Received: 4 April 2025 | Accepted: 21 April 2025

DOI: 10.1002/ecs2.70310

ECOSPHERE
AN ESA OPEN ACCESS JOURNAL

ARTICLE

Vegetation Ecology

Microsite availability, not floral herbivory, limits recruitment in peripheral native thistle populations

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Funding information
Kansas Academy of Science; Colorado Native Plant Society; Wichita State University

Handling Editor: Jaime Madrigal-González

Abstract

Variation in insect herbivory can drive variation in plant fitness and population dynamics. However, our ability to predict the ecological contexts in which insect herbivores will reduce plant fitness or population growth is limited. In theory, populations at the periphery of a plant species' biogeographic range are expected to experience reduced herbivory. Further, in montane landscapes, elevation is expected to drive variation in abiotic conditions and variation in plant–insect interactions. Specifically, less insect herbivory may occur at cooler, higher elevations. To examine these predictions, we quantified effects of inflorescence- and seed-feeding insect herbivores in populations of the short-lived, monocarpic, perennial forb *Cirsium canescens* (Platte thistle) in montane grasslands in Colorado, USA. We asked: (1) Does insect flower head herbivory and pre-dispersal seed predation limit Platte thistle lifetime seed production? (2) Does this insect herbivory limit seedling recruitment? (3) Does ecological context, including spatial—especially elevational—and temporal variation, affect the outcome of these interactions? We conducted insect exclusion experiments in three years at five sites over 52% of Platte thistle's elevation range in our region. We compared both lifetime viable seed production and seedlings recruited between plants with ambient versus insecticide-reduced levels of flower head herbivory. Insect herbivory on flower heads significantly reduced Platte thistle lifetime viable seed production at all sites, independent of elevation. Unexpectedly, however, increasing seed by reducing herbivory did not lead to a proportional increase in seedling recruitment. The relationship between viable seed production and seedling recruitment per plant was non-linear, decelerating across the range of seed production achieved by both plants exposed to and protected from flower head herbivory. While elevation altered Platte thistle flowering phenology, it did not influence insect damage, viable seed production, or seedling recruitment. These results show that flower head- and seed-feeding insect herbivores strongly reduced Platte thistle lifetime viable seed production, a key component of maternal fitness, in these peripheral populations. Yet, the herbivory did not determine population recruitment, suggesting post-dispersal

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processes limit recruitment here. Further, elevation did not drive context-dependent variation in the insect herbivore outcomes.

KEYWORDS
floral herbivory, insect herbivory, insect–plant interactions, montane grasslands, plant demography, plant population dynamics, pre-dispersal seed predation, spatial variation

INTRODUCTION

Insect herbivory and pre-dispersal seed predation can reduce plant fitness (reviews: Bigger & Marvier, 1998, Maron et al., 2014) and limit plant population size (reviews: Katz, 2016; Maron & Crone, 2006). Yet, the consequences of insect herbivory for plant performance and abundance can vary greatly with the abiotic and biotic contexts of the interaction (Bradley et al., 2003; Collinge & Louda, 1988; Louda, 1982, 1983; Miller et al., 2009). For example, Louda and Rodman (1996) showed that *Cardamine cordifolia*'s association with shade beneath shrubs in montane riparian zones in Colorado, USA, resulted from greater insect folivory on sun-exposed plants. Shading reduced drought stress, allowing plants to more effectively produce glucosinolates. In spite of this example, in many instances, factors driving variation in insect herbivores' effects on plants and plant populations remain poorly understood.

To address this knowledge gap, Maron et al. (2014) propose a conceptual framework for incorporating aspects of ecological context into studies of herbivore effects on plant populations. The framework involves quantifying the effect of context on variation in: (1) magnitude of herbivory and pre-dispersal seed predation on plants, (2) plant tolerance for damage, and (3) roles of seed availability versus post-dispersal ecological filters in offspring recruitment. Such filters may include the availability of suitable microsites for germination as well as post-dispersal granivory. Examining how herbivory, plant tolerance, and seed limitation vary with ecological context underlies a mechanistic approach to explaining spatial and temporal variation in herbivores' effects on host plant populations.

The "Species Interactions—Abiotic Stress Hypothesis," formalized by Louthan et al. (2015), proposes that biotic interactions primarily limit species distributions and population growth in benign abiotic contexts, whereas resources and environmental conditions are limiting in stressful contexts. Various environmental stress gradients provide opportunities to evaluate this hypothesis, including gradients from species' biogeographic range center to range edges, sun versus shade boundaries, and intertidal or montane elevation gradients. Montane elevation variation provides an appealing setting for testing the limiting effects of insect herbivory because the abiotic context changes greatly over short distances. As elevation increases, average temperatures decrease, UV radiation increases, and concentrations of both oxygen and carbon dioxide decrease (Körner, 2007), potentially increasing physiological stress for insects and plants. Therefore, in montane landscapes herbivore damage, plant tolerance and recruitment dynamics may vary with elevation.

Insect herbivores are ectothermic, and hence, their damage is expected to decrease with lower temperatures at higher elevation (Moreira et al., 2017; Rasmann et al., 2014). While many studied interactions support this expectation, a substantial minority show different patterns. In their review, Moreira et al. (2017) found that 22% of studies showed increasing insect damage with elevation and 8% showed peak damage at intermediate elevations. Data from additional insect–plant interactions are needed to clarify the conditions under which each pattern between herbivory and elevation occurs. For example, few studies have quantified elevational patterns in damage by endophagous insects (but see: Louda, 1982, 1983). Since internal-feeding insect herbivores develop surrounded by plant tissues, they may be buffered from elevational changes in abiotic conditions compared to free-feeding herbivores (Fernandes & Price, 1988; Kozlov et al., 2022).

While multiple studies have examined patterns of insect herbivory along elevation gradients, few have quantified how the effects of insect herbivory upon key components of individual plant fitness or population parameters vary with elevation (but see, e.g., Dostalek et al., 2018; Louda, 1982, 1983; Miller et al., 2009). If the "Species Interactions—Abiotic Stress" hypothesis holds, insect herbivory should limit plant fitness more under more benign conditions, where damage is predicted to be greater. However, explaining variation in herbivore effects on plant fitness requires integrating variation in insect feeding with variation in plant tolerance for herbivore damage.

The relationship between abiotic stress and plant tolerance for herbivore damage is complicated. Outcomes can depend on plant life-form, type of herbivore damage, and identity of the limiting stress (Hawkes & Sullivan, 2001;

Wise & Abrahamson, 2007). Contrary to predictions based on expected patterns of insect damage, Dostalek et al. (2018) found that folivorous insects limited *Salvia nubicola* population growth more at high elevations than at low elevations in the Himalaya Mountains. They attributed these results to greater tolerance by plants at more resource-rich lower elevations. Such results show the need for additional studies that integrate variation in insect herbivore damage with variation in plant tolerance by quantifying effects on key plant fitness components across elevations.

Even when insect herbivores and pre-dispersal seed predators limit plant seed production, subsequent seedling recruitment and, hence, population growth may not be limited by seed availability. Instead, ecological processes that act after seed dispersal, such as competition and granivory, may prevent viable seeds from germinating (Crawley, 1992; Harper, 1977). In their review, Turnbull et al. (2000) concluded that only about half of the studies that added seeds to plant populations produced an increase in plant population size.

The extent to which seedling recruitment is limited by seed supply versus post-dispersal processes likely reflects plant life history as well as ecological context. Biennial and short-lived, monocarpic perennial plants tend to have a tight link between seed production and seedling recruitment because often they lack clonal reproduction and seed banks (Louda & Potvin, 1995). With respect to ecological context, habitats with more exposed bare ground often are associated with greater seed limitation of seedling recruitment because such habitats allow better seed–soil contact and more light (Maron & Crone, 2006; Turnbull et al., 2000). In montane systems, elevation could mediate the extent to which seedling recruitment is seed-limited. For example, Miller et al. (2009), working in New Mexico, USA, showed that tree cholla (*Opuntia imbricata*) seedling recruitment was more seed-limited in lower elevation desert grasslands than in higher rocky mountain slopes.

Insect consumption of developing inflorescences and seeds is widespread among plant taxa and occurs in many ecosystems (Boaventura et al., 2022; Janzen, 1971; Ortiz et al., 2023). The ecological and evolutionary implications of damage to plant reproductive structures may be distinctive because these herbivores destroy gametes and seeds (Boaventura et al., 2022; Harper, 1977). Further, several experiments demonstrated large reductions in viable seed production due to insect feeding on developing inflorescences and seeds that, in turn, reduced plant population growth (Kelly & Dyer, 2002; Louda, 1982, 1983; Louda & Potvin, 1995; Rose et al., 2011). Nevertheless, Boaventura et al. (2022) conclude in their review that quantification of inflorescence damage and its fitness consequences remains rare under natural conditions.

