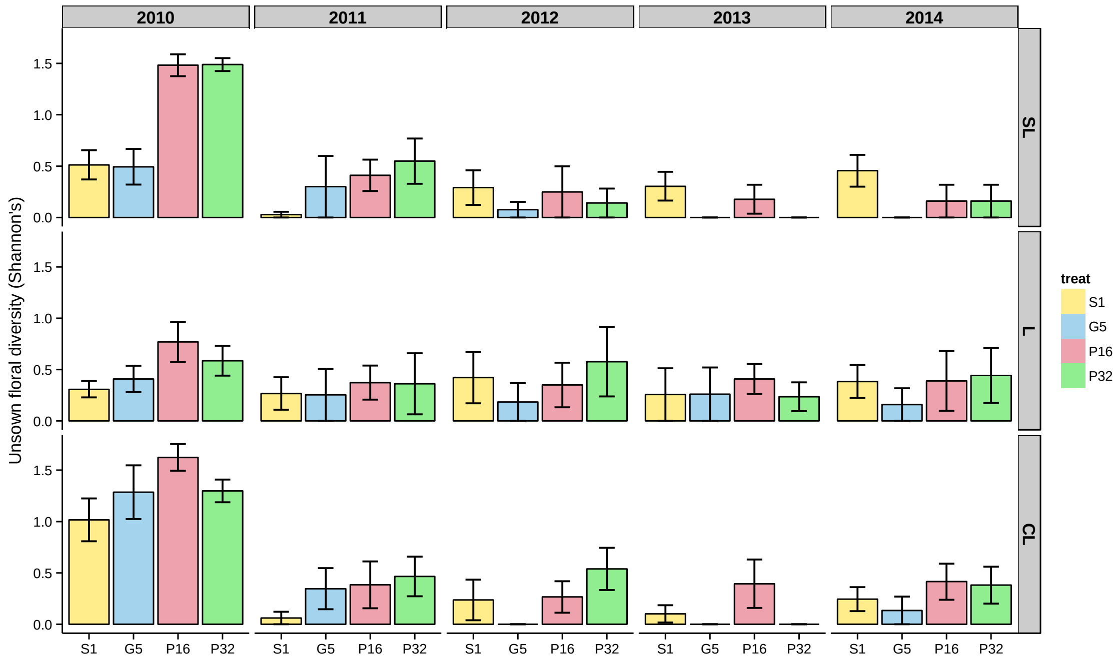


Figure A3 Unsown floral diversity (Shannon's by agrofuel crop, year and soil type (2010-2014)