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Land-use legacies and present fire regimes interact to mediate herbivory by altering the neighboring plant community

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Past and present human activities, such as historic agriculture and fire suppression, are widespread and can create depauperate plant communities. Although many studies show that herbivory on focal plants depends on the density of herbivores or the composition of the surrounding plant community, it is unclear whether anthropogenic changes to plant communities alter herbivory. We tested the hypothesis that human activities that alter the plant community lead to subsequent changes in herbivory. At 20 sites distributed across 80 300 hectares, we conducted a field experiment that manipulated insect herbivore access (full exclosures and pseudo-exclosures) to four focal plant species in longleaf pine woodlands with different land-use histories (post-agricultural sites or non-agricultural sites) and degrees of fire frequency (frequent and infrequent). Plant cover, particularly herbaceous cover, was lower in post-agricultural and fire suppressed woodlands. Density of the dominant insect herbivore at our site (grasshoppers) was positively related to plant cover. Herbivore access reduced biomass of the palatable forb *Solidago odora* in frequently burned post-agricultural sites and in infrequently burned non-agricultural woodlands and increased mortality of another forb (*Pityopsis graminifolia*), but did not affect two other less palatable species (*Schizachyrium scoparium* and *Tephrosia virginiana*). Herbivory on *S. odora* exhibited a hump-shaped response to plant cover, with low herbivory at low and high levels of plant cover. Herbivore density had a weak negative effect on herbivory. These findings suggest that changes in plant cover related to past and present human activities can modify damage rates on focal *S. odora* plants by altering grasshopper foraging behavior rather than by altering local grasshopper density. The resulting changes in herbivory may have the potential to limit natural recovery or restoration efforts by reducing the establishment or performance of palatable plant species.

Human activities affect a large fraction of plant communities across the globe, such as through the introduction of non-native species, habitat fragmentation, historical agriculture, or fire suppression (Wilcove et al. 1998). Herbivores also have important effects on plant population dynamics and community structure in many ecosystems (Crawley 1983). Importantly, the potential exists for human activities to alter the strength of herbivory because human disturbances can directly affect the structure and composition of the plant community, and plant neighbors often alter herbivore attack rates on focal plants (i.e. associational effects, Barbosa et al. 2009). However, although large parts of the biosphere are influenced by both humans and herbivores, it is unclear whether human activities create plant communities that subsequently alter herbivory rates.

Legacies of past agricultural land-use and present fire suppression are two human activities that significantly affect plant communities in ways that could foster changes in herbivory. Agricultural land-use legacies can have persistent effects on plant communities by altering soil conditions and limiting reestablishment of many perennial plant species, resulting in lower diversity and altered community composition for decades to centuries after abandonment (Flinn and Vellend 2005, Vellend et al. 2007). Fire suppression favors growth of woody vegetation relative to herbaceous vegetation and tends to reduce plant diversity (Briggs et al. 2005). Furthermore, many areas that are affected by historic agriculture (Ramankutty and Foley 1999) are in fire-dependent ecosystems that concurrently experience fire suppression (Briggs et al. 2005). Indeed, studies show that both agricultural legacies and fire suppression can interact to alter plant communities (Brudvig and Damschen 2011, Brudvig et al. 2014). Although agricultural legacies and fire suppression cause well-documented, large-scale changes to plant communities, such as changes in productivity or community composition (Briggs et al. 2005, Flinn and Vellend 2005), and these components of plant communities can also mediate herbivore effects (Baraza et al. 2006, Orrock et al. 2008, Caccia et al. 2009), as well as modify herbivore assemblages (Kim and Holt 2012), the degree to which these changes alter the nature of herbivory remains untested.

In this paper, we use a large-scale field study to evaluate how historic and contemporary human disturbances alter the nature of herbivory in longleaf pine *Pinus palustris* woodlands of the southeastern, USA. The longleaf pine ecosystem has a history of intensive agriculture followed more recently by large-scale abandonment (Frost et al. 2006). The legacies of historic agriculture tend to eliminate perennial herbaceous plants and reduce overall vegetation cover and diversity in regenerating longleaf woodlands (Kirkman et al. 2004, Brudvig et al. 2014). Fire suppression can exacerbate the effects of historic agriculture on plant communities by promoting woody plants over herbaceous vegetation (Glitzenstein et al. 2012), resulting in even greater changes to plant community composition (Brudvig and Damschen 2011, Brudvig et al. 2014).

Grasshoppers are common generalist herbivores in longleaf pine woodlands (Knight and Holt 2005, Evans et al. 2012) and grasshoppers are capable of exerting strong top–down effects on plant performance, composition and diversity through selective grazing (Schmitz 2006, Evans et al. 2012, Cease et al. 2012). Furthermore, herbivory by grasshoppers may be altered by past land use or reduced fire frequecy because grasshopper density may be a function of the availability of suitable host plants (Joern and Laws 2013). Alternatively, grasshopper preferences may change as a function of the characteristics of the plant community, such as resource quality (Behmer et al. 2001). Thus, historic agricultural land use and fire suppression could lead to changes in herbivory via two primary mechanisms: 1) changes in total herbivore density or 2) changes in the availability of alternative plant food sources. If agricultural land-use history or fire suppression creates depauperate plant communities that support lower densities of herbivores and if attack rates on palatable focal plants depends on the density of herbivores, this would result in reduced herbivory in habitats that have experienced past agricultural land use or fire suppression (White and Whitham 2000, Rand 2003). On the other hand, herbivory on palatable focal plants could be greater in depauperate plant communities because palatable focal plants are more apparent to herbivores (Atsatt and O'dowd 1976) or because generalist herbivores in depauperate habitats may preferentially target palatable plants to meet particular nutritional needs (Behmer et al. 2001). These changes could lead to herbivores consistently excluding certain species from the plant community, serving to reinforce the maintenance of a particular community state (Parker and Root 1981, Orrock et al. 2010). In the context of the longleaf pine ecosystem, decreased herbivory on focal plants could promote the performance of plants often associated with typical longleaf understory communities. Alternatively, if land-use history and fire suppression increase herbivory on focal plants, herbivory may constrain the re-establishment of plant species of restoration and conservation interest. Increased herbivory rates in depauperate habitats could help to explain the lower cover of herbaceous plants or altered community composition generally found in these habitats (Kirkman et al. 2004, Brudvig and Damschen 2011, Brudvig et al. 2014), although this has not been tested.

Using experimental herbivore exclosures deployed across a landscape with sites varying in land-use history and fire frequency, we evaluated whether past agricultural land use and fire suppression might affect hebivory on focal plants by altering herbivore density or the cover of preferred herbaceous plants. First, we ask 1) does agricultural land-use history or fire suppression alter the plant community or grasshopper density? We also asked: 2) does habitat type (i.e. combinations of land-use history and fire suppression) alter the effect that herbivores have on plant biomass? We then examine the mechanisms whereby land-use history and fire suppression could lead to differences in herbivory by coupling data from our experimental herbivore exclosures with data on grasshopper density and the composition of the neighboring plant community. If herbivory is driven primarily by changes in grasshopper density, we predict herbivory on focal plants will be a function of grasshopper density. If herbivory is primarily affected by changes in herbivore foraging behavior caused by differences in the plants available for consumption, we expect that herbivory will be most strongly explained by the composition of the neighboring plant community, and will not depend upon grasshopper density.