Here, we report a multi-year experiment to evaluate spatial and temporal variation in insect floral herbivory and pre-dispersal seed predation along with its effects on both lifetime seed production and subsequent seedling recruitment for montane populations of the short-lived perennial, monocarpic, native Platte thistle (*Cirsium canescens*). We examine the role of ecological context in the outcome of the plant–insect interaction, specifically in relation to complex elevational variation. We asked: (1) Does insect flower head herbivory and pre-dispersal seed predation limit Platte thistle lifetime seed production? (2) Does insect flower head herbivory and pre-dispersal seed predation limit seedling recruitment? (3) To what extent does the ecological context, including spatial—especially elevational—and temporal variation, influence the outcome of these interactions?

Our montane Platte thistle populations and their insect floral herbivores have multiple features that make them relevant for addressing these questions. First, Platte thistle has a substantial elevation range, >650 m, in our region. Second, Platte thistle's short-lived, monocarpic life history suggests that seedling recruitment should be related to viable seed production. Third, the insects are primarily endophagous, allowing us to address the under-explored topic of how damage by internal-feeding insect herbivores varies with elevation. Finally, our locale in central Colorado, USA, represents the xeric western edge of Platte thistle's natural geographic range.

METHODS

Study species

Platte thistle (*Cirsium canescens* Nutt.) is a short-lived, monocarpic, perennial species that is native to grasslands on sand and gravel soils in the central United States. Its distribution is centered in the Sand Hills of Nebraska, extending west into Wyoming and Colorado (Kaul et al., 2007). It grows as juvenile rosettes for up to 11 years before bolting, flowering, and dying (Louda & Potvin, 1995). Flowering is determinate (Figure 1A). In our montane populations, flowering occurs mid-June to mid-August.

Platte thistle flower heads are used by a suite of native insect herbivores (Louda et al., 1990; Louda & Potvin, 1995). In our region, the most common insect species are two tephritid fly species (*Paracantha gentilis*, *Terellia* (*Orellia*) *occidentalis*) that oviposit into and feed upon the florets, ovules, and maturing seeds of developing flower heads (Figure 1B). These tephritids are characteristic specialists on *Cirsium* spp. in the western United States (D. H. Headrick, personal communication). Further, a

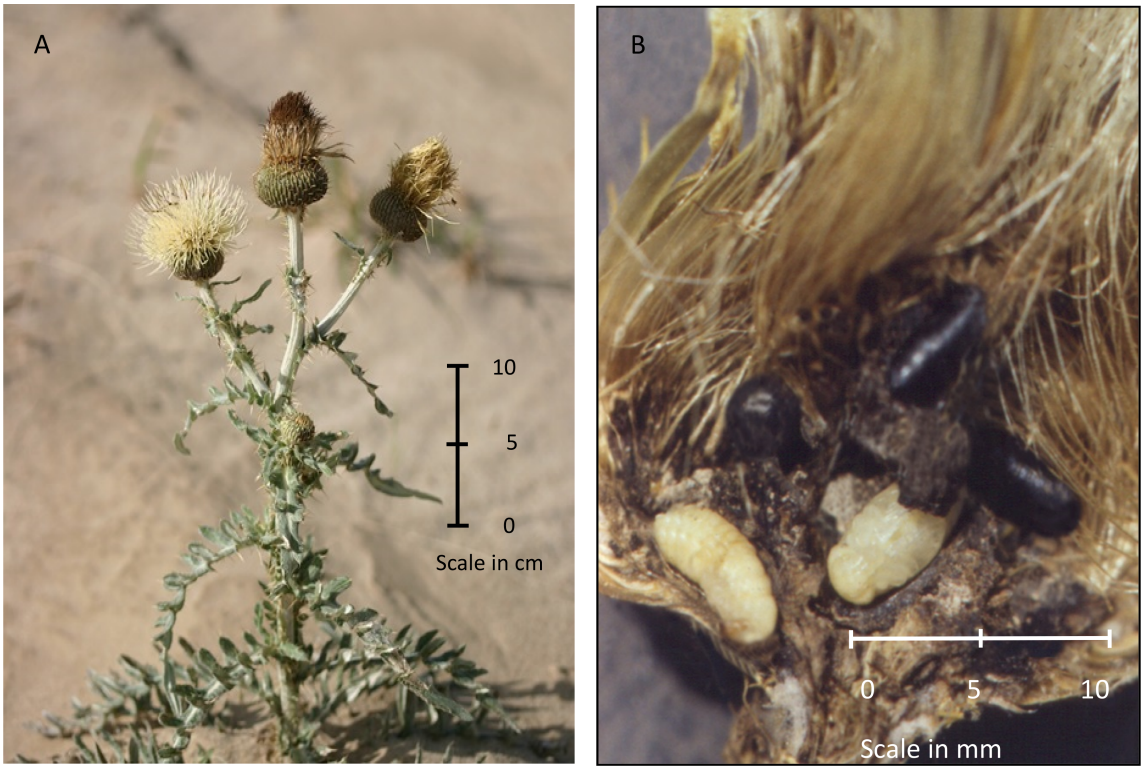


FIGURE 1 Examples of an (A) adult Platte thistle plant and (B) typical insect herbivore damage to developing Platte thistle flower heads. In (A), the reproductive stalk has two flower heads that are post-anthesis: one at anthesis and one in bud. (B) Internal insect feeding, with three black fly pupal cases (Tephritidae), two white *Rhinocyllus conicus* pupae (Curculionidae), and the typical destruction seen to florets, ovules, seeds, and receptacle base within the flower head. Photo credit: S.M. Louda.

polyphagous scarab beetle (*Euphoria inda*) frequently mines into flower heads at full anthesis and destroys ovules and developing seeds (Louda, 1998; S. M. Louda, personal observation).

Platte thistle flower heads in this area also are attacked by a Eurasian weevil, *Rhinocyllus conicus*. This weevil was introduced to North America for biological control of exotic thistles, but it also attacks native thistles (Gassmann & Louda, 2001; Louda, 1998; Louda et al., 1997). Inflorescence oviposition and feeding by *R. conicus* severely reduce both Platte thistle reproductive success (Louda, 1998; Louda et al., 1997) and population growth (Rose et al., 2005) in Great Plains grasslands.

Study sites

We collected data in montane Platte thistle populations in the upper Arkansas River Valley grasslands in Chaffee County, CO, at the western edge of the plant's natural distribution (McMinn et al., 2017). Mean annual temperature in Buena Vista, CO, near our sites, is 6.1°C (1906–2021: Northeast Regional Climate Center). Our years of data collection, 2017–2021, were warmer than average (8.4, 8.1, 6.4, 7.5, and 7.6°C, from 2017 to 2021, respectively). Mean January–June precipitation, the months preceding Platte thistle seedling establishment and seed production, is 103 mm. January–June precipitation was above average in 2017 (106 mm), 2019 (161 mm), and 2021 (158 mm) but below average in 2018 (46 mm) and 2020 (42 mm).

The vegetation of the upper Arkansas River Valley is a mixture of sagebrush scrub and montane grassland. Platte thistle occurs in open gravelly, sandy grasslands and disturbed areas, such as washes and roadsides.

Five study sites were established in 2017, varying in elevation from 2435 to 2776 m (Table 1). From lowest to highest, they are as follows: Site A: Centerville (38°40′55.85″ N, 106°5′13.94″ W); Site B: Brown's Creek (38°41′39.46″ N, 106°7′15.03″ W); Site C: Lost Creek (38°45′29.85″ N, 106°9′26.14″ W); Site D: Forman (38°53′56.68″ N, 106°12′22.75″ W); and Site E: Forest Service (38°49′32.08″ N, 105°59′40.30″ W). Site A is close to the lower elevation limit observed for Platte thistle in the upper Arkansas River valley (2298 m); sparse plant numbers precluded use of lower sites. The highest observed elevation for Platte thistle here is 2954 m; however, extensive *R. conicus* invasion precluded use of the highest sites. Hereafter, we refer to sites as A through E from lowest to highest. Soils at our study sites are

TABLE 1 Abiotic and biotic site characteristics for our five study sites.

Site letter and name	Elevation (m)	Side of valley	Soil moisture (% water by volume)	Soil compaction (Mg/m^2)	Net primary production (g/m^2)	Bare ground (%)	First flowering (days from Jan 1)	Late flowering (% heads, at final census)
Site A, Centerville	2435	East	9.98 (0.72)^{A}	13.08 (1.86)^{A}	104.18 (16.07)	36.97 (3.76)^{A,B,C}	168.4 (0.91)^{A}	1.62 (0.88)
Site B, Brown's Creek	2502	West	5.71 (0.44)^{B}	6.64 (1.17)^{B}	138.04 (19.0)	22 (2.76)^{A}	168.9 (1.33)^{A,B}	1.70 (0.65)
Site C, Lost Creek	2577	West	4.65 (0.69)^{B,C}	13.87 (2.83)^{A}	99.82 (18.51)	44.78 (3.54)^{B,C}	174.6 (1.76)^{B}	2.78 (1.14)
Site D, Forman	2665	West	3.59 (0.59)^{C}	8.4 (1.36)^{A,B}	83.26 (12.71)	58 (5.63)^{B}	173.4 (2.07)^{C}	3.15 (0.83)
Site E, Forest Service	2776	East	10.95 (0.68)^{A}	3.51 (0.49)^{B}	121.18 (20.09)	28.97 (4.62)^{A,C}	172.0 (2.29)^{C}	3.47 (1.24)

Note: Details of methods and statistical analyses are present in Appendix S1. Sites are listed from lowest to highest elevation. Means (SE) are shown where relevant. For each dependent variable (column), statistically significant differences among sites are indicated by different superscript letters.

gravelly sandy loams and fine sandy loams (Soil Survey Staff, Natural Resource Conservation Service, n.d.).

To describe the ecological context of our study sites, we quantified soil and vegetation variables at each site (Appendix S1: Methods and results); we summarize the patterns here. Soil moisture and bare ground cover varied among sites but were unrelated to elevation. Soil moisture ranged from 3.59% (Site D) to 10.95% (Site E) water by volume. Bare ground cover varied from 22% (Site B) to 58% (Site D). Annual net primary productivity was not related to elevation but varied from 83.21 g/m^2 (Site D) to 138.04 g/m^2 (Site B). Soil compaction decreased with elevation. Soil penetrometer force ranged from 13.87 Mg/m^2 (Site C) to 3.51 Mg/m^2 (Site E). Therefore, site contexts varied but not consistently with elevation.

Experimental approach

To quantify the effect of insect floral herbivory and pre-dispersal seed predation on both lifetime seed production and on subsequent seedling establishment, we imposed experimental manipulations in 2017, 2018, and 2019. We counted new seedlings from experimental plants from 2018 through 2021.

In May 2017–2019 at each site, we chose undamaged rosettes that were initiating bolting. We divided plants at each site into two subsets each year: one subset was used in an experiment to quantify viable seed production, and the other subset was used in an experiment to record subsequent seedling establishment around maternal plants. To avoid overlap in seed dispersal shadows among plants used to quantify seedling establishment, these plants could not have another bolting individual within 4 m. We chose this distance because Louda and Potvin (1995) found that 90% of Platte thistle seedlings established <2 m from the maternal plant. Therefore, 4 m (2-m radius + 2-m radius) separating experimental plants largely prevented overlap of their seed shadows.

Treatments

Both the experiment examining Platte thistle viable seed production and the experiment examining seedling establishment had factorial designs. The three crossed independent variables in each experiment were insect herbivore reduction treatment, site, and year.

At each site, we randomly assigned plants in each experiment to one of three levels of the insect herbivore reduction treatment: insecticide spray, water-only spray (water control), and no spray (unmanipulated control). We initially included the "water control" and "unmanipulated control" treatment levels to evaluate whether water added to plants in spraying insecticide affects plant performance. We found that water control and unmanipulated control plants did not differ significantly in key plant performance variables (Appendix S2). Further, when we repeated analyses of treatment effects on seed production per plant and on seedling establishment per plant with and without controls pooled, the results were unchanged. Therefore hereafter, to improve sample sizes for the treatment contrast, we pool "water control" and "unmanipulated control" into a single "control" treatment level.

For insecticide, initially we used Orthene (Ortho), following Louda et al. (1990). However, Orthene did not reduce insect damage. So, in June 2017, we replaced it with Bifen IT (bifenthrin; Control Solutions) and diluted 16 mL insecticide per 1 L water, as recommended. Bifenthrin is neither phytostimulatory nor phytotoxic

(Carson & Root, 1999). In early 2018, we tried to reduce insect herbivory further by also spraying early flower head buds with Tanglefoot (Tanglefoot Co.). Unexpectedly, this killed those flower heads. Thus, from June 2018, we applied only Bifen IT. We applied insecticide using a handheld sprayer. We did not spray flower heads at anthesis, and we found no evidence that insecticide application impaired pollination. Specifically, in dissections of undamaged flower heads 2017–2019, the mean (SE) proportions of ovules that produced viable seed for insecticide-treated ($n = 275$ heads) and control heads ($n = 96$ heads) were 0.528 (0.018) and 0.493 (0.031), respectively.

In 2017 and 2018, to quantify seed production, we treated seven plants with insecticide and had three control plants at each site. To quantify seedling establishment, we treated seven plants with insecticide and had five control plants at each site. Given logistical constraints, we allocated more replicates to the treatment level we expected to be more variable—the insecticide-treated plants.

In 2019, we increased sample sizes and tried to equalize plants among treatment levels. Sites A, B, and D had 10 insecticide plants and 10 control plants in each experiment. At Sites C and E, bolting rosettes were less abundant; so sample sizes in each experiment were 9 insecticide and 9 control plants at Site C and 8 insecticide and 7 control plants at Site E.

Across all sites and years, thus, there were 117 insecticide plants and 76 control plants in the seed production experiment, and 117 insecticide plants and 96 control plants in the seedling establishment experiment. Final sample sizes at the end of growing seasons differed slightly from the numbers of tagged plants due to unnatural causes of plant mortality, such as crushing by vehicles or cattle. Appendix S3 presents final sample sizes in all combinations of the three independent variables (insect reduction treatment, year, and site).

Field measurements

We measured flowering phenology of Platte thistle at our study sites 2017–2019, as one estimate of plant response to elevational variation. Plants in both experiments were checked for flowering weekly starting in May. For early-season flowering phenology, we recorded the date that the first head opened on each plant. For late-season flowering phenology, we recorded the number of flower heads per plant still flowering in early August. Then, we calculated the proportion of flower heads per plant that were flowering in August.

To quantify plant size, we measured flowering stem height. To measure flowering effort, we counted the number of heads in each of three developmental stages (Appendix S4: Table S1): flowering heads (florets exerted), large flower head buds (≥12 mm diameter), and small flower head buds (>2 mm diameter) in monthly censuses. To quantify herbivore damage, we recorded the number of heads with external evidence of feeding in each developmental stage. Common forms of externally apparent insect damage included enveloped flower heads, oviposition scars, unequally exerted florets, loose pappus with frass, and holes in flower heads. The first three damage types indicate tephritid fly oviposition into the developing flower head.

To further quantify development, damage, and seed production, we examined a subset of the main seed-producing flower heads more intensively. These heads were identified using a numbering system based on flower head position, as in previous studies of Platte thistle (Louda & Potvin, 1995; West & Louda, 2021). The apical flower head was identified as 100, and terminal heads on successively lower lateral branches were 200, 300, 400, etc. Axillary flower heads, within each lateral branch, were identified as 210, 220, etc. from the 200 terminal toward the main stem and 310, 320, etc. from the 300 terminal in, and so forth. Intensive data were taken on alternating terminal heads (100, 300, 500, and 700) and on a prominent axillary head (310) in the first week of June, July, and August in each year. These data included diameter, developmental stage, and damage score (defined in Appendix S4: Table S2) consistent with previous studies (above). As each head finished flowering, we covered it with organza fabric to prevent seed dispersal.

For plants used to quantify seed production, we collected bagged heads once seed dispersal had started inside the bag. We dissected each collected head, recording the total number of florets initiated (if possible), the number of viable seeds produced, evidence of insects, and the percentage of the receptacle base damaged (Appendix S4: Table S3). Viable seeds were caramel-to-tan colored, extended and plump, and without evidence of damage.

For plants used to quantify seedling establishment, we sampled production of viable seeds non-destructively in situ since matured flower heads needed to remain on the plant. After seed dispersal had started inside the bag, we removed the bag, pulled a sample of the emerging seeds out, and visually assessed damage and proportion of viable seeds. We then re-inserted the seeds into the flower head to allow natural dispersal. With hindsight, these estimates focused on the central part of a flower head, and they produced significantly higher estimates of seed production than those from comparable dissected

flower heads. So, these data were used only to provide a field-based, head-specific adjustment of the dissection estimate. These adjustments were informative in cases where significant damage or lack of damage was observed for a specific flower head in the field censuses.

Estimating seed produced per plant

For the plants used to quantify seed production, we calculated the total number of viable seeds produced by a plant as the sum of the numbers of viable seeds produced by all heads that flowered. This sum included the numbers of viable seeds counted in the head dissections plus the estimated number of viable seeds produced by the few heads that dispersed before they could be bagged. We estimated viable seed production by flower heads that dispersed before bagging using the position of the flower head within the plant, the level of insect damage recorded for the head in monthly censuses, and head dissection data for that head position (Appendix S5). Estimates of maximum potential seed production for each flower head position were based on the dissections of both our experimental flower heads and of heads in a long-term observational study on Platte thistle seed production in this region (2006–2020: S. M. Louda and F. L. Russell, unpublished data).

For the plants used to quantify seedling establishment, we estimated the total number of viable seeds produced by each plant using field census data, field visual estimates near the end of the season, and dissection data of heads from plants in the parallel seed production experiment in each year. Appendix S5 describes in detail the steps used in calculating viable seed production; the steps are summarized as follows.