Methods

We conducted this study at the Savannah River Site (SRS), an 80 300 hectare National Environmental Research Park in South Carolina, USA (Fig. 1A). We selected 20 upland longleaf pine *Pinus palustris* woodland sites, stratified across land-use histories ('post-agricultural' woodlands that have a history of cultivation or 'non-agricultural' woodlands that have never been cultivated) and fire frequencies (frequent and infrequent), resulting in five replicates of four habitat types (Fig. 1A, Supplementary material Appendix 1 Fig. A1). The habitat types were on similar soils, interspersed across the landscape and all sites were separated by > 500 m. See Brudvig et al. (2014) for additional details about plant diversity and soil conditions at these sites. To avoid edge effects that may arise at the boundary of habitats with different land-use histories or prescribed burn regimes (Knight and Holt 2005), all sites were buffered by > 50 m of similar habitat (i.e. same land-use and fire history). Tilling for agricultural use occurred at SRS between 1865 and 1950, peaking in the 1920s (Kilgo and Blake 2005). Historic farms were small and dispersed across the landscape, leaving many pockets of longleaf woodlands untilled (Kilgo and Blake 2005). Land-use histories were determined from aerial photographs taken in 1951, just prior to agricultural abandonment that followed the creation of SRS. Non-agricultural sites were open woodlands in 1951, whereas agricultural sites were cultivated fields. Although we could not verify land-use history prior to 1951, it is possible that some of the woodland sites were in agriculture prior to 1951. Historic fire return intervals in this system are estimated to be between 1–6 years (Frost et al. 2006). We determined the number of prescribed fires since 1991 from annual fire records and categorized fire frequencies as frequent (≥ 5 burns, mean number of fires: 6.3 ± 0.79 SE) or infrequent (< 5 burns, mean number of fires: 3.2 ± 0.57 SE), as was previously determined to separate these sites based on metrics of the plant community (Brudvig et al. 2014). There was no difference in the total number of burns since 1991 between land-use histories (GLM, poisson distribution: $F_{1,16} = 0.01$, $p = 0.94$). To provide time for recolonization of grasshoppers following prescribed burns (Knight and Holt 2005), we did not use sites that had burned, or were near sites that burned (> 50 m), during the year of the study.

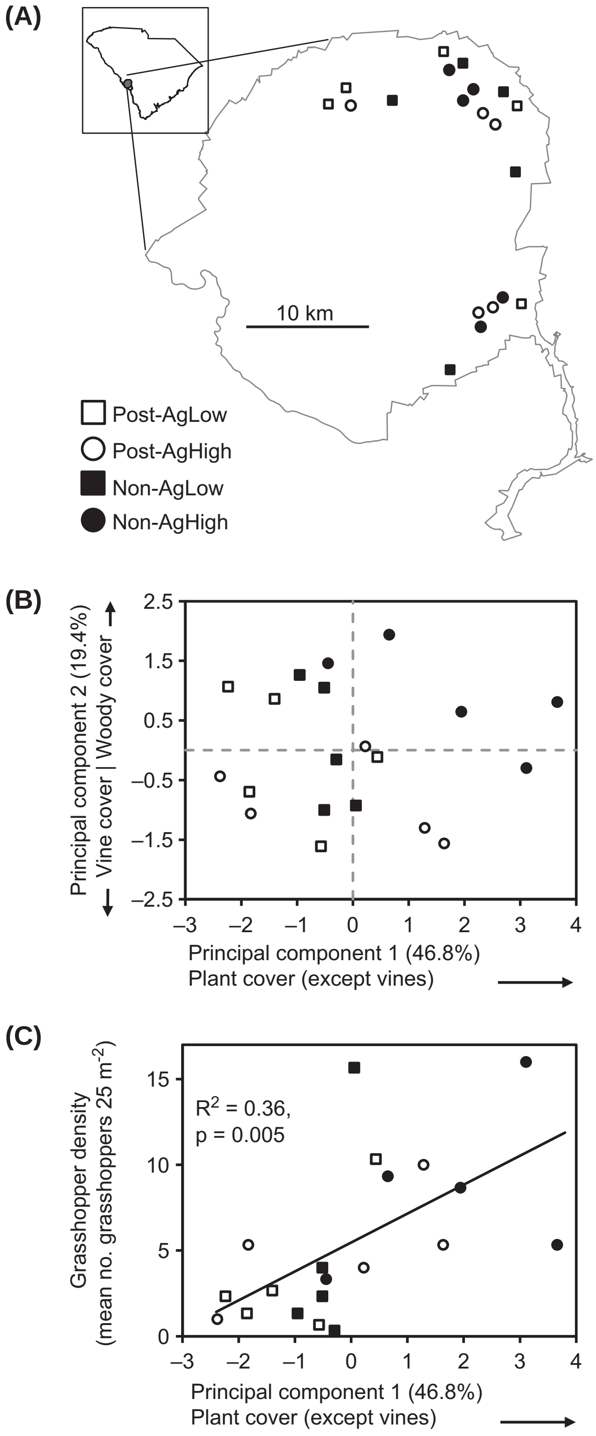

Figure 1. (A) Map of the study sites within the Savannah River Site, SC. (B) Principal component ordination of sites based on plant functional groups. Axis 1 explained 46.8% of the variability in the cover of six plant functional groups and represents an index of plant cover, particularly forb, grass, and legume cover. Axis 1 differed between land use histories ($p = 0.04$) and fire frequency ($p = 0.02$), but the interaction was not significant ($p = 0.30$). (C) Grasshopper density response to herbaceous cover. The trend line and R^2 are for all treatments pooled. Post-Ag = post-agricultural sites, Non-Ag = sites with no history of agriculture, Low = low fire frequency (< 5 burns since 1991), High = high fire frequency (≥ 5 burns since 1991).