First, since extensive insect feeding precluded a direct count of the number of seeds initiated and the number of seeds destroyed in most flower heads, we used the data available to estimate both parameters. To estimate the number of seeds initiated by head position, we used the dissection data from this study and from the long-term observational study on Platte thistle reproduction in our region to determine the mean maximum number of florets, and so potential seeds, observed for each flower head position.

To estimate the viable seed produced by each undissected head, we multiplied the potential number of seeds (above) by the observed proportion of viable seed for a head in that position in the dissection data. When both the field visual inspection estimate and the dissection-based estimate of mean proportion of viable seeds in flower heads in that position were available, the proportion of seeds viable in that head was the average of those two estimates. To obtain viable seed production for each plant, we summed the estimated number of viable seed for all heads that flowered on the plant as recorded in the monthly census data.

In 2018, estimating viable seed production per plant was complicated by the Tanglefoot spray damage to early flower head buds on insecticide-treated plants (range: 1–9 buds/plant, mean (SE) = 3.1 (0.20) buds/plant). To provide data on seed production by these heads, we tagged four replacement adult Platte thistles per site in June and began spraying them with insecticide. Since these plants were later-developing and had suffered some flower head herbivory before we began spraying them, they provide a conservative estimate of the mean viable seed production for each flower head position in the 2018 insecticide treatment for between-year comparisons.

Seedling counts

We quantified seedling emergence around the carcasses of the experimental plants that we allowed to disperse seeds. We counted newly emerged Platte thistle seedlings with a 2-m radius around each maternal plant for two seasons after the plant dispersed seeds, that is, in 2020 and 2021 for the 2019 experiment. We counted seedlings at two-week intervals: May–August in 2018, 2019, 2020, and 2021. Seedlings are distinctive, with cotyledons, tiny size, and initial leaves with rounded leaf margins. We used the total number of seedlings counted over two seasons to estimate the plant's seedling recruitment. We calculated three recruitment parameters: the proportion of plants that established at least one seedling, the total number of seedlings per plant for those that recruited any seedlings, and the average number of viable seeds released per seedling recruited per plant, an estimate of the difficulty of seedling establishment. We found no evidence of a soil seed bank for this species, similar to Louda and Potvin (1995).

Statistical analyses

We used ANOVA to evaluate the effects of explanatory variables upon Platte thistle flowering phenology, insect damage, viable seed production, and seedling establishment. Fixed effects included insect herbivory treatment, site, and year. To examine patterns related to elevation, we evaluated the relationship between the mean value for a dependent variable at a site and the site's elevation, examining both insecticide-treated and control plants, using ANCOVA. We transformed dependent variables to meet ANOVA assumptions. We used Tukey tests for post

hoc comparisons. Analyses were conducted in R 4.2.1, including the lmerTest (Kuznetsova et al., 2017), MuMIn (Barton, 2009), and emmeans packages (Lenth, 2024).

For phenology and damage, we evaluated performance under ambient conditions, so we analyzed only plants in the control treatment level in both experiments. We quantified flowering phenology as date of first flowering and proportion of flower heads still flowering in early August. We quantified insect herbivore damage using proportion of damaged seeds (1 − proportion viable) per flower head and proportion of damaged flower heads per plant. We applied angular transformations to all proportions. The independent variables were site and year. Analyses of proportion of damaged seeds per flower head included "plant" as a random effect. For damage and late season flowering phenology, samples size of control plants for individual sites were 28–37 plants and for individual years were 41–91 plants. For date of first flowering, sample sizes were smaller (sites: 16–37 plants, years: 34–56 plants) because we recorded flowering dates only if flowering appeared to have begun within 2–3 days before our visit. We did not examine two-way interaction effects for these variables because sample sizes of control plants in some site × year combinations were too limited.

We evaluated the effects of herbivore treatment, site, and year and the two-way interactions upon mean number of viable seeds produced per plant (square-root transformed), probability that a plant succeeded in recruiting at least one seedling, mean number of seedlings recruited per successful parent plant (log transformed), and mean rate at which viable seeds produced seedlings (log transformed). We used logistic regression to analyze the probability that a plant recruited at least one seedling. We used ANOVA/ANCOVA to analyze the other dependent variables. Analyses of viable seeds per plant were based on data from plants in both experiments 2017–2019. Analyses of seedling establishment were based solely on the experiment that had natural seed dispersal. Sample sizes for the two-way interactions varied from 21 to 94 plants for seed production and from 8 to 38 plants for seedlings established. We did not evaluate three-way interaction effects because of limited sample sizes for three-way combinations.

To examine the relationship between the availability of bare ground and seedling recruitment, we conducted an ANCOVA with mean bare ground cover at a site and insect herbivore reduction treatment as independent variables. The dependent variable was the mean viable seeds released per seedling recruited at a site.

To further explore the extent to which seedling recruitment was seed-limited versus site-limited, we examined the relationship between the number of seedlings recruiting per plant (log-transformed) and viable seed production per plant, using mixed-model ANCOVA. We included a quadratic term, the square of the number of viable seeds produced per plant, to investigate non-linearity in the seed–seedling relationship. Non-linearity would arise if post-dispersal processes limit seedling recruitment, when increasing seeds released would have a decreasing effect upon seedlings established. In addition to viable seed production per plant and the quadratic term, fixed effects included insect herbivore treatment and two-way interactions among these variables. Site and year were random effects.

RESULTS

Flowering phenology

Date of first flowering varied among sites ($F_{4,123} = 14.25$, $p < 0.001$; Table 1). First flowering was earlier at the two lowest sites: Site A relative to Sites C, D, and E and Site B relative to Sites D and E. All three years differed in the mean date of first flowering, with 2018 earliest (June 12), 2017 intermediate (June 18), and 2019 latest (June 27). In spite of early flowering at the lowest sites, first flowering was not significantly related to site elevation.

The mean proportion of heads still flowering in early August did not differ significantly among sites, although it increased with site elevation ($F_{1,3} = 21.98$, $p = 0.018$; Table 1). Also, years differed ($F_{2,169} = 5.15$, $p = 0.007$). In 2018, a higher proportion of flowering heads was late (mean [SE] 0.048 [0.011]) than in 2017 (0.010 [0.004]) or in 2019 (0.021 [0.006]).

Ambient levels of insect floral and pre-dispersal seed damage

Examining plants exposed to ambient levels of insect herbivory (control plants), 87.2% of florets and seeds per flower head across all sites and years were damaged. However, we found pronounced spatial and temporal variation in levels of insect damage to flower heads (Table 2, Figures 1 and 2A). The proportion of damaged ovules/seeds per flower head varied significantly among sites ($F_{4,154} = 2.52$, $p = 0.043$; Table 2A) and among years ($F_{2,181.07} = 16.85$, $p < 0.001$). The lowest Site (A) had a greater proportion of seeds damaged per flower head than did Site D. Among years, 2019 averaged a significantly higher proportion of damaged seeds per flower head than did either 2017 or 2018, while 2017 and 2018 did not differ from one another. There was a trend toward lower mean proportions of seeds damaged per flower head at higher sites

TABLE 2 Spatial and temporal variation in ambient levels of seed damage (on control plants), both (A) proportion of seed damaged per flower head and (B) proportion of flower heads with external evidence of insect feeding per plant (mean with SE in parentheses).

	Year			
Site (low to high)	**2017**	**2018**	**2019**	**All years**
(A) Proportion of seed damaged/head				
Site A, Centerville	0.86 (0.031)	0.86 (0.029)	0.94 (0.01)	0.91 (0.010)^{A}
Site B, Brown's Creek	0.77 (0.032)	0.84 (0.023)	0.89 (0.014)	0.86 (0.012)^{A,B}
Site C, Lost Creek	0.86 (0.028)	0.85 (0.027)	0.90 (0.012)	0.88 (0.011)^{A,B}
Site D, Forman	0.76 (0.030)	0.85 (0.027)	0.86 (0.017)	0.84 (0.013)^{B}
Site E, Forest Service	0.82 (0.031)	0.87 (0.026)	0.88 (0.021)	0.86 (0.015)^{A,B}
All sites	0.81 (0.014)^{A}	0.85 (0.012)^{A}	0.90 (0.006)^{B}	
(B) Proportion of heads damaged/plant				
Site A, Centerville	0.52 (0.045)	0.66 (0.077)	0.79 (0.028)	0.70 (0.009)^{A,B}
Site B, Brown's Creek	0.55 (0.066)	0.64 (0.07)	0.82 (0.041)	0.72 (0.012)^{A,B}
Site C, Lost Creek	0.60 (0.038)	0.86 (0.044)	0.84 (0.028)	0.79 (0.011)^{A}
SD, Forman	0.48 (0.029)	0.70 (0.097)	0.70 (0.037)	0.64 (0.013)^{B}
Site E, Forest Service	0.68 (0.072)	0.77 (0.058)	0.64 (0.078)	0.69 (0.015)^{A,B}
All sites	0.56 (0.024)^{A}	0.72 (0.034)^{B}	0.77 (0.019)^{B}	

Note: Different letters in superscript indicate statistically significant differences between years and, separately, between sites. Site × year interaction effects were not tested due to sample size constraints.

($t = -1.736$, $p = 0.181$), but this trend did not reach statistical significance.

Across all sites and years, 70.9% of flower heads on control plants showed external evidence of insect damage. Among sites, the proportion of flower heads with external evidence of damage varied marginally significantly ($F_{4,166} = 2.2$, $p = 0.071$; Table 2B). Among years, the proportion of damaged flower heads per plant varied significantly ($F_{2,166} = 15.53$, $p < 0.001$; Figure 2A), being lower in 2017 than in 2018 or in 2019, which did not differ from each other. The mean level of externally apparent damage to flower heads was unrelated to elevation.

Viable seed production

Insecticide application reduced insect damage (Appendix S2). Across all sites and years, insecticide decreased the proportion of damaged ovules and seeds per flower head by 22.6%, as well as the proportion of flower heads with external evidence of damage per plant by 44.9%. Insecticide efficacy did not vary strongly among sites; however, it varied between years with least reduction in damage in 2017, intermediate reduction in 2018, and greatest reduction in 2019.

Flower head-feeding insects strongly reduced Platte thistle lifetime seed production and did so consistently across all sites (Table 3). By contrast, the magnitude of the reduction in viable seed production due to insect floral herbivory and pre-dispersal seed predation varied significantly among years (herbivory × year: $F_{2,388} = 10.32$, $p < 0.001$; Figure 2B). As compared to insecticide-treated plants, ambient levels of insect damage on control plants reduced lifetime seed production by 42.8%, 62.4%, and 68.7% in 2017, 2018, and 2019, respectively (Table 4).

Sites differed in viable seed production per plant, but those differences were not consistent across years (site × year: $F_{8,384} = 4.87$, $p < 0.001$; Table 5). In 2017, plants at Sites B and D had greater viable seed production than those at Sites A and C. In 2018, plants at Sites C and D had greater viable seed production than those at Site A or B. In 2019, plants did not differ significantly among sites.

Site mean value for viable seed production per plant was unrelated to elevation. There was no significant interaction between site elevation and insecticide treatment upon the site mean for viable seeds produced per plant.

Seedling recruitment

Failure to recruit

Of 210 total Platte thistles, 26 plants (12.4%) failed to recruit any seedlings, despite 24 of these 26 thistles producing viable seeds in their one, lifetime reproductive

event. Logistic regression of the probability that an adult Platte thistle produced at least one seedling showed no significant effect of insect herbivory treatment. However, the site × year interaction effect was significant, reflecting a near significant effect of site in 2017 (df = 4, LRT = 9.38, $p = 0.052$) but no effect of site in 2018 or 2019.

We found a trend toward higher proportions of thistles producing at least one seedling at higher elevations ($F_{1,7} = 2.46$, $p = 0.161$). There was no significant insect herbivory × site elevation interaction.

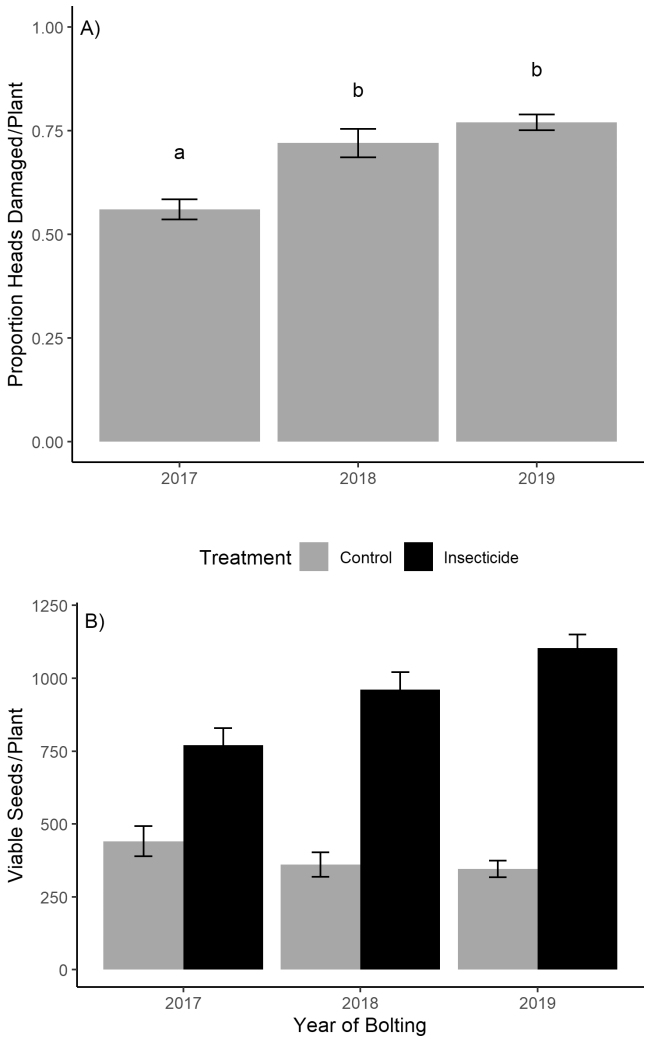


FIGURE 2 (A) Proportion of damaged flower heads on control plants, those exposed to ambient levels of flower head insect herbivory and pre-dispersal seed predation, by year. (B) Viable seeds per plant for each treatment level in each year. Error bars represent SEs.

Seedling recruitment success

Unexpectedly, insecticide treatment did not significantly increase the number of seedlings recruited per successful Platte thistle plant (Tables 6 and 7). The number of seedlings recruited per plant varied significantly among sites ($F_{4,162} = 9.41$ $p < 0.001$; Table 6). Plants at Site B had significantly more seedlings recruited than did those at Site A, D, or E. Also, more seedlings recruited per plant at Sites C and D than at Site A. The number of seedlings recruited per Platte thistle varied significantly among years ($F_{2,162} = 8.64$, $p < 0.001$; Table 7). The 2018 experiment cohort recruited significantly more seedlings than did the 2019 cohort. The 2017 experiment cohort produced marginally significantly more seedlings than did the 2019 cohort (Table 7). The interaction of site × year was marginally significant ($F_{8,162} = 1.82$, $p = 0.077$; Table 7).

We did not find a significant effect of elevation upon the site mean for the number of seedlings recruited per Platte thistle. Similarly, there was no significant interaction effect between insect herbivory treatment and site elevation.

Difficulty of seedling establishment

The number of viable seeds dispersed per seedling established was 122.1% greater for insecticide-treated plants than for control plants ($F_{1,162} = 37.7$, $p < 0.001$; Table 8). The number of viable seeds required for each seedling varied significantly among sites ($F_{4,162} = 6.00$, $p < 0.001$; Table 8). Plants at Sites A and D required more

TABLE 3 Viable seed produced per plant, showing effect of reducing flower head insect herbivory by site (mean with SE in parentheses).

	Site, from low to high elevation					
Treatment	Site A, Centerville	Site B, Brown's Creek	Site C, Lost Creek	Site D, Forman	Site E, Forest Service	All sites
Control	266.6 (31.40)	446.8 (47.46)	336.6 (44.35)	463.2 (58.82)	339.4 (53.40)	371.7 (21.84)^{A}
Insecticide	819.7 (72.00)	977.6 (73.77)	952.0 (74.53)	1127.0 (63.43)	923.7 (73.84)	961.3 (32.39)^{B}
Both treatments	572.4 (51.61)^{A}	750.1 (54.77)^{B,C}	690.4 (57.79)^{A,B}	838.0 (56.62)^{C}	696.5 (59.85)^{A,B}	

Note: Different letters in superscript represent statistically significant differences between levels of the flower head insect herbivory reduction treatment and, separately, between sites.

TABLE 4 Viable seed produced per plant, showing the effect of reducing flower head insect herbivory by year (mean with SE in parentheses).

	Experiment year			
Treatment	**2017**	**2018**	**2019**	**All years**
Control	440.9 (52.34)^{A}	361.1 (42.06)^{A}	345.3 (28.26)^{A}	371.7 (21.84)
Insecticide	770.6 (58.24)^{B}	960.5 (60.01)^{B,C}	1103.9 (46.59)^{C}	961.3 (32.39)
Both treatments	648.8 (44.05)	737.1 (49.21)	730.7 (39.13)	

Note: Different letters in superscript represent statistically significant differences among combinations of the flower head insect herbivory reduction treatment and experimental year because there are statistically significant interaction effects between these independent variables.

TABLE 5 Viable seed produced per plant by year and site (mean with SE in parentheses).