Site characteristics

We measured vegetation cover in 12 1-m^2 quadrats at each site between 7 and 14 July 2011. Quadrats were nested in the corners of three 5 × 5 m plots, separated by approximately 5 m. In each quadrat, we recorded the percent cover of six functional groups – grasses, forbs, legumes, woody plants, vines (woody and herbaceous) and ferns – and averaged the values for each functional group at each site. Maximum vegetation height was recorded in two 1-m^2 plots within 1 m of the herbivore exclosures to quantify the potential for light competition. We also collected grasshoppers between 23 July and 6 August 2011 in the three 5 × 5 m plots. We used a 38 cm diameter sweep net to vigorously sweep through the vegetation for three 3-min periods with approximately 15 min rest periods between sweeps, resulting in each plot being swept three times within a one hour period on a single day. All sweeping was conducted between 09:00–15:00 under sunny conditions on calm days. Sampling under consistent conditions is often used to control for environmental variability when grasshoppers communities are sampled on only one occasion at a site (Squitier and Capinera 2002, Joern 2005). Grasshoppers were stored in a freezer and counted later to get one measure of grasshopper density per site. We averaged the number of grasshoppers collected in the three plots to get one number of grasshopper density (25 m^{-2}) per site. We also identified each individual to species, or genus in some cases, in order to determine grasshopper community composition at each site. A formal analysis of the grasshopper community will be presented in a separate manuscript. Voucher specimens have been deposited in the Insect Research Collection of the Dept of Entomology, Univ. of Wisconsin-Madison. In order to quantify potential differences in canopy closure related to land-use history and fire frequency, we calculated the mean proportion of canopy closure in photos taken with a hemispherical lens 1 m above the ground in three plots per site following standard protocol (Hemiview ver. 2.1). Canopy closure (mean = 58.1%, SD = 8.9%) did not differ with land-use history ($F_{1,16} = 0.98$, $p = 0.34$), fire frequency ($F_{1,16} = 0.15$, $p = 0.70$), or the interaction ($F_{1,16} = 1.59$, $p = 0.23$).

Experimental design

We used four plant species for the field experiment: *Pityopsis graminifolia* (forb), *Schizachyrium scoparium* (grass), *Solidago odora* (forb) and *Tephrosia virginiana* (legume); nomenclature follows the USDA, NRCS (National Plant Data Team, Greensboro, NC, USA). Upland pine savanna is suitable habitat for all four of these species (Kilgo and Blake 2005) and these species were previously found to be indicators of non-agricultural, frequently burned longleaf pine habitat in our study sites at SRS and other locations in the southeast (Brudvig et al. 2014). Plants were selected to represent a variety of functional groups and a range of palatabilities based on the literature (Knight and Holt 2005, Hakes and Cronin 2012) and field observations; this variation in palatability was confirmed with ancillary feeding trials using *Melanoplus angustipennis* (Supplementary material Appendix 2), a common grasshopper species at our experimental sites that was most frequently observed feeding on our experimental plants (Hahn unpubl.). Supplementary material Appendix 3 contains additional information on the relative abundance of grasshopper species collected at our site. Rank-order preference to *M. angustipennis* was: *S. odora*, *P. graminifolia*, *T. virginiana* and *S. scoparium*. Seeds were collected from natural populations at the study site and were planted on 27–29 April 2011, germinated indoors (approximately 20°C), moved to a greenhouse after three weeks and then transplanted into 10.8 × 16.2 cm (height × diameter) plastic containers containing a 50:50 mix of potting soil and sand. Each container received one individual of each species, and each container was randomly assigned to a field site with different combinations of land-use histories and fire frequencies ($n = 5$ replicate sites per habitat type).

On 27–29 June 2011, we deployed the containers to the field and buried them so that the rim of the container was flush with the soil or leaf litter surface. By controlling for soil conditions across sites in our experimental containers (i.e. all plants were in a potting soil/sand mixture), we were able to evaluate herbivory without any potentially confounding effects of differences in plant tissue quality from growing in soils that may differ in nutrients between land-use histories or fire regimes (Verheyen et al. 1999, Fraterrigo et al. 2005). Containers received one of two experimental treatments: full exclosures or pseudo-exclosures. The full exclosure treatment consisted of an herbivore cage ($0.125\ m^2 \times 90$ cm height screen cage) and the pseudo-exclosure treatments consisted of the same cages with ten approximately 10 cm diameter holes to permit grasshopper entrance. Exclosures were buried approximately 5 cm into the soil. This experimental design resulted in a split-plot experiment with habitat type (land-use, fire frequency, and the interaction) as the whole plot factors and the herbivore treatments as the split-plot factor.

These exclosures have been successfully deployed in another study focusing on grasshopper herbivory at our field site (Evans et al. 2012). Field observations suggest that exclosures and pseudo-exclosures worked as intended: grasshoppers were occasionally seen feeding on experimental plants inside the pseudo-exclosures but never on plants in the full exclosures (Hahn unpubl.), and very few leaves had evidence of herbivore damage inside the full exclosures. Both exclosure types excluded large vertebrate herbivores but the pseudo-exclosures admitted rodents. However, only a few species of herbivorous rodents occur at our field sites (Cothran et al. 1991) and only two experimental plants showed evidence of rodent grazing (Hahn unpubl.). To determine how the exclosures affected the light and temperature environments, we deployed light and temperature loggers inside the exclosures (i.e. exclosures and pseudo-exclosures) and adjacent open areas ($n = 4$ replicates). Data were collected in 10 min intervals between 07:00–19:00 hours on 1–3 August 2013 and averaged before analysis. Although both exclosures and pseudo-exclosures slightly reduced light relative to open areas (ln-transformed lux, $t_9 = -7.17$, $p < 0.001$ and $t_9 = -7.45$, $p < 0.001$, means ± 1 SE: exclosure = 10.42 ± 0.05, pseudo-exclosure = 10.40 ± 0.05, open = 10.97 ± 0.05), there was no difference in the light environment between the exclosures and pseudo-exclosures that we used for our experiment ($t_9 = 0.28$, $p = 0.78$). Temperature followed similar patterns (data not shown).

Prior to deployment to the field (25–26 June 2011) and after termination of the experiment (2 August 2011), we measured plant height, length of the longest leaf, and counted the number of fully emerged leaves on each plant. We counted the number of damaged (> 5% of leaf tissue removed by chewing insects) and total leaves on each plant and harvested aboveground biomass at the end of the experiment. Aboveground biomass was weighed after drying for 48h at 60°C. The duration of this experiment is within the range of other experiments investigating the effects of herbivores on leaf damage and plant performance within a growing season (White and Whitham 2000, Palmer et al. 2003, Knight and Holt 2005).

Statistical analysis

Question 1. Does agricultural land-use history or fire suppression alter the plant community or grasshopper density?

We evaluated the effect land-use history and fire suppression on plant community composition (i.e. change the relative abundance of plant functional groups) by first conducting a principal component analysis on the correlation matrix of cover of the six plant functional groups. We performed a square root transformation and then centered the data prior to analysis. We then analyzed the two principal component axes using (separate) linear models (ANOVA) with land-use history (post-agriculture or remnant), fire frequency (high or low), and the interaction as predictor variables. We evaluated the effects of land-use history and fire suppression on grasshopper density using a linear model (ANOVA) to compare grasshopper density among the habitat types with land-use history, fire frequency, and the interaction as predictor variables. We used a second linear model to regress grasshopper density against the first principal component axis to aid in interpretation of the ANOVA.

Question 2. Does habitat type (i.e., combinations of land-use history and fire suppression) alter the effect that herbivores have on plant biomass?