	Site, from low to high elevation					
Year	**Site A, Centerville**	**Site B, Brown's Creek**	**Site C, Lost Creek**	**Site D, Forman**	**Site E, Forest Service**	**All sites**
2017	430.3 (60.07)^{A}	901.8 (120.05)^{B}	415.8 (66.33)^{A}	845.6 (91.87)^{B}	641.7 (98.87)^{A,B,C}	648.8 (44.05)
2018	442.7 (57.84)^{A,C}	504.7 (58.25)^{A,C}	887.0 (80.66)^{B}	840.3 (114.68)^{B}	689.9 (106.88)^{A,B,C}	737.1 (49.21)
2019	670.9 (91.36)^{A,B,C}	790.0 (83.0)^{B,C}	691.6 (91.57)^{A,B,C}	762.3 (77.49)^{B,C}	738.2 (99.18)^{A,B,C}	730.7 (39.13)
All years	572.4 (51.61)	750.1 (54.77)	690.4 (57.79)	838.0 (56.62)	696.5 (59.85)	

Note: Differing superscript letters indicate statistically significant differences among combinations of experimental years and sites because there are statistically significant interaction effects between these independent variables.

TABLE 6 Number of seedlings established per maternal plant, showing effects of reducing flower head insect herbivory at each site (mean with SE in parentheses).

	Site, from low to high					
Treatment	**Site A, Centerville**	**Site B, Brown's Creek**	**Site C, Lost Creek**	**Site D, Forman**	**Site E, Forest Service**	**All sites**
Control	2.5 (0.39)	12.2 (2.26)	10.0 (1.77)	7.6 (2.04)	7.7 (1.52)	8.1 (0.84)
Insecticide	4.6 (1.06)	14.9 (3.65)	10.7 (3.33)	7.5 (1.26)	7.7 (1.89)	9.3 (1.17)
Both treatments	3.6 (0.63)^{A}	13.8 (2.25)^{B}	10.4 (1.98)^{B,C}	7.5 (1.10)^{C}	7.7 (1.22)^{A,B,C}	

Note: Seedlings were counted within a 2-m radius of each maternal plant for two years following the year of seed production. Means are based on maternal plants that produced at least one seedling. Different letters in superscript indicate statistically significant differences between sites.

TABLE 7 Numbers of seedlings established per maternal plant, showing temporal variation in seedling establishment at each site (mean with SE in parentheses).

	Experimental year			
Site	**2017**	**2018**	**2019**	**All years**
Site A, Centerville	2.9 (0.61)	3.7 (1.24)	4.0 (1.11)	3.6 (0.63)^{A}
Site B, Brown's Creek	17.1 (3.09)	21.0 (5.63)	5.3 (1.00)	13.8 (2.25)^{B}
Site C, Lost Creek	8.7 (2.68)	13.2 (2.89)	9.8 (3.76)	10.4 (1.98)^{B,C}
Site D, Forman	8.2 (2.54)	10.4 (2.19)	5.1 (0.98)	7.5 (1.10)^{C}
Site E, Forest Service	7.2 (1.55)	9.7 (2.46)	6.3 (2.30)	7.7 (1.22)^{A,B,C}
All sites	9.1 (1.20)^{A,B}	12.1 (1.70)^{A}	6.1 (0.99)^{B}	

Note: Seedlings were counted within a 2-m radius of each maternal plant for two years following the year of seed production. Means are based on maternal plants that produced at least one seedling. Different letters in superscript indicate statistically significant differences between years and, separately, between sites.

viable seeds per seedling than did plants at Sites B and C. The number of viable seeds required per seedling also varied significantly among years ($F_{2,162} = 13.08$, $p < 0.001$; Table 9). The 2019 experiment plants required more viable seeds per seedling than did the 2017 and 2018 experiment plants, which did not differ. No two-way interaction effects were significant.

The site mean number of viable seeds required to produce a seedling was unrelated to elevation. The interaction of elevation and insecticide treatment was not significant.

Finally, across the five sites, the mean number of viable seeds per seedling was uncorrelated with the mean percent area of bare ground at a site (Figure 3).

Relationship between seed input and seedling recruitment

The relationship between viable seeds released and seedlings recruited per plant was not proportional. Specifically, seedling recruitment per plant increased, but at a decelerating rate, as viable seed production per plant increased ($F_{1,198.6} = 8.49$, $p = 0.004$; Figure 4; Appendix S6). This decelerating relationship is described by a significant quadratic term (square of number of viable seeds produced per plant; $F_{1,197.2} = 4.03$, $p = 0.046$).

Insecticide reduction of floral herbivory did not have a significant effect upon the number of seedlings that established per plant. Visual inspection of the relationships between seed production and seedling recruitment per plant suggests slope and curvature differences between the control versus insecticide treatments. However, tests of two-way interaction effects upon the number of seedlings established per plant were not significant, neither the interaction between treatment and number of viable seeds per plant ($F_{1,197.4} = 2.16$, $p = 0.144$) nor the interaction between treatment and number-squared viable seeds per plant ($F_{1,197.3} = 2.59$, $p = 0.109$).

One insecticide-treated plant that bolted in 2018 was an outlier in the relationship between viable seed produced and number of seedlings recruited (Figure 4), producing 79 seedlings from only an estimated 928 seeds. However, removing the outlier did not change any results.

DISCUSSION

Our results show that flower head-feeding insect herbivores strongly suppress Platte thistle lifetime viable seed production in montane grasslands at the western edge of the species' biogeographic range. Yet, increased seed production with insecticide application did not increase seedling recruitment proportionally. Environmental context, specifically variation among years, affected the amount of insect damage to Platte thistle flower heads and the magnitude of insect herbivores' suppression of

TABLE 8 Number of viable seeds released per plant for each seedling established in the next generation showing the effect of flower head insect herbivory across sites (mean with SE in parentheses).

	Site, from low to high					
Treatment	Site A, Centerville	SB, Brown's Creek	Site C, Lost Creek	Site D, Forman	Site E, Forest Service	All years
Control	143.0 (21.35)	103.3 (45.71)	77.2 (24.12)	173.4 (43.07)	80.5 (24.24)	114.5 (15.32)^{A}
Insecticide	345.7 (79.83)	180.0 (50.55)	154.2 (22.66)	328.8 (66.54)	274.7 (60.14)	254.4 (26.61)^{B}
Both treatments	253.6 (47.49)^{A}	146.6 (34.90)^{B}	119.8 (17.48)^{B}	265.1 (44.34)^{A}	186.0 (37.86)^{A,B}	

Note: The dependent variable, mean number of viable seeds produced per seedling established, is a measure of seedling establishment difficulty. Different letters in superscript indicate statistically significant differences between levels of the flower head insect herbivory reduction treatment and sites.

TABLE 9 Number of viable seeds released per plant for each seedling established in the next generation showing the effect of flower head insect herbivory across years (mean with SE in parentheses).

	Experimental year			
Treatment	2017	2018	2019	All years
Control	103.1 (23.31)	61.1 (14.71)	153.2 (28.06)	114.5 (15.32)^{A}
Insecticide	217.0 (40.89)	180.2 (39.42)	347.6 (50.42)	254.4 (26.61)^{B}
Both treatments	172.8 (27.34)^{A}	130.7 (25.04)^{A}	251.7 (30.99)^{B}	

Note: The dependent variable, mean number of viable seeds produced per seedling established, is a measure of seedling establishment difficulty. Different letters in superscript indicate statistically significant differences between levels of the flower head insect herbivory reduction treatment and years.

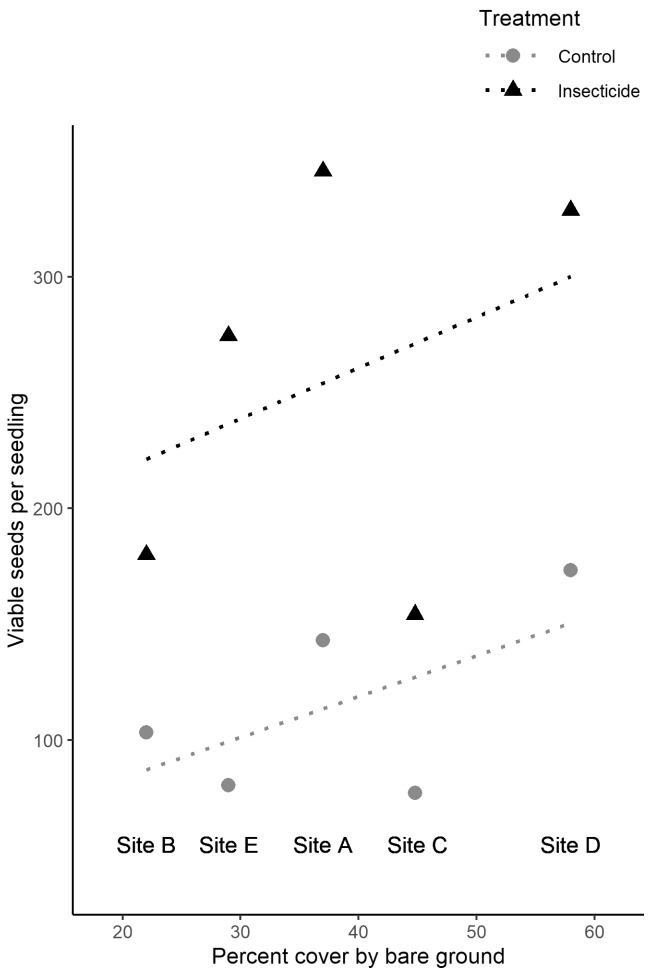


FIGURE 3 Relationship between the mean number of viable seeds required to produce a seedling at a site (difficulty of establishment) and mean cover by bare ground at that site. Dotted lines are used to illustrate trends that were short of statistical significance. Relationships between seeds per seedling versus percent bare ground are shown separately for control plants and for insecticide-treated plants at each site. Higher ratios of seed input to seedlings established suggest greater limitation of recruitment by post-dispersal ecological filters.

viable seed production. Elevation did not drive variation in the insect herbivore–Platte thistle interaction.

Insect floral herbivory and pre-dispersal seed damage limit lifetime seed production

Consistent with previous studies on thistles (*Cirsium* spp.) (Louda et al., 1990; Louda & Potvin, 1995; Maron et al., 2002; Palmisano & Fox, 1997; Rose et al., 2011) and other Asteraceae (Kelly & Dyer, 2002; Louda, 1982, 1983), inflorescence insect herbivory and destruction of developing seed substantially reduced viable seed matured by Platte thistle in these peripheral montane populations. These reductions in lifetime seed production are important because, even in microsite-limited plant populations, they likely represent reduced maternal fitness. In plant populations where microsite availability limits seedling establishment, plants that produce more seeds have more opportunities to colonize the limited number of suitable microsites.

Reduced viable seed production reflects intense insect damage to flower heads in these populations at the periphery of Platte thistle's biogeographic range, and that finding contradicts expectation based on theory. Metapopulation theory (Hanski & Ovaskianen, 2000) and empirical results (Fletcher et al., 2018) suggest that landscapes with more isolated host populations might be expected to support smaller abundances of specialist natural enemies (Alexander et al., 2007). We know that Platte thistle populations near the range edge, including in our region, are less frequent than in interior portions of the range (McMinn et al., 2017). Further, severe insect damage to Platte thistle in the Upper Arkansas River Valley does not reflect the availability of alternate hosts; no other *Cirsium* species that would sustain oligophagous thistle feeders are common in these montane grasslands (S. M. Louda, personal observation).

Two hypotheses may explain intense specialist herbivore damage to Platte thistle at the edge of its range. First, persistence of specialist insect herbivore populations may depend more upon plant density within Platte thistle populations, possibly reflecting patch quality (Thomas et al., 2001), than upon the frequency of populations across the landscape. Within-population plant densities at the edges of Platte thistle's range do not differ from those within the interior of the distribution (McMinn et al., 2017). Second, as a perennial plant species in which individuals vary in the number of years required to reach reproduction (F. L. Russell et al., unpublished data), Platte thistle populations may be a reliable resource for specialist insect herbivores, sustaining local insect populations. Years of poor seedling recruitment do not necessarily produce a future failure of resources for flower head-feeding insects because thistles from other cohorts with precocious or delayed reproduction extend flower head availability.

Although flower head feeding by insects reduced seed production in all three experimental years across all five sites, the magnitude of the insect herbivore effect varied substantially among years. Our data suggest that this variation in insect effects reflected inter-annual variation in insect pressure and in seed production by insecticide-protected plants.

First, temporal variation in ambient levels of insect damage was evident. Control plants were most successful in producing seed in 2017, when flower head damage

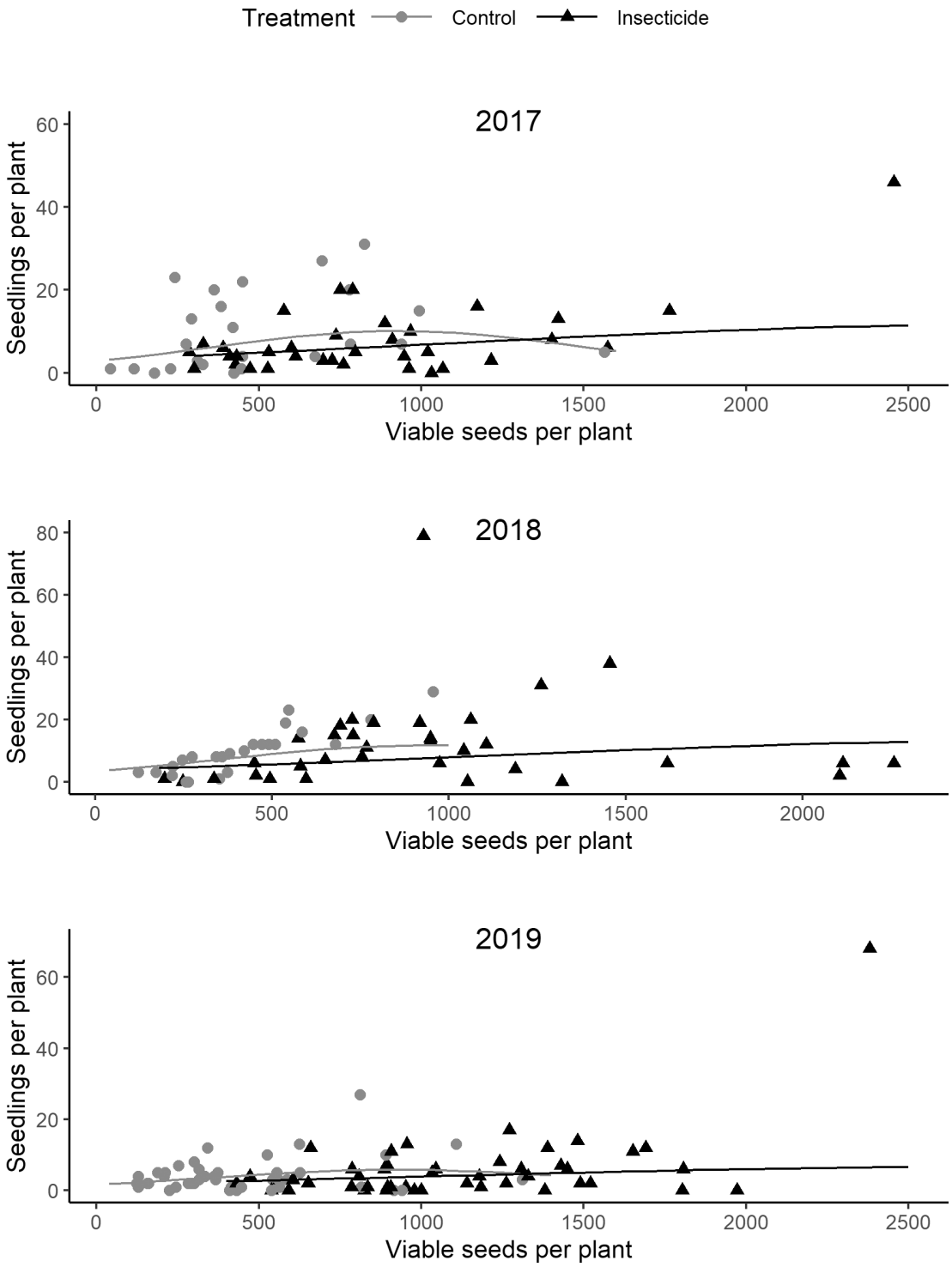


FIGURE 4 Relationship between number of viable seeds produced per plant and number of seedlings established by treatment level (insect herbivore reduction and control with ambient herbivory). Data are shown separately for Platte thistles that were adults in each experimental year: 2017, 2018, and 2019. Curves represent model-predicted number of seedlings per plant from the mixed model ANCOVA for control plants and for insecticide-treated plants. "Year" and "site" are random effects in these models. Year-specific curves are shown for Site E with the median number of seedlings germinated per adult plant. In 2017, the relationship for seedlings per plant for control plants was $1.0521 + 0.0027$ (viable seeds per plant) $- 1.456\ e^{-6}$ (viable seeds per plant)^2; and for insecticide-treated plants: $1.1700 + 0.0009$ (viable seeds per plant) $- 1.59\ e^{-7}$ (viable seeds per plant)^2. The curves for 2018 and 2019 only differed from 2017 curves in their intercepts. For 2018, the intercept was 1.1968 for control plants and 1.3148 for insecticide-treated plants. For 2019, the intercept was 0.5024 for control plants and 0.6203 for insecticide-treated plants.

was least, and least successful in 2019, when damage was greatest. Since ambient levels of feeding on control plants reflect variation in insect numbers and activity, the results suggest that herbivore populations and pressure varied between years.