We evaluated the effect of agricultural land-use history and fire suppression on herbivory on focal plants by comparing dried aboveground plant biomass between exclosure and pseudo-exclosure treatments across sites that varied in their land-use history and fire frequency. Separate models were conducted for each of the four plant species. These linear mixed models included plant biomass as the response variable, with land-use history, fire frequency, herbivore treatment (exclosure or pseudo-exclosure) and all interactions as fixed effects. Covariates that could affect final plant biomass (stem height measured prior to the experiment, canopy closure and height of natural vegetation within 1 m of the experimental plants, as well as their interactions with main effects) were included in the biomass models when the addition of the covariate reduced the AICc by > 2 (Burnham and Anderson 2002). We evaluated each covariate (and interactions) separately to maintain statistical power (i.e. we did not have enough degrees of freedom to run models that included all potential covariates and interaction terms), while still allowing us to detect higher-order interactions among covariates and main effects. When significant herbivore effects (i.e. factor × herbivore treatment) were detected, linear contrasts were conducted to compare the exclosure and pseudo-exclosure herbivore treatments within habitat types. To ensure that there were no differences in biomass among the habitat types, we compared plant biomass within the exclosure treatments among the four habitat types. We conducted similar analyses comparing herbivore damage (number of leaves damaged) on plants exposed to herbivores (i.e. pseudo-exclosure treatment). We also evaluated whether herbivore damage was linked to plant biomass using linear regressions with the proportion of leaves damaged as the predictor variable and plant biomass on surviving plants in the pseudo-exclosure treatments as the response variable. We focus on the biomass results, rather than herbivore damage, because it is more closely linked to fitness than is herbivore damage per se (Zangerl et al. 2002) and is often used as a proxy for fitness (Hakes and Cronin 2012).

Evaluating the mechanisms whereby land-use history and fire suppression could alter herbivory

We evaluated the effects of herbivore density and plant community composition on herbivory rates among sites that vary in land-use history and fire frequency using a linear model with the difference in biomass between the full exclosure and pseudo-exclosure as the response variable. Predictor variables included grasshopper density and the first two principal components from the vegetation analysis (see Question 1 above), as well as quadratic terms for all variables to account for potential non-linear trends. We used a stepwise AIC model selection procedure for model reduction. These analyses were conducted only for plant species significantly affected by herbivores (see Question 2 above). All linear models and multivariate analyses were conducted in R (R ver. 3.0.1) and linear mixed-models were conducted in SAS (SAS ver. 9.3). We examined residual plots to ensure that all models adequately fit the data. We conducted analyses similar to ones described above using fire frequency as a continuous variable. These results were not qualitatively different, so we present the results that treat fire as a categorical variable.

Data available from the Dryad Digital Repository: <http://dx.doi.org/10.5061/dryad.142m0> (Hahn and Orrock 2015).

Results

Question 1. Does agricultural land-use history or fire suppression alter the plant community or grasshopper density?

The first principal component axis explained 46.8% of the original variance in the cover of the square root-transformed matrix of the six plant functional groups and was a general index of plant cover, except for vines, with forb, grass and legume cover having the highest loadings (Table 1, Fig. 1B). This axis differed with land-use history and fire frequency, but not the interaction (Table 1). The second principal component axis explained an additional 19.4% of the original variance and was a contrast between vine cover and woody/fern cover (Table 1). This axis differed between land-use history, but not fire frequency, although the interaction was marginally significant (Table 1).

Table 1. Loadings for the first three principal components from the analysis on the plant functional group matrix. % variance indicates the percent of variance in the original matrix explained by that principal component. Land-use, Fire and Interaction rows show F-values from univariate ANOVAs.

Variable	PC1	PC2	PC3
Grass	0.48	−0.33	0.07
Forb	0.54	−0.08	−0.10
Legume	0.47	−0.38	0.17
Woody	0.26	0.49	−0.70
Vine	−0.16	−0.60	−0.68
Fern	0.41	0.38	0.03
% variance	46.8	19.4	15.7
Land-use	4.80*	5.18*	0.00
Fire	6.61*	0.02	0.75
Interaction	1.16	3.73†	1.59

$^{\dagger}p < 0.1$, $^{*}p < 0.05$.

Grasshopper density did not differ with land-use history (ANOVA: $F_{1,16} = 1.23$, $p = 0.28$), fire frequency (ANOVA: $F_{1,16} = 1.69$, $p = 0.21$), or the interaction (ANOVA: $F_{1,16} = 0.26$, $p = 0.62$). However, grasshopper density increased with the first principal component (i.e. plant cover) (linear regression: $F_{1,18} = 10.03$, $p = 0.005$, $R^2 = 0.36$, Fig. 1C).

Question 2: Does habitat type (i.e. combinations of land-use history and fire suppression) alter the effect that herbivores have on plant biomass?

Details on pre-deployment plant sizes are provided in Supplementary material Appendix 4. Several of the forty initial plants per species died from unknown causes during the experiment: *Pityopsis graminifolia*, n = 11; *Schizachyrium scoparium*, n = 2; *Solidago odora*, n = 1; *Tephrosia virginiana*, n = 2. Dead plants were treated as zeros in the biomass analyses, although the results were similar when they were deleted.

Solidago odora biomass was affected by the interaction between land-use history, fire frequency and exclosure treatment, after adjusting for initial plant height (ANCOVA: $F_{1,15} = 6.05$, $p = 0.03$, Table 2, Fig. 2C). Including initial plant height and the initial plant height-by-land-use history interaction term improved the model fit (AICc = 88.0 with covariates; AICc = 93.2 without covariates), but canopy closure, height of vegetation within 1 m of the exclosures and other interaction terms between initial plant height and main effects did not. Biomass in exclosures was approximately 1.5 times greater than in pseudo-exclosure treatments in high fire post-agricultural sites (linear contrast: mean difference = 0.85 g, SE = 0.4, t = 2.13, $p = 0.05$) and nearly two times greater in low fire remnant woodlands (linear contrast: mean difference = 1.01 g, SE = 0.3, t = 2.97, $p = 0.01$). We found no differences in *S. odora* biomass across the four habitat types within the exclosure treatment (simple effects of land-use history, fire frequency, and the interaction, given the 'exclosure' level of the herbivory treatment: all $p > 0.12$). The proportion of leaves damaged by chewing invertebrates on *S. odora* varied among the land-use history and fire frequency sites (Supplementary material Appendix 5 Fig. A2) and was associated with lower final biomass of plants in

Table 2. ANOVA table for linear mixed models of plant biomass after the herbivory experiment.

	Pityopsis graminifolia		*Schizachyrium scoparium*		*Solidago odora*		*Tephrosia virginiana*	
Effect	F	p	F	p	F	p	F	p
Land use history	0.29	0.60	0.32	0.58	6.41	0.02	0.29	0.60
Fire frequency	0.14	0.72	0.40	0.54	0.64	0.43	0.13	0.73
Land use × Fire	0.32	0.58	1.96	0.18	4.14	0.06	0.10	0.76
Herbivores	0.91	0.35	2.80	0.12	7.74	0.02	1.25	0.28
Herb × Land use	0.51	0.49	1.52	0.24	0.38	0.55	1.29	0.27
Herb × Fire	0.27	0.61	0.68	0.42	0.02	0.89	0.50	0.49
Herb × Land use × Fire	0.09	0.77	0.10	0.75	6.05	0.03	0.77	0.39
Height (cm)	5.63	0.03	42.70	<.001	6.18	0.03	–	–
Height × Land use	–	–	–	–	4.48	0.05	–	–

pseudo-exclosures (linear regression: $F_{1,17} = 7.90$, $p = 0.01$, Supplementary material Appendix 6 Fig. A3).

Biomass of the other three plant species were not different between the herbivore exclusion treatments, land-use histories, or fire frequencies (Table 2), although several of the species did have lower mean biomass in the pseudo-exclosures than in the exclosures (Fig. 2A, B, D). Three dead *P. graminfolia* had evidence of damage by leaf chewing insects (all in pseudo-exclosures), and mortality for *P. graminifolia* was greater for plants exposed to herbivores (GLMM, binomial response: $F_{1,19} = 5.72$, $p = 0.027$; proportion mortality: 0.44 ± 0.12 SE in pseudo-exclosures and 0.09 ± 0.06 in exclosures), but did not differ with land-use history, fire frequency or any interactions (all $p > 0.29$). The proportion of leaves damaged on *P. graminfolia* was negatively related to biomass (linear regression: $F_{1,10} = 20.32$, $p = 0.001$, Supplementary material Appendix 6 Fig. A3) of surviving plants damaged by herbivores in pseudo-exclosures. Biomass of *S. scoparium* (linear regression: $F_{1,14} = 0.23$, $p = 0.64$, Supplementary material Appendix 6 Fig. A3) and *T. virginiana* (linear regression: $F_{1,12} = 0.49$, $p = 0.50$, Supplementary material Appendix 6 Fig. A3) was not related to the proportion of leaves damaged.

Evaluating the mechanisms whereby land-use history and fire suppression could alter herbivory

Solidago odora was the only species that had reduced biomass when exposed to herbivores (Fig. 2). The stepwise model selection procedure selected a model (multiple

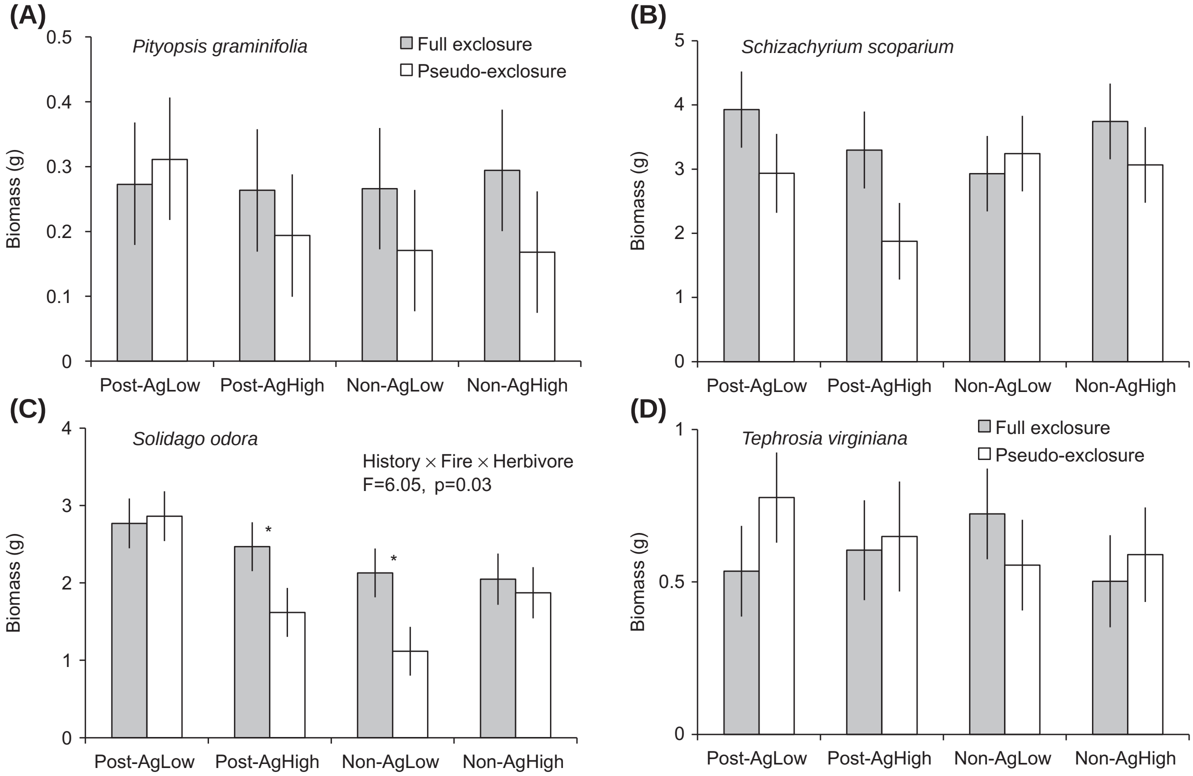

Figure 2. Biomass (g) at the end of the five week field experiment, after adjusting for initial plant height. Plants were protected from herbivores with mesh screen (Exclosure) or mesh screen with holes to allow invertebrate herbivores to enter (Pseudo-exclosure). Post-Ag = post-agricultural sites, Non-Ag = sites with no history of agriculture, Low = low fire frequency (< 5 burns since 1991), High = high fire frequency (≥ 5 burns since 1991). Significant ($p < 0.05$) herbivore effects (i.e. factor × exclosure interactions) are listed on each panel. Asterisks (*) indicate significant ($p < 0.05$) difference between exclosure and pseudo-exclosure treatments. Bars are ± 1 SE. Note the value changes on the y-axis among the different species.

linear regression: $F_{3,15} = 4.90$, $p = 0.01$, $R^2 = 0.48$, Fig. 3) that included the first principal component from the vegetation analysis (slope = 0.22, t = 1.55, p = 0.14), the quadratic term of this principal component (slope = −0.18, t = −3.32, p = 0.004), and grasshopper density (slope = −0.09, t = −2.04, p = 0.058). Because we found a relationship between the first principal component from the vegetation analysis and grasshopper density (Fig. 1C), we tested for collinearity in this model. The variance inflation factors were 1.96, 1.30, and 1.59 respectively, suggesting that collinearity was not problematic. We also conducted the same analysis, but included the density of the three dominant grasshopper species, or grasshopper community composition, instead of overall grasshopper density, to test for the effects of individual grasshopper species or overall community composition. Overall, these models were similar, but worse than when we used overall grasshopper density (Supplementary material Appendix 3).