Second, we found temporal variation in successful seed production even among insecticide-treated plants. This likely reflects variation both in insecticide efficacy, with greatest efficacy in 2019, and in moisture availability. We hypothesize that protected plants can mature more seed in wetter years. January–June precipitation was 52.6% greater in 2019, the year with the greatest seed production by insecticide-treated plants, than in 2017 and 252.8% greater than in 2018. Thus, our data suggest that the intense insect herbivore pressure on Platte thistle floral resources in these montane populations is strongly influenced by multiple factors that vary among years, especially herbivore pressure and moisture availability.

Greater seed availability did not increase seedling recruitment

Although flower head-feeding insects reduced Platte thistle seed production, experimentally lessening insect damage to seeds did not increase seedling recruitment. Seedling recruitment was not directly proportional to viable seed released, and it did not differ between treatments. Some insecticide-treated plants produced very large numbers of viable seeds (>1600 seeds per plant); yet, these additional seeds resulted in only minimal increases in seedling recruitment. These results suggest that post-dispersal ecological processes limited seedling recruitment below the seed availability threshold, especially at higher levels of seed input.

The relationship between seed input and seedling establishment in dry montane grasslands likely is governed by different processes than in mesic grasslands. In mesic systems, exposed bare ground often strengthens the connection between seed production and seedling establishment (Maron & Crone, 2006; Turnbull et al., 2000) by alleviating light limitation (Grman, 2013) and deleterious effects of plant litter (Jessen et al., 2023). However, in our dry grasslands, extensive bare ground is common, yet seedling establishment was low, suggesting physically open sites are not limiting. In this system, water limitation and/or ant granivory in large patches of bare ground may explain the lack of proportional seed to seedling recruitment; further tests are warranted.

Interspecific interactions are expected to shift from competitive to facilitative as abiotic stress increases (Bertness & Callaway, 1994). This shift appears to be strongest when stress arises from scarce belowground resources (Adams et al., 2021). Dry montane grasslands likely are abiotically stressful for Platte thistle, with soil water availability limiting recruitment. During our experiment, the highest seedling establishment coincided with the greatest January–June precipitation. Platte thistles that produced seed in 2018 had the highest seedling establishment and their first-year seedlings, which dominate Platte thistle seedling recruitment (F. L. Russell et al., unpublished data), germinated in the wettest winter–spring of our five-year study. January–June precipitation in 2019 was >3.5× greater than that in either 2018 or 2020, the first year for seedling establishment from the 2017 and 2019 experiments, respectively. Under dry conditions, shading and litter accumulation around neighbor plants may increase soil water availability (Fick et al., 2016; Loydi et al., 2013), more than it increases competition; this hypothesis merits examination in this system.

Second, granivory can significantly reduce plant recruitment in dry systems (Brown et al., 1979). *Pogonomyrmex occidentalis*, a granivorous ant species common to our study sites (F. L. Russell et al., unpublished data), efficiently removes seeds from exposed soil (Fick et al., 2016). Seeds that are embedded in litter adjacent to neighboring plants may escape ant harvesting. We hypothesize that ant granivory is important in limiting Platte thistle seedling establishment on bare ground in this montane environment. This hypothesis is supported by comparing our measure of the difficulty of seedling establishment versus the availability of bare ground across our study sites. A positive correlation between the difficulty of seedling establishment (ratio of viable seeds produced to seedlings established) and the amount of bare ground was observed among four sites; only one site, Site C, interrupted this trend. Site C had both extensive bare ground and high seedling establishment. Notably, site C had the fewest *P. occidentalis* mounds (F. L. Russell et al., unpublished data). Therefore, reducing ants may reduce the negative effects of bare ground.

Elevation did not mediate the effects of insect herbivory on plant performance

Ecological context in the form of inter-annual variation in resources, likely precipitation before the growing season, affected some aspects of the Platte thistle–insect interaction. However, predictable spatial variation in the interaction across the elevational gradient was weak. Specifically, elevation did not influence insect herbivore: (1) damage to Platte thistle flower heads, (2) effects upon viable seed production, and (3) effects on seedling recruitment. This surprised us since elevation is expected to

represent a systematic change in key environmental variables, including temperature, UV radiation, and concentrations of oxygen and carbon dioxide (Körner, 2007).

Why was elevation insufficient to explain the patterns of herbivore–plant interaction in this study? One hypothesis is that our study sites did not encompass an adequate range of elevations. This explanation seems unlikely. Our study sites spanned most of the elevation range of Platte thistle in the upper Arkansas River valley. Further, reproductive phenology in our Platte thistle populations was correlated with site elevation. More likely, we hypothesize that any influence of elevation was reduced by (1) internal feeding by Platte thistle flower head insects, (2) complexity of factors influencing montane weather and resource availability, and (3) habitat constraints for Platte thistle in montane grasslands at the periphery of its geographic range.

First, while external-feeding insects are fully exposed to colder temperatures at higher elevations, endophagous insects may be insulated by the plant tissues that surround them, extending their range. The fact that the flower head-feeding specialist insects on Platte thistle are internal feeders is a novel aspect of our research because most tests of the relationship between elevation and insect herbivory have involved external-feeding folivores (Kozlov et al., 2022). Our results are consistent with Kozlov et al. (2022) who found no relationship between damage by internal-feeding folivores (gallers and miners) across six Eurasian mountain ranges, even though damage by external-feeding insects decreased with elevation.

Second, the influence of elevation on plant performance and species interactions may be obscured by variation in other environmental factors, including orographic rain shadow effects, slope aspect, and edaphic conditions. These complexities are common in the Colorado Rocky Mountains and result in the importance of local topographic variation driving weather on the small scale (Nelson, 2007). A rain shadow effect likely overrides some of the direct effects associated with elevational variation in the Upper Arkansas River valley, which sits just east of the Rocky Mountain continental divide. Weather systems primarily travel west to east, passing over the >4000 m peaks of the Collegiate Peaks mountain range (Nelson, 2007). Consistent with a rain shadow effect, our three study sites on the western (lee) side of the valley, which were intermediate in elevation, had lower soil moisture availability than our two sites on the eastern side of the valley, the lowest and highest sites. By affecting moisture availability, rain shadows likely influence Platte thistle seedling establishment and survival in this region.

Finally, Platte thistle is associated with a specific habitat, montane grasslands on gravelly sand soils, across its full elevational range in our region. This habitat specificity likely diminishes the effects of elevation on its interactions with insect herbivores. Montane grasslands tend to occur under similar abiotic conditions across elevations. In contrast to our results, Miller et al. (2009) found that elevation modified insect herbivore effects on *Opuntia imbricata* populations over a smaller elevation range (e.g., 120 m) than separated our study sites. However, *Opuntia imbricata* occurred in distinct plant communities, from lower desert grassland to upper rocky slopes, along the elevation gradient.

CONCLUSION

Our study demonstrates that, even in populations at the periphery of a species' biogeographic range, feeding by specialized insects in developing flower heads can substantially reduce key components of plant maternal fitness. However, these effects are not the main factor limiting Platte thistle population growth in this region. Dry conditions at the species range edge and ant granivory likely restricted availability of "safe sites" for seedling establishment, disconnecting seedling recruitment from viable seed production above minimal levels. In general, if peripheral plant populations experience abiotically stressful conditions, as predicted by the Abundant Center Hypothesis (Brown, 1984), then seedlings, with their small root systems, are the life stage that is likely to be most affected. As a result, for many plant species, seed production often may exceed "safe site" availability in their peripheral populations.

While insect herbivore effects on individual plants and on plant populations often vary among ecological contexts (Maron et al., 2014), elevation did not have a strong or predictable influence on the insect herbivore–Platte thistle interaction. Viewed through the conceptual framework proposed by Maron et al. (2014), our system showed that spatial and, especially, temporal context influenced the magnitude of herbivore damage. However, this variation in context did not fundamentally change the herbivores' strong effect on individual plant performance or their weak influence on population recruitment and, so, local density.

AUTHOR CONTRIBUTIONS

F. Leland Russell and Svata M. Louda designed the study. Mason R. Taylor and F. Leland Russell carried out the fieldwork. All three authors participated in the data management, statistical analysis, and writing of the manuscript.

ACKNOWLEDGMENTS

We thank the Bureau of Land Management, especially Cathy Hagen; the United States Forest Service, especially Jim Pitts; the Colorado State Land Board; and a private landowner, Annie Vohs, for encouragement, assistance, and access to study sites. We are grateful to Rodney W. Otley and Mary Liz Jameson for logistical support and for many fruitful discussions that benefited our project. Financial support was generously provided by Wichita State University, the Kansas Academy of Science Student Research Grants Program, the Colorado Native Plant Society John W. Marr Grant Program, and it was supplemented by personal funds.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

Data (Russell et al., 2025) are available on Dryad: https://doi.org/10.5061/dryad.kh18932dc.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Russell, F. Leland, Mason R. Taylor, and Svata M. Louda. 2025. "Microsite Availability, Not Floral Herbivory, Limits Recruitment in Peripheral Native Thistle Populations." *Ecosphere* 16(6): e70310. https://doi.org/10.1002/ecs2.70310