Discussion

The results from our large-scale study show that agricultural legacies and fire regimes can interact to affect herbivory and suggest that these effects arise because changes in the plant community affect herbivore foraging patterns, rather than through changes in herbivore density. Historic agriculture and reduced fire frequency were associated with altered plant communities, and herbivory exhibited a hump-shaped relationship to plant cover (mainly herbaceous cover), where the difference in plant biomass between the herbivore exclosures increased at low levels of plant cover but leveled off and decreased at higher levels of plant cover, suggesting a saturating effect of neighboring plant cover on herbivory of our focal plants. We also found a weak, negative relationship between the difference in plant biomass between the herbivore exclosures and grasshopper density, suggesting lower per plant herbivory rates at higher grasshopper density, further supporting the saturating effect of plant cover. Furthermore, these effects were consistent with herbivore feeding preferences, as we found strong herbivore effects on the biomass of one palatable forb species (*Solidago odora*), weak herbivore effects on mortality and biomass of another forb (*Pityopsis graminifolia*), but no effect of herbivores on less preferred plants (*Schizachyrium scoparium*, grass; and *Tephrosia virginiana*, legume). Across a multi-site landscape, our results demonstrate how past and present human disturbance can alter species interactions, which have previously been hypothesized to be potentially important for regulating plant species occurrences in post-agricultural landscapes (Flinn and Vellend 2005, Cramer et al. 2008). Below we discuss how our results may generate the patterns in herbivory we observed in anthropogenically modified habitats by applying a novel perspective to three themes in plant–herbivore interactions: 1) that neighboring plants matter in affecting herbivory (Atsatt and O'dowd 1976, Palmer et al. 2003, Barbosa et al. 2009), 2) that herbivore density is a poor predictor of herbivory in multi-species plant communities (Cronin et al. 2010), and 3) herbivore preference can affect plant–herbivore interactions (Schmitz 2006, Allan and Crawley 2011).

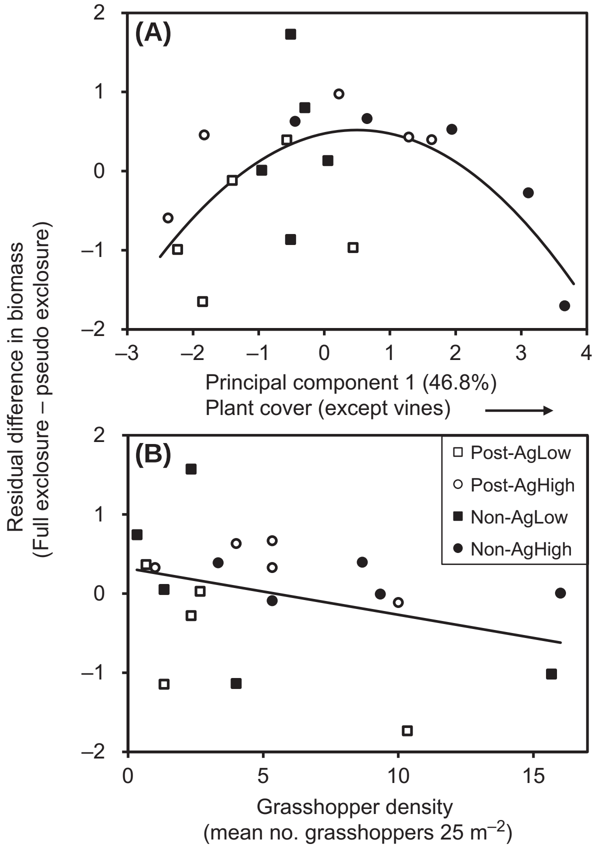

Figure 3. Effect of (A) the first principal component from the analysis of the plant community and its quadratic term and (B) grasshopper density on the (residual) difference in biomass between the full exclosure minus the pseudo-exclosure for *Solidago odora*. The full model included the principal component, the quadratic term of the principal component, and grasshopper density ($F = 4.90$, $p = 0.01$, $R^2 = 0.48$). The graphs show each variable regressed against the residuals from a model containing the other term.

Neighboring plants matter for affecting herbivory

Herbivore effects on plant communities can depend on herbivore density, foraging behavior, or composition of the herbivore community (Crawley 1983). In our experiment, high herbaceous cover might be satiating herbivores, or making focal plants less apparent, resulting in lower per plant attack rates on focal plants, more often in frequently burned remnant woodlands. This result is similar to theoretical models (Hambäck et al. 2014) and other field studies that have examined herbivory rates in plots with varying amounts of herbaceous plant cover. For example, Hakes and Cronin (2012) found greater herbivore damage on transplanted *Solidago altissima* in late successional fields, where encroachment of woody plants reduced herbaceous cover, compared to early successional fields, which had greater herbaceous cover. One potential explanation is that the herbaceous over present in the old fields reduced per plant herbivory to the focal plants (Hakes and Cronin 2012). Similarly, Underwood and Halpern (2012) found a hump-shaped relationship between neighboring plant density and herbivory rates on focal *Solanum carolinense*, likely due to herbivore satiation at high plant density. In infrequently burned non-agricultural woodlands and frequently burned post-agricultural woodlands, we found that herbivores grazed more intensively on the palatable forb *S. odora* likely because herbaceous plants supply superior nutrition to grasshoppers over the woody foliage (Joern 1983, Behmer et al. 2001) that is common at these sites (Fig. 1B). In other words, herbivores may persist on suboptimal resources, and preferentially consume herbaceous plants when available. Collectively, these results highlight one way in which human activities that alter the structure of the plant community can modify plant–herbivore interactions.

Herbivore density is a poor predictor of herbivory

Although palatable plants might alter herbivory by increasing herbivore density in some systems (White and Whitham 2000, Rand 2003), we found a weak negative effect of grasshopper density on herbivory. One explanation for this is that grasshoppers may not be able to build up to levels capable of reducing plant cover in the years following fires, relative to post-fire regeneration rates of plant cover (Knight and Holt 2005, Kim and Holt 2012). Thus, it could be expected that grasshoppers would not be able to consume all the available plant material, particularity at sites with high pre-fire plant cover, resulting in a saturating effect of plant cover on overall herbivory rates. Furthermore, this effect should be stronger in non-agricultural sites with high pre-fire vegetation cover, where we documented lower reductions in plant biomass from herbivory. The exception in our study might be at low to medium densities of herbivore densities and plant cover, as we found greater levels of herbivory at low grasshopper densities (Fig. 3B) and herbivory increased with increases at low levels of plant cover (Fig. 3A). In our post-agricultural sites with low to medium levels of plant cover, herbivory may act synergistically with reduced fire frequency by decreasing plant cover and potentially establishment in the short term. However, in the long term, the effect that herbivores have on plant biomass or establishment should diminish as a deepening litter layer (Hiers et al. 2007) and closing canopy (Glitzenstein et al. 2012) cause concurrent reductions in plant cover and establishment, as well as grasshopper density (Fig. 1C).

Changes to grasshopper community composition related to land-use history and fire frequency (Joern 2005) might also contribute to differences in herbivory rates. However, we found no effect, or weak negative effects, of the densities of three dominant species of grasshopper or grasshopper community composition (Supplementary material Appendix 2), suggesting that herbivore community composition probably contributes little to the herbivory rates we documented. Orthopterans comprised 40.7% of all non-Aranae arthropods collected in our sweep net samples and are the dominant insect herbivore in our system (Knight and Holt 2005, Evans et al. 2012), so the majority of leaf chewing damage was likely from orthoperans. Other insects, such as chrysomelid beetles or other insects associated with *Solidago* species (Root and Cappuccino 1992), likely contributed only a minor amount of chewing damage to our plants and were never observed on our experimental plants (Hahn unpubl.). Although our data point to the important role of herbivore choice in generating different patterns in herbivory, future studies that directly manipulate herbivore communities within human modified landscapes will be necessary to understand whether herbivore choice is changing solely due to vegetative cover, or whether interactions among herbivores and other organisms (i.e. competition or predation) are also important for generating the differential patterns of herbivory we observed.

Herbivore preference can affect plant–herbivore interactions

Feeding preferences of the common grasshopper species at our study site might help explain why the effect of herbivores differed among the four focal species in this study. The grasshoppers *Melanoplus angustipennis*, *M.* sp. (*scudderi* and *puer* group) (both subfamily Melanoplinae) and *Schistocerca* species (subfamily Cyrtacanthacridinae) are abundant at our site and all are classified as mixed feeders that will consume grasses, forbs, and some woody foliage (Joern 1983). In six ancillary feeding trials involving twelve plant species, *M. angustipennis* consumed *S. odora* (Asteraceae) in 50% of the trials, *P. graminifolia* (Asteraceae) in 33%, *T. virginiana* (Fabaceae) in 17%, and *S. scoparium* (Poaceae) in none of the feeding trials (Supplementary material Appendix 2), suggesting a strong preference for forb species used in our experiment. In our field experiment, the biomass of the two most palatable species decreased with increasing herbivore damage (Supplementary material Appendix 6 Fig. A3), suggesting that palatable plants receive damage levels that are high enough to reduce plant biomass. Finally, it is notable that *M. angustipennis* consumed several species of woody plants and vines, albeit infrequently, in our feedings trials (Supplementary material Appendix 2), suggesting that grasshoppers may be able to persist in habitats dominated by woody plants and vines, but preferentially feed on herbaceous plants when they are available. This effect could lead to herbivores maintaining the degraded states created by human disturbances if herbivores are having disproportionate effects on plant performance of rare, palatable plants over less preferred, dominant plants.

Future directions for studying species interactions in post-agricultural systems

Because past agricultural use and fire suppression affect a significant fraction of the terrestrial biosphere (Briggs et al. 2005, Flinn and Vellend 2005), our results suggest that plant-herbivore interactions may be modified in many ecosystems. Although our study cannot separate the effects of disturbance from plant community composition per se, our results are informative because disturbance and plant community composition are inextricably linked in contemporary landscapes (Flinn and Vellend 2005, Cramer et al. 2008, Brudvig and Damschen 2011). Furthermore, both land-use history and fire can alter other factors that could affect plant-herbivore interactions, such as soil nutrients (Verheyen et al. 1999, Fraterrigo et al. 2005), plant tissue quality (Fraterrigo et al. 2006, Kim and Holt 2012), and herbivore community composition (Kim and Holt 2012, Joern 2005). We avoided any potential effects of soil nutrient differences among land-use histories and fire frequency sites by using standard soil in all experimental plots in order to focus explicitly on differences in herbivory. These topics likely provide profitable avenues for future investigation.

Our results document a link between human modified systems and herbivory, driven in part by altered plant communities related to historical agriculture and contemporary fire regimes. Importantly, shifts in herbivory we documented were only found for one of the four study species, suggesting that the effect of human disturbance on herbivory is likely to be idiosyncratic. Furthermore, our results suggest a threshold in the relationship between herbivory and plant cover, where herbivore density may be an important driver of herbivory rates at low to medium plant cover, but at higher levels of plant cover herbivores become satiated resulting in lower per plant herbivory rates on palatable plants. Although our results highlight a link between human activities and altered herbivory rates, invertebrate herbivory can limit plant establishment into new habitats (Parker and Root 1981, Branson and Sword 2009, Hahn and Dornbush 2012) and herbivory may be an important factor limiting plant establishment recently documented in post-agricultural systems (Baeten et al. 2009). Thus, while our results provide a mechanism that may generate differential herbivory on palatable herbaceous plants in post-agricultural habitats (i.e. altered herbivory rates), future studies should evaluate whether these differences in herbivory can limit plant establishment and contribute to the maintenance of degraded plant communities in post-agricultural habitats. In particular, our results suggest that herbivory would be most likely to limit plant establishment in regenerating systems or during ecological restoration where existing plant cover is low. In a broader sense, since many ecological investigations either avoid or ignore historic land-use (Martin et al. 2012), unexplained variation in previous studies may be attributed to legacy effects on ecological interactions.

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References

Allan, E. and Crawley, M. J. 2011. Contrasting effects of insect and molluscan herbivores on plant diversity in a long-term field experiment. – Ecol. Lett. 14: 1246–1253.

Atsatt, P. and O'dowd, D. 1976. Plant defense guilds. – Science 193: 24–29.

Baeten, L. et al. 2009. Low recruitment across life stages partly accounts for the slow colonization of forest herbs. – J. Ecol. 97: 109–117.

Baraza, E. et al. 2006. Conditional outcomes in plant–herbivore interactions: neighbors matter. – Oikos 113: 148–156.

Barbosa, P. et al. 2009. Associational resistance and associational susceptibility: having right or wrong neighbors. – Annu. Rev. Ecol. Evol. Syst. 40: 1–20.

Behmer, S. T. et al. 2001. Frequency-dependent food selection in locusts: a geometric analysis of the role of nutrient balancing. – Anim. Behav. 61: 995–1005.

Branson, D. H. and Sword, G. A. 2009. Grasshopper herbivory affects native plant diversity and abundance in a grassland dominated by the exotic grass *Agropyron cristatum*. – Restor. Ecol. 17: 89–96.

Briggs, J. M. et al. 2005. An ecosystem in transition: causes and consequences of the conversion of mesic grassland to shrubland. – BioScience 55: 243–254.

Brudvig, L. A. and Damschen, E. I. 2011. Land-use history, historical connectivity, and land management interact to determine longleaf pine woodland understory richness and composition. – Ecography 34: 257–266.

Brudvig, L. A. et al. 2014. Land-use history and contemporary management inform an ecological reference model for longleaf pine woodland understory plant communities. – PLoS ONE: 9: e86604.

Burnham, K. P. and Anderson, D. A. 2002. Model selection and multimodel inference: a practical information-theoretic approach. – Springer.

Caccia, F. D. et al. 2009. Direct and indirect effects of understory bamboo shape tree regeneration niches in mixed temperate forest. – Oecologia 161: 771–780.

Cease, A. J. et al. 2012. Heavy livestock grazing promotes locust outbreaks by lowering plant nitrogen content. – Science 335: 467–469.

Cothran, E. G. et al. 1991. Mammals of the Savannah River Site. – Dept of Energy's Savannah River Site, Aiken, SC.

Cramer, V. A. et al. 2008. What's new about old fields? Land abandonment and ecosystem assembly. – Trends Ecol. Evol. 23: 104–12.

Crawley, M. J. 1983. Herbivory: the dynamics of animal–plant interactions. – Blackwell.

Cronin, J. P. et al. 2010. A simultaneous test of trophic interaction models: which vegetation characteristic explains herbivore control over plant community mass? – Ecol. Lett. 13: 202–212.

Evans, D. et al. 2012. Habitat patch shape, not corridors, determines herbivory and fruit production of an annual plant. – Ecology 93: 1016–1025.

Flinn, K. and Vellend, M. 2005. Recovery of forest plant communities in post-agricultural landscapes. – Front. Ecol. Environ. 3: 243–250.

Fraterrigo, J. M. et al. 2005. Effects of past land use on spatial heterogeneity of soil nutrients in southern Appalachian forests. – Ecol. Monogr. 75: 215–230.

Fraterrigo, J. M. et al. 2006. Previous land use alters plant allocation and growth in forest herbs. – J. Ecol. 94: 548–557.

Frost, C. et al. 2006. History and future of the longleaf pine ecosystem. – In: Jose, S. et al. (eds), The longleaf pine ecosystem: ecology, silviculture and restoration. Springer, pp. 9–42.

Glitzenstein, J. S. et al. 2012. Fire-frequency effects on vegetation in north Florida pinelands: another look at the long-term Stoddard Fire Research Plots at Tall Timbers Research Station. – For. Ecol. Manage. 264: 197–209.

Hahn, P. G. and Dornbush, M. E. 2012. Exotic consumers interact with exotic plants to mediate native plant survival in a Midwestern forest herb layer. – Biol. Invas. 14: 449–460.

Hahn, P. G. and Orrock, J. L. 2015. Data from: Land-use legacies and present fire regimes interact to mediate herbivory by altering the neighboring plant community. – Dryad Digital Repository, <http://dx.doi.org/10.5061/dryad.142m0>.

Hakes, A. and Cronin, J. T. 2012. Successional changes in plant resistance and tolerance to herbivory. – Ecology 93: 1059–1070.

Hambäck, P. A. et al. 2014. Effects of plant neighborhoods on plant–herbivore interactions: resource dilution and associational effects. – Ecology 95:1370–1383.

Hiers, J. K. et al. 2007. Forest floor depth mediates understory vigor in xeric *Pinus palustris* ecosystems. – Ecol. Appl. 17: 806–814.

Joern, A. 1983. Host plant utilization by grasshoppers (Orthoptera: Acrididae) from a sandhills prairie. – J. Range Manage. 36: 793–797.

Joern, A. 2005. Disturbance by fire frequency and bison grazing modulate grasshopper assemblages in tallgrass prairie. – Ecology 86: 861–873.

Joern, A. and Laws, A. N. 2013. Ecological mechanisms underlying arthropod species diversity in grasslands. – Annu. Rev. Entomol. 58: 19–36.

Kilgo, J. and Blake, J. I. 2005. Ecology and management of a forested landscape: fifty years on the Savannah River Site. – Island Press.

Kim, T. N. and Holt, R. D. 2012. The direct and indirect effects of fire on the assembly of insect herbivore communities: examples from the Florida scrub habitat. – Oecologia 168: 997–1012.

Kirkman, L. et al. 2004. Ground cover recovery patterns and life-history traits: implications for restoration obstacles and opportunities in a species-rich savanna. – J. Ecol. 92: 409–421.

Knight, T. M. and Holt, R. D. 2005. Fire generates spatial gradients in herbivory: an example from a Florida sandhill ecosystem. – Ecology 86: 587–593.

Martin, L. J. et al. 2012. Mapping where ecologists work: biases in the global distribution of terrestrial ecological observations. – Front. Ecol. Environ. 10: 195–201.

Orrock, J. L. et al. 2008. Apparent competition with an exotic plant reduces native plant establishment. – Ecology 89: 1168–1174.

Orrock, J. L. et al. 2010. Refuge-mediated apparent competition in plant–consumer interactions. – Ecol. Lett. 13: 11–20.

Palmer, S. C. F. et al. 2003. The perils of having tasty neighbors: grazing impacts of large herbivores at vegetation boundaries. – Ecology 84: 2877–2890.

Parker, M. A. and Root, R. B. 1981. Insect herbivores limit habitat distribution of a native composite, *Machaeranthera canescens*. – Ecology 62: 1390–1392.

Ramankutty, N. and Foley, J. A. 1999. Estimating historical changes in land cover: North American croplands from 1850 to 1992. – Global Ecol. Biogeogr. 8: 381–396.

Rand, T. A. 2003. Herbivore-mediated apparent competition between two salt marsh forbs. – Ecology 84: 1517–1526.

Root, R. B. and Cappuccino, N. 1992. Patterns in population change and the organization of the insect community assocated with goldenrod. – Ecol. Monogr. 62: 393–420.

Schmitz, O. J. 2006. Predators have large effects on ecosystem properties by changing plant diversity, not plant biomass. – Ecology 87: 1432–1437.

Squitier, J. M. and J. L. Capinera. 2002. Habitat associations of Florida grasshoppers (Orthoptera: Acrididae). – Fla Entomol. 85: 235–244.

Underwood, N. and S. T. Halpern. 2012. Insect herbivores, density dependence, and the performance of the perennial herb *Solanum carolinense*. – Ecology 93: 1026–1035.

Vellend, M. et al. 2007. Homogenization of forest plant communities and weakening of species–environment relationships via agricultural land use. – J. Ecol. 95: 565–573.

Verheyen, K. et al. 1999. The land use history (1278–1990) of a mixed hardwood forest in western Belgium and its relationship with chemical soil characteristics. – J. Biogeogr. 26: 1115–1128.

White, J. A. and Whitham, T. G. 2000. Associational susceptibility of cottonwood to a box elder herbivore. – Ecology 81: 1795–1803.

Wilcove, D. S. et al. 1998. Quantifying threats to imperiled species in the United States. – Bioscience 48: 607–615.

Zangerl, A. R. et al. 2002. Impact of folivory on photosynthesis is greater than the sum of its holes. – Proc. Natl Acad. Sci. USA 99: 1088–1091.

Supplementary material (available as Appendix oik.01445 at <www.oikosjournal.org/readers/appendix>). Appendix 1–6.